Mortality Hazard Functions as Related to Neutropenia at Different Times After Marrow Transplantation

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We characterized the relationship between severe neutropenia and risk of death in 2,276 patients after marrow transplantation to define objective and clinically relevant criteria that could be used to judge the timing and potential value of interventions designed to improve survival in patients with delayed initial engraftment. Proportional hazards models were used to estimate the relative risk of death before day 100 among patients alive on any given day with an absolute neutrophil count (ANC) less than 100/μL compared with those alive on the same day with an ANC ≥ 100/μL. Between day 10 and 14, the risk ratio remained close to 1.0, indicating that the risk of death before day 100 for patients with an ANC less than 100/μL was similar to that for patients with an ANC ≥ 100/μL. Between day 15, when 38% of patients had an ANC less than 100/μL, and day 26, when 3.8% of patients had an ANC less than 100/μL, the risk ratio showed an overall upward trend, indicating that patients with an ANC less than 100/μL had a higher risk of death before day 100 than those with an ANC ≥ 100/μL. Thereafter, the risk ratio fluctuated between 2.01 and 5.78, indicating consistently higher risks of mortality in patients with severe neutropenia. However, allogeneic and autologous transplant recipients each had distinctive risk ratio patterns. These results could be helpful in deciding the appropriate timing for treatment given to improve graft function after marrow transplantation.

ENGRAFTMENT AFTER marrow transplantation is often defined as an event characterized by the number of days elapsed before the absolute neutrophil count (ANC) surpasses a specified level.[1-4] Because the ANC often fluctuates before starting to increase consistently, engraftment is usually defined as the first in a series of consecutive days with the ANC above some minimum value. Although this type of definition allows engraftment to be recognized retrospectively, the clinician faced with a patient who has an ANC above the specified minimum on any given day cannot predict whether the ANC will exceed that level on subsequent days and therefore cannot determine whether engraftment has yet occurred.

Prolonged neutropenia is known to be associated with poor clinical outcome after marrow transplantation. The present study was intended not to demonstrate this well-accepted phenomenon, but rather to estimate the time at which neutropenia changes from being an unavoidable initial phase of engraftment to becoming a real clinical concern in its own right. Uncertainty concerning the timing of this transition has made it difficult to judge when interventions should be made to improve engraftment in patients with prolonged neutropenia. Thus, we have characterized the relationship between neutropenia and the risk of death to estimate the appropriate timing and potential benefits of treatment to improve graft function after marrow transplantation.

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PATIENTS AND METHODS

Patients. Patients (N = 2,276) transplanted between 1987 and 1995 at the Fred Hutchinson Cancer Research Center were evaluated in this study (Table 1). Patients were divided into three groups: those with human leukocyte antigen (HLA)-matched sibling or identical twin donors, those with HLA-mismatched related donors or unrelated donors, and autologous transplant recipients. Indications for marrow transplantation and risk categories are summarized in Table 1.

Transplant procedure. All protocols and consent forms used in patients in this study were approved by the institutional review board. Patients received myeloablative pretransplant regimens that contained various combinations of cyclophosphamide, busulfan, thiotepa, etoposide, carmustine, and total-body irradiation. The choice of conditioning regimen and the administration of intrathecal methotrexate before and after transplant depended on diagnosis, disease status, and type of donor. Patients who had major ABO incompatibility with the donor underwent plasma exchange to reduce the titer of anti-A or anti-B isoagglutinins, or the grafts were red blood cell–depleted. Allogeneic transplant recipients were treated with graft-versus-host disease (GVHD) prophylaxis regimens that contained cyclosporine, methotrexate, or the combination, with or without glucocorticoids. All patients treated in laminar air-flow isolation had enteric decontamination with oral nonabsorbable antibiotics. When the ANC decreased to less than 500/μL, patients received prophylactic intravenous antibiotics (ceftizoxime and mezlocillin until April 1991 and ceftazidime thereafter). Acyclovir or gancyclovir was given for viral prophylaxis, fluconazole was given for fungal prophylaxis, intravenous immunoglobulin was given as supplementation or replacement, and treatment with hematopoietic growth factor was given according to clinical trials or standard practices active at the time of transplantation.

Study design. The ANC was assessed daily for inpatients and at least twice weekly for outpatients. On any given day, surviving patients were divided in two groups: those whose most recent ANC was ≥100/μL and those with values less than 100/μL. By not incorporating any information about the subsequent course of engraftment, we sought to mimic the information available to clinicians faced with the question of whether to intervene on behalf of patients with severe neutropenia persisting at any given time point after transplantation. Conditional hazards ratios and 95% confidence limits were estimated using proportional hazards models[5] that included all deaths from the day of evaluation to day 100 regardless of the causes. Censoring at day 100 follows from the premise that most deaths related to delayed engraftment would be expected to occur

Table 1. Patient Characteristics (N = 2,276)

Characteristic	No.	%
Patient age		
Mean	33	
Range	3.6 mo-68 yr	
Male	1,267	56
Female	1,009	44
Type of transplant		
Identical twin	28	1
HLA-matched related	1,053	46
HLA-mismatched related	321	14
Unrelated	454	20
Autologous*	420	18
Diagnosis		
Acute myeloid leukemia	494	22
Acute lymphoid leukemia	292	13
Chronic myeloid leukemia	665	29
Non-Hodgkin's lymphoma	234	10
Hodgkin's disease	83	4
Myelodysplasia	166	7
Aplastic anemia	78	3
Myeloma	85	4
Breast cancer	71	3
Other	108	5
Transplant risk category†		
Low risk	1,310	58
High risk	966	42

* Marrow for 32 patients with acute myeloid leukemia was treated with 4-hydroperoxycyclophosphamide.

† The high-risk category included acute leukemia or lymphoma in relapse, chronic myeloid leukemia in accelerated phase or blast crisis, and myeloma. All other diagnoses were categorized as low risk.

before day 100. Results from the models indicate the relative risk of death before day 100 among patients with an ANC less than 100/μL on a given day as compared with those with an ANC $\geq$ 100/μL on the same day.

RESULTS

Mortality hazard as related to neutropenia. A method was devised to analyze mortality hazard as related to the presence of severe neutropenia at different times after marrow transplantation. To illustrate this method, Fig 1 compares cumulative mortality rates for patients with an ANC $\geq$ or less than 100/μL on days 13 and 26 after transplantation. The data are presented as the negative logarithm of the proportion of patients in each group who remained alive on sequential days after days 13 and 26, respectively. The slopes of these curves represent the mortality hazards and appear to be indistinguishable for the two groups on day 13, but different on day 26 when the group with ANCs less than 100/μL clearly had a higher mortality hazard than the group with ANCs $\geq$ 100/μL.

To determine when the mortality hazards diverged for patients with an ANC less than or $\geq$100/μL, a similar analysis was undertaken for each day after transplantation between days 10 and 35 (Fig 2A). Between day 10 and day 14, the risk ratio or instantaneous relative risk remained close to 1.0, which indicates that the risk of death before day 100 was similar for patients with an ANC less than 100/μL and for those with an ANC $\geq$ 100/μL. Between day 15, when 38% of patients had an ANC less than 100/μL, and day 26, when 3.8% of patients had an ANC less than 100/μL, the risk ratio showed an overall upward trend, which indicates that patients with an ANC less than 100/μL had a higher risk of death before day 100 than those with an ANC $\geq$ 100/μL. Between days 27 and 35, the risk ratio fluctuated between 2.21 and 2.64. Risk ratios on days 40, 45, 50, 55, 60, 65, 70, and 75 ranged between 2.01 and 5.78 (mean, 3.78), and the lower limit of the 95% confidence interval for the risk ratio was greater than 1.0 on 6 of the 8 days tested during this time frame, which suggests that the risk of death before day 100 was consistently higher for patients with ANCs less than 100/μL than for those with ANCs $\geq$ 100/μL (data not shown).

Hazard analysis with different types of transplants. Results were not uniform for patients with different types of transplants. With HLA-matched allotransplants, the risk ratio increased gradually until a strong upward trend became apparent on day 20, when 12% of patients had an ANC less than 100/μL (Fig 2B). The correlation between neutropenia and mortality was more striking in this subgroup than in the entire population of patients. Risk ratios at 5-day intervals between days 40 and 75 ranged between 0 and 8.15 (mean, 3.17) (data not shown). The lower limit of the 95% confidence interval for the risk ratio was greater than 1.0 on only 2 of the 8 days tested. Risk ratio estimates after day 35 fluctuated widely because the number of patients with an ANC less than 100/μL was too small for valid analysis.

With unrelated or HLA-mismatched related transplants, the risk ratio remained between 1.37 and 1.62 after day 18 until an overall upward trend began on day 23, when 9.9% of patients had ANCs less than 100/μL (Fig 2C). Risk ratios at 5-day intervals between days 40 and 65 ranged between 3.33 and 8.70 (mean, 4.78) (data not shown). The lower limit of the 95% confidence interval for the risk ratio was greater than 1.0 until day 55, when estimates of the risk ratio became less precise because very few patients had ANCs less than 100/μL.

With autologous transplants, the risk ratio did not show a consistent rising trend as observed with allogeneic transplants. Instead, the risk ratio fluctuated between 0.91 and 1.74 (mean, 1.31) from day 13 to 35 (Fig 2D). The proportion of patients with severe neutropenia after day 25 was greater in this group than in the other two groups. Risk ratios at 5-day intervals between days 40 and 75 ranged between 1.62 and 7.27 (mean, 4.25) (data not shown). Risk ratios for all but one of the days tested during this interval were consistently $\geq$2.5, and the lower limit of the 95% confidence intervals for the risk ratio estimate was greater than 1.0 on 4 of the 8 days tested during this time frame, which suggests that severe neutropenia at times beyond 40 days after transplantation represents a mortality risk for autologous recipients.

Risk of mortality as related to severity of neutropenia. To assess the relationship between severity of neutropenia and risk of mortality, we analyzed results for patients who were alive on day 25 after transplantation from an HLA-

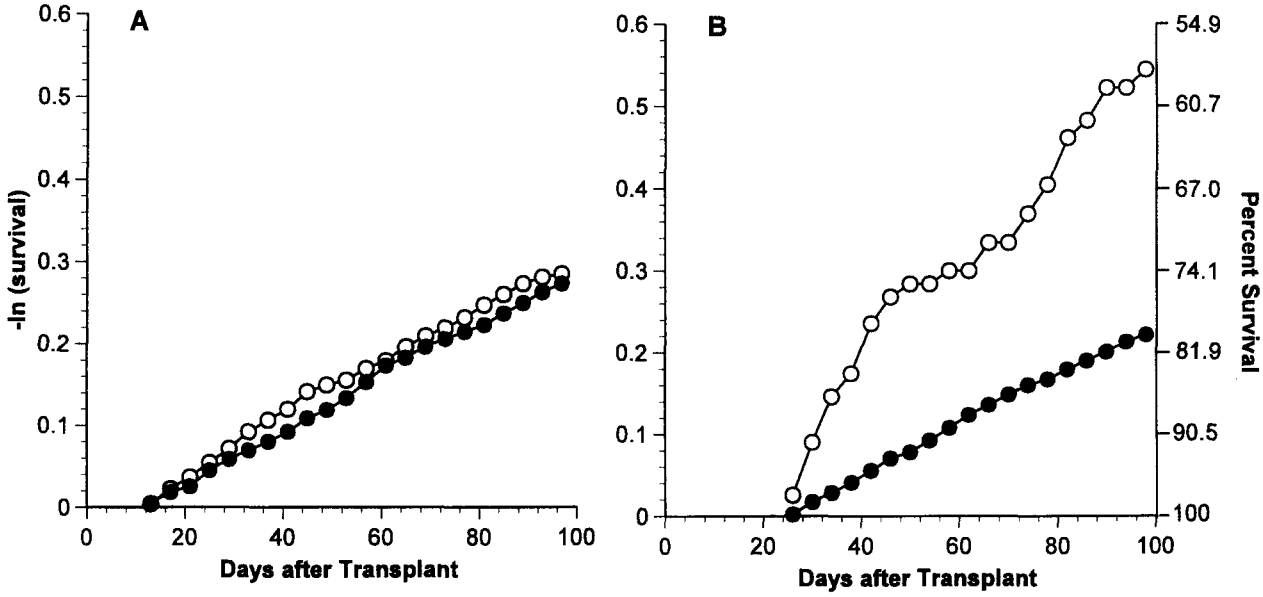

Fig 1. Negative logarithmic plots of survival among patients alive on day 13 (A) and 26 (B) after transplantation. Results for patients with an ANC < 100/μL (●) on the indicated day are compared with those for patients with an ANC ≥ 100/μL (○). The axis on the right of the figure indicates percent survival corresponding to the values for −ln (survival) shown on the axis on the left.

identical related donor. This population was selected because of the relatively large number of patients available for analysis and the strong association between severe neutropenia on day 25 and risk of mortality before day 100 (Fig 2B). Mortality risks for patients with ANCs between 100 and 199/μL, 200 and 499/μL, and 500 and 999/μL on day 25 were closely similar (Fig 3). Patients with an ANC greater than 1,000/μL appeared to have a slightly higher risk than those with an ANC of 100 to 999/μL. Patients with an ANC less than 100/μL on day 25 had a much higher risk than all other groups, which suggests that an ANC of 100/μL represents a critical threshold for the assessment of initial graft function after marrow transplantation.

DISCUSSION

Delayed initial engraftment after marrow transplantation has often been defined according to criteria such as an ANC less than 100/μL on day 21 or day 28.[1,4] Criteria such as these are based on observations that most patients have evidence of engraftment by these time points. The present study was motivated by the need to define more objective and clinically relevant criteria that could be used to judge the appropriate timing and potential value of treatment for patients with delayed initial engraftment after marrow transplantation. The choice of an ANC ≥ or less than 100/μL as the threshold for analysis in this study was based on a desire to define delayed initial engraftment at the earliest possible time after transplantation. Based on the strong association between survival and ANC ≥ or less than 100/μL for patients alive on day 25 after transplantation from an HLA-identical related donor, a threshold value of 100/μL represents an appropriate criterion to consider in evaluating the potential benefits of treatment for patients with delayed engraftment.

Because we defined groups according to the ANC for each specific day without considering the values for any previous days, our results represent only a first approximation of the relationship between the duration of neutropenia and the risk of mortality after marrow transplantation. Neutrophil counts can fluctuate repeatedly above and below 100/μL before definitive engraftment occurs. Patients with such fluctuations in ANC might have a lower risk of mortality than those with an ANC consistently less than 100/μL.

Several explanations might account for differences in mortality hazard patterns among patients with delayed engraftment after allogeneic or autologous marrow transplantation. We surmise that the extremely high mortality associated with severe neutropenia after allogeneic transplantation partly reflects the often irreversible nature of graft failure caused by rejection. GVHD and the exacerbation of mucosal injury by GVHD[6,7] could predispose allogeneic recipients to higher risks of fatal infection during neutropenia as compared with autologous transplant recipients, while other causes of death such as regimen-related toxicity and recurrent malignancy might diminish the relative importance of neutropenia in autologous recipients.[8-12] Alternatively, it is possible that granulocyte counts show more fluctuation above and below 100/μL before definitive engraftment in autologous recipients than in allogeneic recipients.

Our results suggest that intervention to improve graft function earlier than day 21 after transplantation could be warranted in patients with an ANC less than 100/μL. While it is clear that severe neutropenia cannot be singled out as a mortality risk factor during the first 2 weeks after transplantation, results for the entire population analyzed in the present study suggest that a small survival benefit could be gained by treatments that reliably increase the ANC to ≥100/

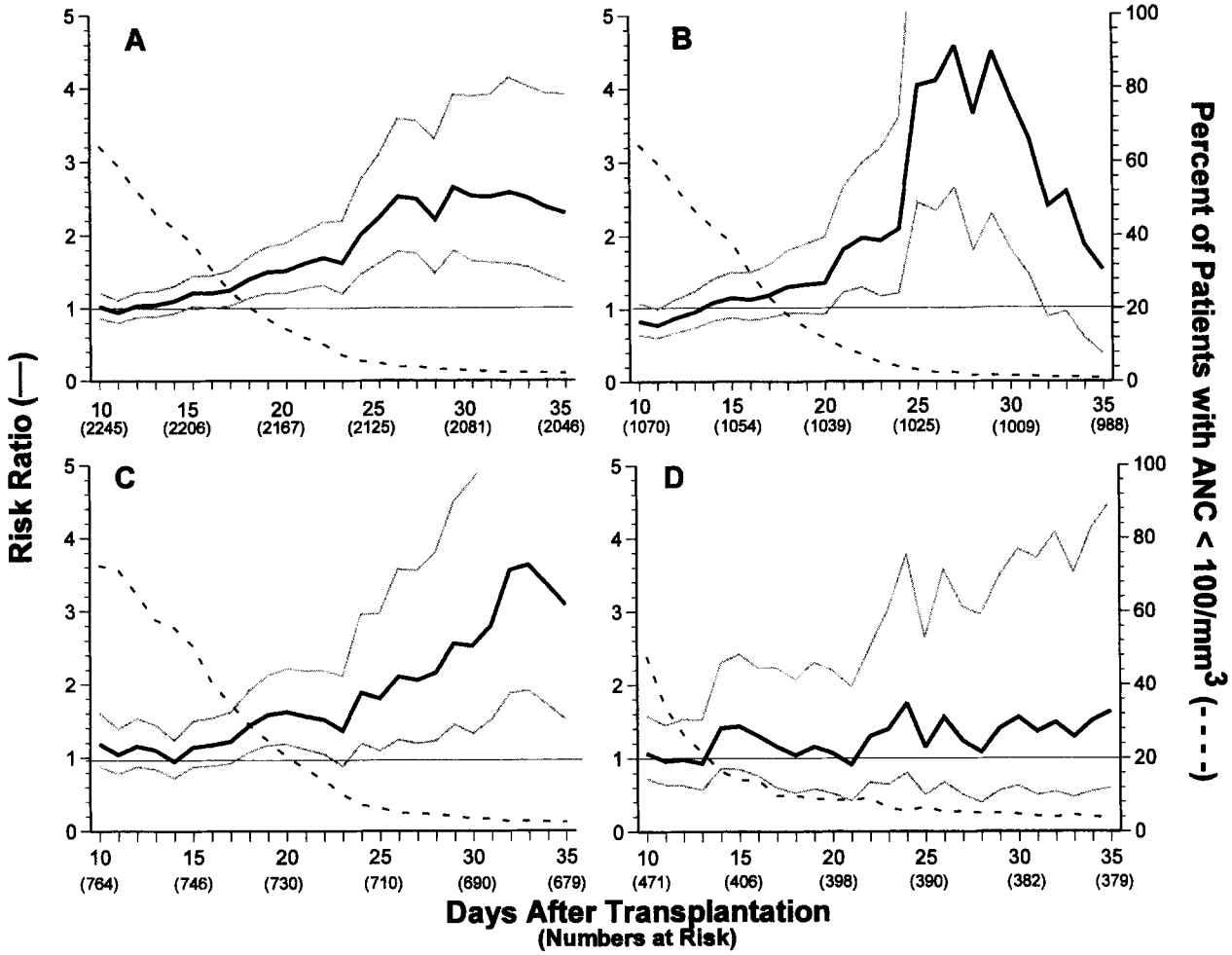

Fig 2. Conditional risk ratios (——) and 95% confidence limits (——) for death before day 100 in patients with an ANC < 100/μL compared with patients with an ANC ≥ 100/μL. Risk ratios for the scale shown at the left were calculated on consecutive days for the entire patient group (A), for those with HLA-identical related donors (B), for those with unrelated or HLA-mismatched related donors (C), and for autologous transplant recipients (D). The ratio of 1.0 delineated in each panel indicates equivalent risks for patients with an ANC < 100/μL and for those with an ANC ≥ 100/μL. The scale shown at the right depicts the proportions of patients with an ANC < 100/μL on each day after transplantation (- - -). Numbers of patients at risk at 5-day intervals beginning on day 10 are indicated in parentheses at the bottom of each panel.

μL after day 14. The degree of benefit would likely be greater if treatment were initiated at later time points when survival differences between patients with ANCs ≥ or less than 100/μL are more striking. However, given that the response to treatment might not occur immediately, an argument can be made for early intervention, especially in patients transplanted with allogeneic marrow, in whom the relative risk of mortality associated with delayed engraftment appears to be greater than in autologous transplant recipients. Although early intervention would not be expected to improve survival in autologous recipients, such intervention might yield other benefits such as a reduced risk of infection.

If an ANC less than 100/μL at day 14 after transplantation were used as the deciding criterion, approximately 40% to 50% of allogeneic transplant recipients could qualify for preemptive treatment to improve graft function. Our retrospective results suggest that the benefit of this approach would be maximized if treatment were able to induce a prompt increase in ANC to levels consistently greater than 100/μL in a large fraction of patients, especially in those who would otherwise persist with severe neutropenia beyond 25 days after transplantation. However, this expectation must be tempered by the limitation that treatment with an agent such as a hematopoietic growth factor would likely not improve survival in patients destined to have prolonged severe neutropenia resulting from rejection. In addition, correction of neutropenia would likely not improve survival in patients with severe regimen-related toxicity, multiorgan failure, or other irreversible life-threatening complications.[8-12]

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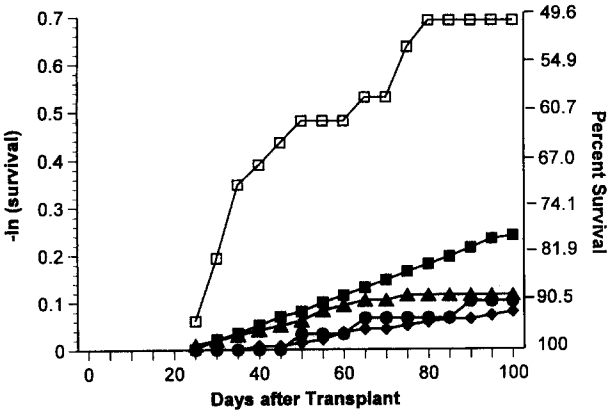

Fig 3. Negative logarithmic plots of survival among patients alive on day 25 after transplantation from an HLA-identical related donor. Results for patients with an ANC < 100/μL (□) (n = 34) on day 25 are compared with those for patients with ANC values of 100 to 199/μL (●) (n = 31), 200 to 999/μL (▲) (n = 101), 500 to 999/μL (♦) (n = 144), and ≥ 1,000/μL (■) (n = 715).

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