

## REVIEW

# Full-intensity and reduced-intensity allogeneic stem cell transplantation in AML

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**Allogeneic stem cell transplantation represents the most active form of anti-leukaemic therapy in acute myeloid leukaemia (AML). Advances in transplant technology and supportive care have resulted in improved outcomes in patients allografted using a myeloablative conditioning regimen. At the same time the use of reduced-intensity conditioning regimens has allowed an immunologically mediated graft-versus-leukaemia effect to be exploited in older patients who were previously ineligible for transplantation on the grounds of age or comorbidity. This coupled with the increased availability of alternative stem cell sources, in the form of either unrelated or cord blood donations, has established allogeneic transplantation as a key therapeutic strategy in the treatment of both younger and older adults with AML.**

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## Introduction

Acute myeloid leukaemia (AML) is now the commonest indication for allogeneic transplantation in adults.<sup>1</sup> This reflects both the continued inadequacy of conventional chemotherapy and the growing realization that a potent graft-versus-leukaemia (GVL) may be exploited in patients allografted for AML. In the past decade it has become clear that not only has the benefit of myeloablative allogeneic transplantation in younger patients been consistently underestimated but also that the use of reduced-intensity conditioning (RIC) regimens now permits the delivery of a potentially curative GVL effect in older patients whose outcome with conventional chemotherapy would be dismal.<sup>2,3</sup>

The limitations of conventional chemotherapy are particularly marked in patients over the age of 50 where fewer than 20% of patients can be expected to be cured with current regimens.<sup>2–5</sup> Until the advent of RIC regimens the potent anti-leukaemic activity of allogeneic transplantation had been restricted to the small number of adults young enough to tolerate the toxicity of a myeloablative conditioning regimen.<sup>6</sup> In a disease whose incidence rises sharply with age this effectively prevented the delivery of the most effective form of anti-leukaemic therapy to the majority of patients with AML.<sup>5</sup> Consequently, the demonstration that reduced-intensity regimens permit the extension of a GVL effect to the majority of patients with AML represents arguably the most important therapeutic advance since the introduction of conventional chemotherapy.<sup>7–10</sup> At the same time the ability to access suitable stem cell donors for patients who lack a matched sibling—whether through the increased size of volunteer unrelated registries or the development of umbilical cord blood banks—now makes the long-term aim of incorporating an allograft as a key component of the management plan for all patients with high-risk AML a realistic prospect for the coming decade.<sup>11–13</sup>

## Emerging therapeutic strategies in AML

### *Improving outcome of conventional chemotherapy*

The remarkable clinical activity of the ABL kinase inhibitor imatinib in the treatment of patients with chronic myeloid leukaemia (CML) has demonstrated the therapeutic potential of targeting the molecular abnormalities causing myeloid leukaemias.<sup>14</sup> However, the molecular complexity of AML has limited the development of a similar therapeutic strategy.<sup>15,16</sup> Thus, although there has been much interest in the development of small molecule inhibitors which target dysregulated signalling pathways, such as FLT3 inhibitors or farnesyltransferase inhibitors, these drugs are largely ineffective in both newly diagnosed and relapsed patients with AML.<sup>17,18</sup> Whilst disappointing, these results are not in themselves surprising given the remarkable degree of molecular heterogeneity of adult AML as demonstrated by microarray and single nucleotide polymorphism analyses.<sup>16,19</sup> The notion that targeting a single pathway is likely to be an effective treatment strategy

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in AML is also undermined by the demonstration that the pathogenesis of this disease is a multi-step process in which the acquisition of mutations in both differentiation and proliferative pathways is required.<sup>15,20</sup> It has therefore become clear that the prospect of currently available targeted therapies transforming the outcome for large numbers of adults with AML is as remote as it is economically daunting.

The identification of a small subpopulation of CD34+ CD38- cells within the bulk population of leukaemic blasts, which are responsible for the continued growth and propagation of leukaemic cells in murine transplantation models has led to a re-evaluation of the target cell population in AML.<sup>21</sup> Importantly, a number of groups have demonstrated that this population of leukaemic stem cells (LSC) demonstrate differences in their transcriptional profile compared with the bulk population of leukaemic blasts.<sup>22</sup> Consequently, strategies which target molecular pathways central to the survival of long-term repopulating LSC are required. Such an approach, which offers the prospect of specifically targeting the LSC compartment, is of genuine therapeutic promise but is currently limited by our incomplete understanding of the molecular determinants of the LSC phenotype. Nonetheless, inhibitors of the nuclear factor (NF)κB pathway, mTOR, and the WNT/β-catenin pathways, all of which have been shown to be abnormally regulated in LSC, are currently in Phase I/II trials<sup>23–26</sup> and offer the possibility of developing a more effective targeted therapeutic strategy.

#### *Optimizing a GVL effect in AML*

The growing realization of the limitations of targeting intracellular abnormalities within AML blasts has led to renewed interest in the development of immunotherapeutic strategies which instead target immunodominant epitopes on the surface of leukaemic blasts. Harnessing a GVL effect represents an increasingly important treatment option in AML thanks to the development of RIC regimens. In addition to classical data showing a correlation between disease relapse and the presence of GVHD,<sup>27,28</sup> some of the most compelling evidence supporting the presence of a significant GVL effect in patients allografted for AML is the demonstration that the intensity of post transplant immunosuppression has a major impact on relapse risk.<sup>29</sup> Remarkably reducing the cyclosporin A (CsA) dose administered in the first 20 days post transplant is sufficient to reduce the relapse rate by more than 30%.<sup>30,31</sup> However, exploitation of a GVL effect—an approach which can bypass the intracellular mechanisms of drug resistance which characterize adult AML—has been hampered by the risk of attendant severe GVHD.

Candidate antigens on AML blasts and LSC recognized in a GVL reaction include minor histocompatibility antigens and leukaemia-specific antigens such as WT1 and PR-1.<sup>32,33</sup> Immunotherapeutic strategies that aim to exert an anti-tumour effect through the induction of a donor response to a disparate host HA-1 antigen are limited by the rare frequency of this allele and its HLA restriction.<sup>34,35</sup> In contrast, antigens such as WT1 represent an attractive option given its overexpression in up to 70%

of patients with AML.<sup>32</sup> Recently, the demonstration of immune responses to leukaemia-specific antigens such as WT1 and PR-1 in patients with AML and ALL has led to studies that aim to induce an autologous T-cell response using peptide vaccination or TCR gene transfer.<sup>36,37</sup> It is clear however that further characterization of the antigens recognized in the GVL reaction will be vital if we are to improve current immunotherapeutic strategies in high-risk AML.

#### **Results of myeloablative allogeneic stem cell transplantation in AML**

A myeloablative sibling allograft delivers the most effective form of anti-leukaemic therapy in patients with AML. In patients transplanted in first CR long-term disease-free survival (DFS) rates in the region of 60–70% are routinely achieved and the risk of disease relapse is reduced by 25–30% compared with patients treated with chemotherapy alone.<sup>6,38</sup> In patients with advanced disease (CR2 or greater), whose outcome with salvage chemotherapy alone is usually dismal, DFS rates in the region of 40–50% can be expected after a sibling or unrelated donor transplant.<sup>39</sup> Remarkably, even patients with primary refractory disease can achieve durable remissions after a myeloablative allograft.<sup>40</sup> The major causes of treatment failure in patients allografted using a myeloablative conditioning regimen remain transplant-related mortality (TRM), principally GVHD, and, in patients with advanced-phase disease, relapse. Consequently, the effective treatment of patients in first CR has been dependent on the development of a risk stratification model which allows allogeneic transplantation, with its attendant morbidity and mortality, to be restricted to patients whose outcome with chemotherapy alone will be poor. In contrast, in patients with advanced disease the demand has been for transplant technologies that further reduce the risk of disease relapse either through intensifying the conditioning regimen or optimizing a GVL effect. Importantly, although there can be increasing confidence that RIC regimens possess the capacity to produce durable remissions their ability to deliver long-term DFS has not yet been conclusively demonstrated. Consequently, a myeloablative conditioning regimen should still be viewed as the 'gold standard' preparative regimen in eligible patients undergoing allogeneic transplantation in AML until longer-term follow-up of outcome of RIC allografts and results of a formal comparison of both regimens is obtained.

#### **The role of allogeneic transplantation in management of patients in first CR**

After 20 years of heated debate there is now convincing evidence that sibling allogeneic transplantation improves DFS and overall survival (OS) in patients with AML in first CR.<sup>6,41–43</sup> The controversy surrounding this issue has in part been caused by the long-standing mutual antagonism of transplant enthusiasts and their more sceptical

colleagues and also reflects the difficulties of assessing the benefits of transplantation in a statistically rigorous fashion. These include the fact that physician and patient bias make it impossible to perform a randomized controlled trial and the inevitability that the time delays associated with allogeneic transplantation allow some of the highest risk patients to relapse before a transplant can be performed. An alternative method to assess the benefit of transplantation in first CR is the use of a 'donor-vs-no donor' analysis in which the availability of a matched sibling donor—a random event—is used as a substitute for formal randomization. Although such an approach is imperfect, since it will underestimate the benefit (or detriment) of transplantation because not all patients with an available donor will actually be transplanted, this form of analysis has proved the best way of overcoming the multiple potential biases associated with the decision whether to allograft a patient.<sup>44</sup> Five major studies using a donor-vs-no donor analysis have now been reported in patients with AML in first CR.<sup>6,41–43,45</sup> While they do not individually show any benefit in terms of overall survival (OS) all but one shows an improvement in DFS in patients with an available sibling donor (Table 1). The absence of a statistically significant survival benefit in the individual studies probably reflects the fact that a sizeable proportion of patients in the non-transplant arm who relapsed received salvage chemotherapy and a subsequent allogeneic transplant. However, a recent meta-analysis of more than 4000 patients entered into these trials confirms improved survival, as well as DFS, in patients with an available HLA identical sibling donor.<sup>6</sup>

Given the toxicity of allogeneic transplantation and the ability of current chemotherapy regimens to cure a proportion of patients it is essential to identify patients who are likely to be cured by chemotherapy alone. To date the most effective form of risk stratification has been based on presentation karyotype. Thus, while allografting improves outcome in patients with intermediate or high-risk cytogenetics it is not of benefit in patients with good-risk cytogenetics.<sup>6</sup> The recent demonstration that relapse risk can be more accurately quantified using molecular analysis—including mutational analysis of

FLT3 and NPM1 genes—offers the prospect of further accuracy in defining which patients with intermediate-risk cytogenetics will benefit from allogeneic transplantation using a sibling, or possibly unrelated, donor.<sup>46</sup> The other critical assessment to be made in patients who are candidates for allogeneic transplantation relates to the risk of transplantation. Gratwohl's seminal study<sup>47</sup> of patients transplanted for CML identified patient age, disease duration, donor sex and HLA disparity as predictors of TRM. In patients undergoing a sibling allograft for AML patient age has emerged as the most significant determinant of TRM.<sup>6,41</sup> The demonstration that the TRM after a myeloablative allograft in patients with AML in first CR rises from 19% in patients under the age of 20 to 39% in patients over the age of 35 supports the exploration of the use of RIC allografts in this population. Recent improvements in tissue typing and transplant technology have also resulted in markedly improved outcomes after unrelated donor transplantation in AML.<sup>48</sup> Consequently, in patients with high-risk cytogenetics who lack a matched sibling the poor outcome with standard chemotherapy justifies the emergence of transplantation using a volunteer unrelated donor (VUD) as standard practice.<sup>49,50</sup>

#### Role of allogeneic transplantation in the management of patients in second CR and with advanced-phase disease

The fact that no prospective studies have examined the role of allogeneic transplantation in patients with AML beyond first CR is testament to the inability of chemotherapy alone to produce durable remissions in the great majority of patients with relapsed disease and the consequent absence of an ethically acceptable control arm. In contrast, registry data demonstrate DFS rates in the region of 30–50% in patients in second CR and therefore it seems reasonable to recommend that all patients with a sibling donor should proceed swiftly to transplant once they have achieved remission.<sup>39</sup> In patients lacking a matched family donor DFS rates in the region of 35–40% can be achieved using a well-matched VUD and in young patients with a short CR1 duration and intermediate- or poor-risk cytogenetics this is the treatment of choice.<sup>48–50</sup> However, it should be remembered that autologous transplantation has been reported to produce similar long-term survival rates in patients with first CR of long-duration (>18 months) and good-risk cytogenetics.<sup>39,51,52</sup> Autologous transplantation is recommended for patients with relapsed acute promyelocytic leukaemia (APML) who have achieved a molecular remission and have a molecularly negative stem cell product<sup>53,54</sup> and should also be considered in all patients with good-risk cytogenetics lacking a well-matched unrelated donor. In contrast allogeneic, rather than autologous, transplantation should be performed, where possible, in patients with relapsed APML who fail to achieve a molecular remission with salvage chemotherapy.<sup>55</sup> The encouraging recent results of RIC allografts in patients in second CR using sibling or unrelated donors make this an important area for future study particularly in patients estimated to have a high TRM using a myeloablative regimen. Patients with primary refractory AML

**Table 1** Donor-vs-no donor analysis of impact of sibling allogeneic transplantation on OS and DFS in patients with AML in first CR

Study	OS (%)			DFS (%)		
	Donor	No donor	P	Donor	No donor	P
GOELAM	53	53	NS	44	38	0.6
MRC	56	50	NS	50	42	0.001
EORTC	48	40	NS	46	33	0.01
BGMT	65	51	NS	66	42	<0.05
HOVON	54	46	NS	48	37	<0.001

Abbreviations: BGMT = Bordeaux-Grenoble-Marseille-Toulouse; DFS = disease-free survival; EORTC = European Organisation for Research and Treatment of Cancer; GOELAM = Groupe Ouest Est Leucémies Aigues Myéloblastiques; HOVON = Hematology and Oncology Society for Adults in the Netherlands; MRC, Medical Research Council; NS = not significant; OS = overall survival.

who fail to achieve CR after induction chemotherapy should also be considered for a myeloablative transplant now that a number of centres have confirmed long-term DFS rates of between 20 and 30% in patients fit enough to tolerate a myeloablative conditioning regimen.<sup>40,56,57</sup>

### Optimizing outcome after myeloablative allogeneic transplantation in AML

Recent efforts to improve the outcome of myeloablative allografts in AML have focused on reducing TRM and relapse risk. The two most widely used myeloablative conditioning regimens in AML combine either cyclophosphamide and total body irradiation (Cy/TBI) or busulphan (Bu) and Cy (Bu/Cy). A number of randomized studies have compared these regimens in myeloid malignancies.<sup>58–63</sup> While there would appear to be broad equivalence of both regimens in patients with CML and early phase AML, studies using oral preparations of Bu suggest superiority of TBI-based regimens in patients with advanced-phase AML.<sup>64</sup> However, the pharmacokinetics of oral Bu are unpredictable resulting in highly variable blood levels. Given the higher relapse risk observed in patients with low Bu levels and the impact of high Bu levels on transplant toxicity—principally veno-occlusive disease—there has been intense interest in improving delivery of this highly effective anti-leukaemic drug.<sup>65,66</sup> Consequently, historical results obtained using oral Bu must be re-evaluated in the light of the significantly improved results achievable using newer methods of Bu delivery. These include the use of area under the curve measurements to determine dosing of oral Bu, which allows a defined blood Bu level to be reliably delivered (targeted <sup>T</sup>Bu/Cy) or the use of an intravenous Bu preparation (i.v. Bu/Cy).<sup>67–69</sup> Using a <sup>T</sup>Bu/Cy regimen the Seattle group have reported markedly reduced transplant toxicity and relapse rates in patients allografted for CML and secondary AML.<sup>70</sup> Similar encouraging results have been reported using an i.v. Bu/Cy regimen and there is now a compelling case for a randomized trial of one of these regimens with a Cy/TBI preparative regimen.

There has been much interest in whether the outcome after sibling allogeneic transplantation in AML can be improved by the use of peripheral blood stem cells (PBSC) as opposed to bone marrow progenitors. While unquestionably associated with faster neutrophil and platelet engraftment the impact of the use of PBSC on long-term outcome remains controversial. There are, however, reports which demonstrate improved survival, consequent upon a reduction in both TRM in patients with advanced-phase AML.<sup>71</sup> This benefit may be offset by an increased risk of chronic GVHD in PBSC recipients and in children there is evidence that this translates into decreased OS.<sup>72</sup> It has also been hypothesized that the use of PBSC provides a mechanism by which a GVL effect may be augmented as a result of manipulation of the content of the stem cell inoculum. Such a notion is supported by separate evidence that the CD34+ content of grafts may be a critical determinant of relapse risk, possibly through an effect on numbers of antigen-presenting cells.<sup>73,74</sup>

### Reduced-intensity allogeneic transplants in AML

The ability of reduced-intensity allografts to deliver durable remissions in older patients with high-risk AML has now been demonstrated by a number of groups.<sup>7,9,10,75–81</sup> Although longer-term follow-up is still required on these cohorts it is likely that RIC allografts represent a major advance in the treatment of AML in older patients and important questions concerning the optimal reduced-intensity regimen and the integration of RIC allografts into the overall management of older patients must now be addressed.

Leukaemia-free survival rates in the region of 40–60% of patients are achievable using a variety of reduced-intensity regimens in patients with AML in their fifth and sixth decade. A number of conclusions can be drawn from these studies. First, the outcome of patients with active disease at the time of transplant is poor and these patients should not be considered for RIC transplant unless a morphological remission can be achieved with salvage chemotherapy. Second, the outcome of patients undergoing a sibling or unrelated donor transplant is broadly equivalent—a significant observation given the scarcity of fit sibling donors in this older population. Lastly, the incidence of acute and chronic GVHD after T-cell-replete RIC allografts in AML is considerable with up to 60% of patients developing chronic GVHD.<sup>9,79,81</sup> In addition to representing the major cause of morbidity and mortality the presence of active GVHD limits opportunities to manipulate a GVL effect post transplant using donor lymphocyte infusion (DLI) or withdrawal of immunosuppression. Use of the T-cell-depleting antibodies anti-thymocyte globulin (ATG) or alemtuzemab as part of the preparative regimen has been shown to be effective at reducing the risk of severe acute GVHD and extensive chronic GVHD.<sup>10,12,82,83</sup> However, the attendant T-cell depletion is associated with impaired immune reconstitution and an increased risk of infectious complications—particularly cytomegalovirus reactivation.<sup>84,85</sup> More importantly, such regimens are associated with a higher risk of disease relapse and this represents the major cause of treatment failure in patients undergoing a reduced-intensity transplant for myeloid malignancies.<sup>10</sup> Therefore, the development of strategies which decrease relapse risk in the context of T-cell-depleted reduced-intensity allograft is now a priority.

### What is the current role of RIC allografts in the management of AML?

The challenges of confirming the benefit of myeloablative allografts in AML have already been referred to and the extent to which the encouraging preliminary results with RIC allografts in AML reflect patient selection should be borne in mind. Two prospective studies have recently confirmed that it is possible to transplant a substantial proportion of older patients with AML and one study has demonstrated improved outcome in patients allografted using a donor-vs-no donor analysis.<sup>76,78</sup> However, larger studies are still required of the impact of RIC allografts,

using both sibling and unrelated donors, on the outcome of patients over the age of 50 with newly diagnosed AML.

### Strategies to improve outcome after RIC allografts

#### *Reduction of disease relapse*

Disease relapse occurs in 30–40% of patients transplanted in first or second CR. The kinetics of relapse after RIC allografts appears to differ from that observed after myeloablative transplants occurring, in the great majority of patients, in the first year post transplant.<sup>10</sup> Consequently, strategies aimed at decreasing the relapse rate must either focus on reducing the disease burden prior to transplant, intensifying the preparative regimen without a concomitant increase in toxicity or optimizing the GVL effect in the first few months post transplant.

The observation that almost all patients undergoing an RIC allograft with active disease at the time of transplant relapse raises the possibility that a more precise assessment of disease load in patients in morphological CR at the time of transplant might identify patients at a higher risk of relapse and this needs to be formally addressed, using immunophenotypic measures of minimal residual disease (MRD), in future prospective studies. Two approaches currently under investigation by which the leukaemic burden at the time of transplant can be minimized are the use of an additional cycle of chemotherapy (usually a third course) or even an autograft prior to allogeneic transplantation<sup>7</sup> or modification of the preparative regimen to increase its anti-leukaemic activity. *i.v.* Bu is currently being explored in conjunction with fludarabine as a novel preparative regimen.<sup>69,86</sup> Alternatively, radioimmunotherapy may prove an effective mechanism of targeting the anti-leukaemic properties of radionuclides such as yttrium or iodine to the marrow cavity without increasing extramedullary toxicity. In animal models radiolabelled anti-CD45 has also been shown to be immunosuppressive raising the possibility that such an approach might be sufficient to deliver durable engraftment as well.<sup>87</sup>

A number of approaches can be used to augment a GVL effect post transplant. CsA exposure is likely to play a central role in determining relapse risk after a RIC transplant.<sup>29,30</sup> While the already substantial risk of severe acute and chronic GVHD in recipients of T-cell-replete RIC allografts makes it difficult to contemplate a reduction in the intensity of post transplant immunosuppression with any degree of equanimity, particularly in recipients of unrelated donor transplants, a randomized study examining the impact of reducing the dose or duration of CsA on disease relapse is important in recipients of T-cell-depleted RIC allografts. Similarly, data are lacking concerning the optimal form of T-cell depletion in RIC allografts for AML. A prospective study comparing the impact of alemtuzemab or ATG dose on relapse and GVHD risk and the results of dose de-escalation in recipients of alemtuzemab-based RIC allografts would be of value in developing more effective reduced-intensity regimens.

Despite the clear evidence of GVL in AML, DLI is not usually effective in patients who relapse after a RIC

allograft. Consequently, there is interest in the administration of prophylactic DLI in patients deemed to be at a higher risk of relapse as a strategy to optimize the GVL effect post transplant.<sup>88</sup> The presence of mixed T-cell chimerism after a reduced-intensity allograft is taken to indicate bidirectional tolerance and it can be argued that DLI should be administered in these patients in the first few months post transplant with the aim of achieving full donor T-cell chimerism.<sup>89</sup> Other groups have explored the possibility of restricting the use of DLI to patients who demonstrate MRD positivity post transplant. This approach is hampered by the lack of informative molecular markers of MRD in the majority of patients with AML—in contrast to acute lymphoblastic leukaemia—and the concern that patients may proceed rapidly to a frank morphological relapse. Nonetheless, this approach may be of value in a number of patients particularly if markers such as WT1 prove to be accurate markers of incipient relapse.<sup>90</sup>

The major factor limiting the administration of DLI in the first year post transplant is the risk of GVHD—particularly in unrelated donor transplant recipients. It is still not clear why the toxicity of early DLI is so high but possibilities include the presence of residual host antigen-presenting cells and more rapid kinetics of lymphocyte expansion in a lymphopenic environment.<sup>91</sup> Whatever the mechanism it is necessary to administer much lower doses of lymphocytes than would be used in patients who relapse late and as a result it may be difficult to administer an effective lymphocyte dose.<sup>92</sup> Consequently, there is interest in the development of strategies which allow the postponement of DLI after RIC allografts. One such approach might be the adjunctive use of a drug such as azacitidine with activity against residual host leukaemic blasts to manipulate the kinetics of disease relapse and postpone or eliminate the requirement for DLI.<sup>93</sup>

### The future of RIC allogeneic SCT in AML

The advent of reduced-intensity preparative regimens has transformed the landscape of allogeneic transplantation for AML in less than a decade. Therefore, there is now a case for a prospective comparison of outcome after RIC and myeloablative conditioning regimens in younger patients with AML—particularly those aged between 35 and 50 whose TRM we know to be unacceptably high. Moreover, the encouraging outcomes achieved with RIC allografts in older patients with AML make it possible to contemplate a future where an allograft becomes standard of care for patients with AML over the age of 50. Because of the lack of effective chemotherapy for so many patients with high-risk AML it is possible that this fundamental change in practice will occur before the results of rigorous prospective studies are available—if indeed it ever proves practicable to perform them. However, three major challenges remain (Table 2). First, it is clear that the optimal reduced-intensity regimen has yet to be defined and prospective randomized studies will be required to address this issue. Second, although the expansion of unrelated donor registries and development of high resolution tissue typing has

**Table 2** Unresolved issues in reduced-intensity allogeneic stem cell transplantation amenable to analysis by randomized trials

- Role of T-cell depletion on relapse and overall survival
- Impact of minimal residual disease status at the time of transplantation on relapse risk
- Optimal dose and duration of post transplant immunosuppression
- Role of donor lymphocyte infusion in patients with isolated mixed T-cell chimerism in the first 12 months post transplant

transformed the availability and outcome of unrelated donor transplants a significant proportion of patients still lack a suitable donor. However, encouraging results have been reported using umbilical cord blood, particularly when two cord blood units are used, and this may represent an important new stem cell source for RIC allografts in AML.<sup>94</sup> Lastly, the failure of up to 30% of older patients with high-risk AML to achieve a CR of sufficient duration to allow them to proceed to an allograft with induction chemotherapy means that regimens which improve outcome in patients with resistant or relapsed disease are required. The FLAMSA regimen which incorporates an additional block of cytoreductive therapy prior to transplant followed by early DLI appears promising in this regard.<sup>95,96</sup>

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### References

- 1 Gratwohl A, Baldomero H, Frauendorfer K, Urbano-Ispizua A. EBMT activity survey 2004 and changes in disease indication over the past 15 years. *Bone Marrow Transplantation* 2006; **37**: 1069–1085.
- 2 Buchner T, Berdel WE, Wormann B, Schoch C, Haferlach T, Schnittger S *et al.* Treatment of older patients with AML. *Crit Rev Oncol Hematol* 2005; **56**: 247–259.
- 3 Leith CP, Kopecky KJ, Godwin J, McConnell T, Slovak ML, Chen IM *et al.* Acute myeloid leukemia in the elderly: assessment of multidrug resistance (MDR1) and cytogenetics distinguishes biologic subgroups with remarkably distinct responses to standard chemotherapy. A Southwest Oncology Group study. *Blood* 1997; **89**: 3323–3329.
- 4 Grimwade D, Walker H, Harrison G, Oliver F, Chatters S, Harrison CJ *et al.* The predictive value of hierarchical cytogenetic classification in older adults with acute myeloid leukemia (AML): analysis of 1065 patients entered into the United Kingdom Medical Research Council AML11 trial. *Blood* 2001; **98**: 1312–1320.
- 5 Frohling S, Schlenk RF, Kayser S, Morhardt M, Benner A, Dohner K *et al.* Cytogenetics and age are major determinants of outcome in intensively treated acute myeloid leukemia patients older than 60 years: results from AMLSG trial AML HD98-B. *Blood* 2006; **108**: 3280–3288.
- 6 Cornelissen JJ, van Putten WL, Verdonck LF, Theobald M, Jacky E, Daenen SM *et al.* Results of a HOVON/SAKK donor versus no-donor analysis of myeloablative HLA-identical sibling stem cell transplantation in first remission acute

myeloid leukemia in young and middle-aged adults: benefits for whom? *Blood* 2007; **109**: 3658–3666.

- 7 Blaise D, Bay JO, Faucher C, Michallet M, Boiron JM, Choufi B *et al.* Reduced-intensity preparative regimen and allogeneic stem cell transplantation for advanced solid tumors. *Blood* 2004; **103**: 435–441.
- 8 de Lima M, Anagnostopoulos A, Munsell M, Shahjahan M, Ueno N, Ippoliti C *et al.* Nonablative versus reduced-intensity conditioning regimens in the treatment of acute myeloid leukemia and high-risk myelodysplastic syndrome: dose is relevant for long-term disease control after allogeneic hematopoietic stem cell transplantation. *Blood* 2004; **104**: 865–872.
- 9 Hegenbart U, Niederwieser D, Sandmaier BM, Maris MB, Shizuru JA, Greinix H *et al.* Treatment for acute myelogenous leukemia by low-dose, total-body, irradiation-based conditioning and hematopoietic cell transplantation from related and unrelated donors. *J Clin Oncol* 2006; **24**: 444–453.
- 10 Tauro S, Craddock C, Peggs K, Begum G, Mahendra P, Cook G *et al.* Allogeneic stem-cell transplantation using a reduced-intensity conditioning regimen has the capacity to produce durable remissions and long-term disease-free survival in patients with high-risk acute myeloid leukemia and myelodysplasia. *J Clin Oncol* 2005; **23**: 9387–9393.
- 11 Lacerda JF, Martins C, Lourenco F, Carmo JA, Juncal C, Oliveira JJ *et al.* Unrelated stem cell transplantation after a reduced intensity conditioning regimen containing high-dose thymoglobulin leads to controllable graft-versus-host disease. *Biol Blood Marrow Transplant* 2007; **13**: 494–497.
- 12 Chakraverty R, Peggs K, Chopra R, Milligan DW, Kottaridis PD, Verfuere S *et al.* Limiting transplantation-related mortality following unrelated donor stem cell transplantation by using a nonmyeloablative conditioning regimen. *Blood* 2002; **99**: 1071–1078.
- 13 Bertz H, Potthoff K, Finke J. Allogeneic stem-cell transplantation from related and unrelated donors in older patients with myeloid leukemia. *J Clin Oncol* 2003; **21**: 1480–1484.
- 14 Druker BJ, Guilhot F, O'Brien SG, Gathmann I, Kantarjian H, Gattermann N *et al.* Five-year follow-up of patients receiving imatinib for chronic myeloid leukemia. *N Engl J Med* 2006; **355**: 2408–2417.
- 15 Frohling S, Scholl C, Gilliland DG, Levine RL. Genetics of myeloid malignancies: pathogenetic and clinical implications. *J Clin Oncol* 2005; **23**: 6285–6295.
- 16 Bullinger L, Valk PJ. Gene expression profiling in acute myeloid leukemia. *J Clin Oncol* 2005; **23**: 6296–6305.
- 17 Knapper S, Burnett AK, Littlewood T, Kell WJ, Agrawal S, Chopra R *et al.* A phase 2 trial of the FLT3 inhibitor lestaurtinib (CEP701) as first-line treatment for older patients with acute myeloid leukemia not considered fit for intensive chemotherapy. *Blood* 2006; **108**: 3262–3270.
- 18 Lancet JE, Gojo I, Gotlib J, Feldman EJ, Greer J, Liesveld JL *et al.* A phase 2 study of the farnesyltransferase inhibitor tipifarnib in poor-risk and elderly patients with previously untreated acute myelogenous leukemia. *Blood* 2007; **109**: 1387–1394.
- 19 Valk PJ, Verhaak RG, Beijnen MA, Erpelinck CA, Barjesteh van Waalwijk van Doorn-Khosrovani S, Boer JM *et al.* Prognostically useful gene-expression profiles in acute myeloid leukemia. *N Engl J Med* 2004; **350**: 1617–1628.
- 20 Tallman MS, Gilliland DG, Rowe JM. Drug therapy for acute myeloid leukemia. *Blood* 2005; **106**: 1154–1163.
- 21 Bonnet D, Dick JE. Human acute myeloid leukemia is organized as a hierarchy that originates from a primitive hematopoietic cell. *Nat Med* 1997; **3**: 730–737.
- 22 Krivtsov AV, Twomey D, Feng Z, Stubbs MC, Wang Y, Faber J *et al.* Transformation from committed progenitor to

- leukaemia stem cell initiated by MLL-AF9. *Nature* 2006; **442**: 818–822.
- 23 Kirstetter P, Anderson K, Porse BT, Jacobsen SE, Nerlov C. Activation of the canonical Wnt pathway leads to loss of hematopoietic stem cell repopulation and multilineage differentiation block. *Nat Immunol* 2006; **7**: 1048–1056.
- 24 Guzman ML, Neering SJ, Upchurch D, Grimes B, Howard DS, Rizzieri DA *et al*. Nuclear factor-kappaB is constitutively activated in primitive human acute myelogenous leukemia cells. *Blood* 2001; **98**: 2301–2307.
- 25 Guzman ML, Swiderski CF, Howard DS, Grimes BA, Rossi RM, Szilvassy SJ *et al*. Preferential induction of apoptosis for primary human leukemic stem cells. *Proc Natl Acad Sci USA* 2002; **99**: 16220–16225.
- 26 Xu Q, Thompson JE, Carroll M. mTOR regulates cell survival after etoposide treatment in primary AML cells. *Blood* 2005; **106**: 4261–4268.
- 27 Sullivan KM, Weiden PL, Storb R, Witherspoon RP, Fefer A, Fisher L *et al*. Influence of acute and chronic graft-versus-host disease on relapse and survival after bone marrow transplantation from HLA-identical siblings as treatment of acute and chronic leukemia. *Blood* 1989; **73**: 1720–1728.
- 28 Weiden PL, Sullivan KM, Flournoy N, Storb R, Thomas ED. Antileukemic effect of chronic graft-versus-host disease: contribution to improved survival after allogeneic marrow transplantation. *N Engl J Med* 1981; **304**: 1529–1533.
- 29 Weaver CH, Clift RA, Deeg HJ, Storb R, Appelbaum FR, Bensinger W *et al*. Effect of graft-versus-host disease prophylaxis on relapse in patients transplanted for acute myeloid leukemia. *Bone Marrow Transplantation* 1994; **14**: 885–893.
- 30 Bacigalupo A, Van Lint MT, Occhini D, Gualandi F, Lamparelli T, Sogno G *et al*. Increased risk of leukemia relapse with high-dose cyclosporine A after allogeneic marrow transplantation for acute leukemia. *Blood* 1991; **77**: 1423–1428.
- 31 Bacigalupo A, Vitale V, Corvo R, Barra S, Lamparelli T, Gualandi F *et al*. The combined effect of total body irradiation (TBI) and cyclosporin A (CyA) on the risk of relapse in patients with acute myeloid leukaemia undergoing allogeneic bone marrow transplantation. *Br J Haematol* 2000; **108**: 99–104.
- 32 Greiner J, Schmitt M, Li L, Giannopoulos K, Bosch K, Schmitt A *et al*. Expression of tumor-associated antigens in acute myeloid leukemia: implications for specific immunotherapeutic approaches. *Blood* 2006; **108**: 4109–4117.
- 33 Scheibenbogen C, Letsch A, Thiel E, Schmittel A, Mailaender V, Baerwolf S *et al*. CD8 T-cell responses to Wilms tumor gene product WT1 and proteinase 3 in patients with acute myeloid leukemia. *Blood* 2002; **100**: 2132–2137.
- 34 Di Terlizzi S, Zino E, Mazzi B, Magnani C, Tresoldi C, Perna SK *et al*. Therapeutic and diagnostic applications of minor histocompatibility antigen HA-1 and HA-2 disparities in allogeneic hematopoietic stem cell transplantation: a survey of different populations. *Biol Blood Marrow Transplant* 2006; **12**: 95–101.
- 35 Mutis T, Verdijk R, Schrama E, Esendam B, Brand A, Goulmy E. Feasibility of immunotherapy of relapsed leukemia with *ex vivo*-generated cytotoxic T lymphocytes specific for hematopoietic system-restricted minor histocompatibility antigens. *Blood* 1999; **93**: 2336–2341.
- 36 Rezvani K, Yong AS, Savani BN, Mielke S, Keyvanfar K, Gostick E *et al*. Graft-versus-leukemia effects associated with detectable Wilms tumor-1 specific T lymphocytes following allogeneic stem cell transplantation for acute lymphoblastic leukemia (ALL). *Blood* 2007; **110**: 1924–1932.
- 37 Xue SA, Gao L, Hart D, Gillmore R, Qasim W, Thrasher A *et al*. Elimination of human leukemia cells in NOD/SCID mice by WT1-TCR gene-transduced human T cells. *Blood* 2005; **106**: 3062–3067.
- 38 Vicente D, Lamparelli T, Gualandi F, Occhini D, Raiola AM, Ibatici A *et al*. Improved outcome in young adults with *de novo* acute myeloid leukemia in first remission, undergoing an allogeneic bone marrow transplant. *Bone Marrow Transplantation* 2007; **40**: 349–354.
- 39 Gorin NC, Labopin M, Fouillard L, Meloni G, Frassoni F, Iriondo A *et al*. Retrospective evaluation of autologous bone marrow transplantation vs allogeneic bone marrow transplantation from an HLA identical related donor in acute myelocytic leukemia. A study of the European Cooperative Group for Blood and Marrow Transplantation (EBMT). *Bone Marrow Transplantation* 1996; **18**: 111–117.
- 40 Fung HC, Stein A, Slovak M, O'Donnell M R, Snyder DS, Cohen S *et al*. A long-term follow-up report on allogeneic stem cell transplantation for patients with primary refractory acute myelogenous leukemia: impact of cytogenetic characteristics on transplantation outcome. *Biol Blood Marrow Transplant* 2003; **9**: 766–771.
- 41 Burnett AK, Wheatley K, Goldstone AH, Stevens RF, Hann IM, Rees JH *et al*. The value of allogeneic bone marrow transplant in patients with acute myeloid leukaemia at differing risk of relapse: results of the UK MRC AML 10 trial. *Br J Haematol* 2002; **118**: 385–400.
- 42 Suci S, Mandelli F, de Witte T, Zittoun R, Gallo E, Labar B *et al*. Allogeneic compared with autologous stem cell transplantation in the treatment of patients younger than 46 years with acute myeloid leukemia (AML) in first complete remission (CR1): an intention-to-treat analysis of the EORTC/GIMEMAAML-10 trial. *Blood* 2003; **102**: 1232–1240.
- 43 Jourdan E, Boiron JM, Dastugue N, Vey N, Marit G, Rigal-Huguet F *et al*. Early allogeneic stem-cell transplantation for young adults with acute myeloblastic leukemia in first complete remission: an intent-to-treat long-term analysis of the BGMT experience. *J Clin Oncol* 2005; **23**: 7676–7684.
- 44 Burnett AK. Current controversies: which patients with acute myeloid leukaemia should receive a bone marrow transplantation?—an adult treater's view. *Br J Haematol* 2002; **118**: 357–364.
- 45 Harousseau JL, Cahn JY, Pignon B, Witz F, Milpied N, Delain M *et al*. Comparison of autologous bone marrow transplantation and intensive chemotherapy as postremission therapy in adult acute myeloid leukemia. The Groupe Ouest Est Leucemies Aigues Myeloblastiques (GOELAM). *Blood* 1997; **90**: 2978–2986.
- 46 Dohner K, Schlenk RF, Habdank M, Scholl C, Rucker FG, Corbacioglu A *et al*. Mutant nucleophosmin (NPM1) predicts favorable prognosis in younger adults with acute myeloid leukemia and normal cytogenetics: interaction with other gene mutations. *Blood* 2005; **106**: 3740–3746.
- 47 Gratwohl A, Hermans J, Goldman JM, Arcese W, Carreras E, Devergie A *et al*. Risk assessment for patients with chronic myeloid leukaemia before allogeneic blood or marrow transplantation. Chronic Leukemia Working Party of the European Group for Blood and Marrow Transplantation. *Lancet* 1998; **352**: 1087–1092.
- 48 Sierra J, Storer B, Hansen JA, Martin PJ, Petersdorf EW, Woolfrey A *et al*. Unrelated donor marrow transplantation for acute myeloid leukemia: an update of the Seattle experience. *Bone Marrow Transplantation* 2000; **26**: 397–404.
- 49 Tallman MS, Dewald GW, Gandham S, Logan BR, Keating A, Lazarus HM *et al*. Impact of cytogenetics on outcome of matched unrelated donor hematopoietic stem cell transplantation for acute myeloid leukemia in first or second complete remission. *Blood* 2007; **110**: 409–417.

- 50 Das-Gupta EP, Russell NH, Shaw BE, Pearce RM, Byrne JL. Long-term outcome of unrelated donor transplantation for AML using myeloablative conditioning incorporating pre-transplant alemtuzumab. *Biol Blood Marrow Transplant* 2007; **13**: 724–733.
- 51 Chantry AD, Snowden JA, Craddock C, Peggs K, Roddie C, Craig JI *et al*. Long-term outcomes of myeloablation and autologous transplantation of relapsed acute myeloid leukemia in second remission: a British Society of Blood and Marrow Transplantation Registry study. *Biol Blood Marrow Transplant* 2006; **12**: 1310–1317.
- 52 Lazarus HM, Perez WS, Klein JP, Kollman C, Bate-Boyle B, Bredeson CN *et al*. Autotransplantation versus HLA-matched unrelated donor transplantation for acute myeloid leukaemia: a retrospective analysis from the Center for International Blood and Marrow Transplant Research. *Br J Haematol* 2006; **132**: 755–769.
- 53 Meloni G, Diverio D, Vignetti M, Avvisati G, Capria S, Petti MC *et al*. Autologous bone marrow transplantation for acute promyelocytic leukemia in second remission: prognostic relevance of pretransplant minimal residual disease assessment by reverse-transcription polymerase chain reaction of the PML/RAR alpha fusion gene. *Blood* 1997; **90**: 1321–1325.
- 54 Petti MC, Pinazzi MB, Diverio D, Romano A, Petrucci MT, De Santis S *et al*. Prolonged molecular remission in advanced acute promyelocytic leukaemia after treatment with gemtuzumab ozogamicin (Mylotarg CMA-676). *Br J Haematol* 2001; **115**: 63–65.
- 55 Lo-Coco F, Romano A, Mengarelli A, Diverio D, Iori AP, Moleti ML *et al*. Allogeneic stem cell transplantation for advanced acute promyelocytic leukemia: results in patients treated in second molecular remission or with molecularly persistent disease. *Leukemia* 2003; **17**: 1930–1933.
- 56 Biggs JC, Horowitz MM, Gale RP, Ash RC, Atkinson K, Helbig W *et al*. Bone marrow transplants may cure patients with acute leukemia never achieving remission with chemotherapy. *Blood* 1992; **80**: 1090–1093.
- 57 Oyekunle AA, Kroger N, Zabelina T, Ayuk F, Schieder H, Renges H *et al*. Allogeneic stem-cell transplantation in patients with refractory acute leukemia: a long-term follow-up. *Bone Marrow Transplantation* 2006; **37**: 45–50.
- 58 Blaise D, Maraninchi D, Archimbaud E, Reiffers J, Devergie A, Jouet JP *et al*. Allogeneic bone marrow transplantation for acute myeloid leukemia in first remission: a randomized trial of a busulfan-cytosin versus cytosin-total body irradiation as preparative regimen: a report from the Group d'Etudes de la Greffe de Moelle Osseuse. *Blood* 1992; **79**: 2578–2582.
- 59 Blaise D, Maraninchi D, Michallet M, Reiffers J, Jouet JP, Milpied N *et al*. Long-term follow-up of a randomized trial comparing the combination of cyclophosphamide with total body irradiation or busulfan as conditioning regimen for patients receiving HLA-identical marrow grafts for acute myeloblastic leukemia in first complete remission. *Blood* 2001; **97**: 3669–3671.
- 60 Clift RA, Buckner CD, Thomas ED, Bensinger WI, Bowden R, Bryant E *et al*. Marrow transplantation for chronic myeloid leukemia: a randomized study comparing cyclophosphamide and total body irradiation with busulfan and cyclophosphamide. *Blood* 1994; **84**: 2036–2043.
- 61 Ringden O, Labopin M, Tura S, Arcese W, Iriando A, Zittoun R *et al*. A comparison of busulphan versus total body irradiation combined with cyclophosphamide as conditioning for autograft or allograft bone marrow transplantation in patients with acute leukaemia. Acute Leukaemia Working Party of the European Group for Blood and Marrow Transplantation (EBMT). *Br J Haematol* 1996; **93**: 637–645.
- 62 Ringden O, Remberger M, Ruutu T, Nikoskelainen J, Volin L, Vindelov L *et al*. Increased risk of chronic graft-versus-host disease, obstructive bronchiolitis, and alopecia with busulfan versus total body irradiation: long-term results of a randomized trial in allogeneic marrow recipients with leukemia. Nordic Bone Marrow Transplantation Group. *Blood* 1999; **93**: 2196–2201.
- 63 Ringden O, Ruutu T, Remberger M, Nikoskelainen J, Volin L, Vindelov L *et al*. A randomized trial comparing busulfan with total body irradiation as conditioning in allogeneic marrow transplant recipients with leukemia: a report from the Nordic Bone Marrow Transplantation Group. *Blood* 1994; **83**: 2723–2730.
- 64 Socie G, Clift RA, Blaise D, Devergie A, Ringden O, Martin PJ *et al*. Busulfan plus cyclophosphamide compared with total-body irradiation plus cyclophosphamide before marrow transplantation for myeloid leukemia: long-term follow-up of 4 randomized studies. *Blood* 2001; **98**: 3569–3574.
- 65 Ljungman P, Hassan M, Bekassy AN, Ringden O, Oberg G. High busulfan concentrations are associated with increased transplant-related mortality in allogeneic bone marrow transplant patients. *Bone Marrow Transplantation* 1997; **20**: 909–913.
- 66 Slattery JT, Clift RA, Buckner CD, Radich J, Storer B, Bensinger WI *et al*. Marrow transplantation for chronic myeloid leukemia: the influence of plasma busulfan levels on the outcome of transplantation. *Blood* 1997; **89**: 3055–3060.
- 67 Deeg HJ, Storer B, Slattery JT, Anasetti C, Doney KC, Hansen JA *et al*. Conditioning with targeted busulfan and cyclophosphamide for hemopoietic stem cell transplantation from related and unrelated donors in patients with myelodysplastic syndrome. *Blood* 2002; **100**: 1201–1207.
- 68 Madden T, de Lima M, Thapar N, Nguyen J, Roberson S, Couriel D *et al*. Pharmacokinetics of once-daily IV busulfan as part of pretransplantation preparative regimens: a comparison with an every 6-hour dosing schedule. *Biol Blood Marrow Transplant* 2007; **13**: 56–64.
- 69 Bornhauser M, Storer B, Slattery JT, Appelbaum FR, Deeg HJ, Hansen J *et al*. Conditioning with fludarabine and targeted busulfan for transplantation of allogeneic hematopoietic stem cells. *Blood* 2003; **102**: 820–826.
- 70 Chang C, Storer BE, Scott BL, Bryant EM, Shulman HM, Flowers ME *et al*. Hematopoietic cell transplantation in patients with myelodysplastic syndrome or acute myeloid leukemia arising from myelodysplastic syndrome: similar outcomes in patients with *de novo* disease and disease following prior therapy or antecedent hematologic disorders. *Blood* 2007; **110**: 1379–1387.
- 71 Bensinger WI, Martin PJ, Storer B, Clift R, Forman SJ, Negrin R *et al*. Transplantation of bone marrow as compared with peripheral-blood cells from HLA-identical relatives in patients with hematologic cancers. *N Engl J Med* 2001; **344**: 175–181.
- 72 Eapen M, Horowitz MM, Klein JP, Champlin RE, Loberiza Jr FR, Ringden O *et al*. Higher mortality after allogeneic peripheral-blood transplantation compared with bone marrow in children and adolescents: the Histocompatibility and Alternate Stem Cell Source Working Committee of the International Bone Marrow Transplant Registry. *J Clin Oncol* 2004; **22**: 4872–4880.
- 73 Bahceci E, Read EJ, Leitman S, Childs R, Dunbar C, Young NS *et al*. CD34+ cell dose predicts relapse and survival after T-cell-depleted HLA-identical haematopoietic stem cell transplantation (HSCT) for haematological malignancies. *Br J Haematol* 2000; **108**: 408–414.
- 74 Nakamura R, Bahceci E, Read EJ, Leitman SF, Carter CS, Childs R *et al*. Transplant dose of CD34(+) and CD3(+) cells

- predicts outcome in patients with haematological malignancies undergoing T cell-depleted peripheral blood stem cell transplants with delayed donor lymphocyte add-back. *Br J Haematol* 2001; **115**: 95–104.
- 75 de Lavallade H, Faucher C, Furst S, El-Cheikh J, Vey N, Coso D *et al*. Allogeneic stem cell transplantation after reduced-intensity conditioning in a patient with T-cell prolymphocytic leukemia: graft-versus-tumor effect and long-term remission. *Bone Marrow Transplantation* 2006; **37**: 709–710.
- 76 Grigg AP, Gibson J, Bardy PG, Reynolds J, Shuttleworth P, Koelmeyer RL *et al*. A prospective multicenter trial of peripheral blood stem cell sibling allografts for acute myeloid leukemia in first complete remission using fludarabine-cyclophosphamide reduced intensity conditioning. *Biol Blood Marrow Transplant* 2007; **13**: 560–567.
- 77 Ho AY, Pagliuca A, Kenyon M, Parker JE, Mijovic A, Devereux S *et al*. Reduced-intensity allogeneic hematopoietic stem cell transplantation for myelodysplastic syndrome and acute myeloid leukemia with multilineage dysplasia using fludarabine, busulphan, and alemtuzumab (FBC) conditioning. *Blood* 2004; **104**: 1616–1623.
- 78 Mohty M, de Lavallade H, Ladaique P, Faucher C, Vey N, Coso D *et al*. The role of reduced intensity conditioning allogeneic stem cell transplantation in patients with acute myeloid leukemia: a donor vs no donor comparison. *Leukemia* 2005; **19**: 916–920.
- 79 Taussig DC, Davies AJ, Cavenagh JD, Oakervee H, Syndercombe-Court D, Kelsey S *et al*. Durable remissions of myelodysplastic syndrome and acute myeloid leukemia after reduced-intensity allografting. *J Clin Oncol* 2003; **21**: 3060–3065.
- 80 van Besien K, Artz A, Smith S, Cao D, Rich S, Godley L *et al*. Fludarabine, melphalan, and alemtuzumab conditioning in adults with standard-risk advanced acute myeloid leukemia and myelodysplastic syndrome. *J Clin Oncol* 2005; **23**: 5728–5738.
- 81 Wong R, Giralt SA, Martin T, Couriel DR, Anagnostopoulos A, Hosing C *et al*. Reduced-intensity conditioning for unrelated donor hematopoietic stem cell transplantation as treatment for myeloid malignancies in patients older than 55 years. *Blood* 2003; **102**: 3052–3059.
- 82 Kottaridis PD, Milligan DW, Chopra R, Chakraverty RK, Chakrabarti S, Robinson S *et al*. *In vivo* CAMPATH-1H prevents GvHD following nonmyeloablative stem-cell transplantation. *Cytotherapy* 2001; **3**: 197–201.
- 83 Mohty M, Bay JO, Faucher C, Choufi B, Bilger K, Tournilhac O *et al*. Graft-versus-host disease following allogeneic transplantation from HLA-identical sibling with antithymocyte globulin-based reduced-intensity preparative regimen. *Blood* 2003; **102**: 470–476.
- 84 Mohty M, Jacot W, Faucher C, Bay JO, Zandotti C, Collet L *et al*. Infectious complications following allogeneic HLA-identical sibling transplantation with antithymocyte globulin-based reduced intensity preparative regimen. *Leukemia* 2003; **17**: 2168–2177.
- 85 Perez-Simon JA, Kottaridis PD, Martino R, Craddock C, Caballero D, Chopra R *et al*. Nonmyeloablative transplantation with or without alemtuzumab: comparison between 2 prospective studies in patients with lymphoproliferative disorders. *Blood* 2002; **100**: 3121–3127.
- 86 de Lima M, Couriel D, Thall PF, Wang X, Madden T, Jones R *et al*. Once-daily intravenous busulfan and fludarabine: clinical and pharmacokinetic results of a myeloablative, reduced-toxicity conditioning regimen for allogeneic stem cell transplantation in AML and MDS. *Blood* 2004; **104**: 857–864.
- 87 Matthews DC, Martin PJ, Nourigat C, Appelbaum FR, Fisher DR, Bernstein ID. Marrow ablative and immunosuppressive effects of 131I-anti-CD45 antibody in congenic and H2-mismatched murine transplant models. *Blood* 1999; **93**: 737–745.
- 88 Dominiotto A, Pozzi S, Miglino M, Albarracin F, Piaggio G, Bertolotti F *et al*. Donor lymphocyte infusions for the treatment of minimal residual disease in acute leukemia. *Blood* 2007; **109**: 5063–5064.
- 89 Dey BR, McAfee S, Colby C, Sackstein R, Saidman S, Tarbell N *et al*. Impact of prophylactic donor leukocyte infusions on mixed chimerism, graft-versus-host disease, and antitumor response in patients with advanced hematologic malignancies treated with nonmyeloablative conditioning and allogeneic bone marrow transplantation. *Biol Blood Marrow Transplant* 2003; **9**: 320–329.
- 90 Ogawa H, Tamaki H, Ikegame K, Soma T, Kawakami M, Tsuboi A *et al*. The usefulness of monitoring WT1 gene transcripts for the prediction and management of relapse following allogeneic stem cell transplantation in acute type leukemia. *Blood* 2003; **101**: 1698–1704.
- 91 Miller JS, Weisdorf DJ, Burns LJ, Slungaard A, Wagner JE, Verneris MR *et al*. Lymphodepletion followed by donor lymphocyte infusion (DLI) causes significantly more acute graft-versus-host disease than DLI alone. *Blood* 2007; **110**: 2761–2763.
- 92 Marks DI, Lush R, Cavenagh J, Milligan DW, Schey S, Parker A *et al*. The toxicity and efficacy of donor lymphocyte infusions given after reduced-intensity conditioning allogeneic stem cell transplantation. *Blood* 2002; **100**: 3108–3114.
- 93 Olavarria E, Siddique S, Griffiths MJ, Avery S, Byrne JL, Piper KP *et al*. Posttransplantation imatinib as a strategy to postpone the requirement for immunotherapy in patients undergoing reduced-intensity allografts for chronic myeloid leukemia. *Blood* 2007; **110**: 4614–4617.
- 94 Barker JN, Weisdorf DJ, DeFor TE, Blazar BR, Miller JS, Wagner JE. Rapid and complete donor chimerism in adult recipients of unrelated donor umbilical cord blood transplantation after reduced-intensity conditioning. *Blood* 2003; **102**: 1915–1919.
- 95 Schmid C, Schleuning M, Ledderose G, Tischer J, Kolb HJ. Sequential regimen of chemotherapy, reduced-intensity conditioning for allogeneic stem-cell transplantation, and prophylactic donor lymphocyte transfusion in high-risk acute myeloid leukemia and myelodysplastic syndrome. *J Clin Oncol* 2005; **23**: 5675–5687.
- 96 Schmid C, Schleuning M, Schwerdtfeger R, Hertenstein B, Mischak-Weissinger E, Bunjes D *et al*. Long-term survival in refractory acute myeloid leukemia after sequential treatment with chemotherapy and reduced-intensity conditioning for allogeneic stem cell transplantation. *Blood* 2006; **108**: 1092–1099.