Successful hematopoietic stem cell transplantation (HSCT), both autologous and allogeneic, requires a rapid and durable engraftment, with neutrophil (>500/μL) and platelet (>20,000/μL) reconstitution. Factors influencing engraftment after autologous or allogeneic HSCT were investigated in 65 patients: 25 autologous peripheral stem cell transplantation (PBSCT) and 40 allogeneic bone marrow transplantation (BMT) patients. The major factor affecting engraftment was the graft source for HSCT. Neutrophil and platelet recovery were more rapid in autologous PBSCT than in allogeneic BMT [neutrophil occurring in median on day 10.00 (09.00/11.00) and 19.00 (16.00/23.00) and platelet on day 11.00 (10.00/13.00) and 21.00 (18.00/25.00), respectively; p < 0.0001]. The type of disease also affected engraftment, where multiple myeloma (MM) and lymphoma showed faster engraftment when compared with leukemia, syndrome myelodysplastic (SMD) and aplastic anemia (AA) and MM presented the best overall survival (OS) in a period of 12 months. Other factors included the drug used in the conditioning regimen (CR), where CBV, melphalan (M-200) and FluCy showed faster engraftment and M-200 presented the best OS, in a period of 12 months and age, where 50–59 years demonstrated faster engraftment. Sex did not influence neutrophil and platelet recovery.

Introduction

Hematopoietic stem cell transplantation (HSCT) is a widely accepted therapeutic modality for a number of malignant, hematologic, immunologic and genetic diseases.1 This therapy consists of the intravenous infusion of hematopoietic progenitor cells to reestablish marrow function in patients with damaged or defective bone marrow. Allogeneic HSCT involves the transfer of marrow from a donor to another person. It has been preferentially performed in patients with under 60 years of age, because of a higher incidence of graft-versus-host disease in older patients. Autologous HSCT involves the use of the patient’s own marrow to reestablish hematopoietic cell function after the administration of high-dose chemotherapy and can be safely performed in older patients, because there is no risk of graft-versus-host disease as a complication.2

Autologous transplantation has been performed particularly in patients with multiple myeloma or lymphoma3,4 whereas allogeneic transplantation is preferred for patients with leukemias or myeloproliferative diseases.5

Graft failure or graft rejection after HSCT remains a severe complication and may be manifested as either lack of initial engraftment of donor cells, or loss of donor cells after initial engraftment. Rejection is a major cause of graft failure. Other possible causes include viral infections, drug toxicity and sepsis. The use of granulocyte-colony-stimulating factor (G-CSF) mobilized peripheral blood stem cell transplantation (PBSCT) instead of bone marrow, may reduce the rates of graft rejection,6 enhanced engraftment and accelerated hematopoietic recovery7,9 because PBSCT affords about 10-fold more T cells,10,11 compared to bone marrow, and a 2-fold higher CD34+ cell dose. However, most studies have reported a higher risk of chronic graft-versus-host disease (GVHD) with allogeneic PBSCTs.12,13 In autologous HSCT, the major problem is that malignant cells, with their inherent resistance to chemotherapy, might survive and their reinfusion probably contributes to the high incidence of relapse observed after this therapy.14

Administration of high doses of chemotherapy (including busulfan, cyclophosphamide and etoposide), with or without
total body irradiation (TBI), is a feature of HSCT protocols, for malignant and non-malignant diseases. These myeloablative CRs can be more or less toxic, depending on the drug utilized. In addition, the speed and durability of engraftment are important for successful HSCT with both neutrophil (>500/μL) and platelet (>20,000/μL) reconstitution. It is important to improve our knowledge about the different factors affecting hematopoetic recovery after HSCT to further enhance the safety of this procedure. The objective of this study is to identify predictive factors affecting a rapid engraftment in patients undergoing autologous and allogeneic HSCT.

Results

In 65 (100%) patients examined, 3 (04.62%) did not present engraftment and in 62 (93.94%) neutrophil and platelet engraftment occurred. Kruskal-Wallis ANOVA followed by Mann-Whitney U test demonstrated that in terms of neutrophil and platelet engraftment there was no significant difference between the sexes. The Fisher test also showed no difference between the sexes in terms of OS in the periods of <6 months and 6–12 months (Table 2).

There were statistically significant differences in neutrophil and platelet engraftment between patients ≤20 years old (neutrophil) and 20–29 years old (platelet) from 50–59 years old, where neutrophil engraftment occurred more rapidly in the 50–59 year-old group, occurring in median on day 11.00 (10.00/18.00) and 19.00 (16.00/29.00), respectively and platelet engraftment in median on day 13.00 (10.00/21.00) and 22.00 (13.00/25.00), respectively (p < 0.005), (Table 3).

The effect of disease type on engraftment was investigated. Neutrophil and platelet engraftment occurred more rapidly in MM than in leukemias, SMD and AA, neutrophil engraftment occurring in median on day 11.00 (10.00/18.00) and 19.00 (16.00/20.00), respectively and platelet engraftment in median on day 11.00 (10.00/13.00), 21.00 (17.00/25.00), 23.00 (21.00/26.50) and 24.00 (20.00/29.00), respectively (p < 0.005); Neutrophil and platelet engraftment occurred more rapidly for lymphomas than for leukemias, SMD and AA, neutrophil engraftment occurring on day 10.00 (09.00/15.50), 18.00 (16.00/20.00), 20.50 (18.50/25.50) and 22.00 (18.00/29.00), respectively and platelet engraftment occurring on day 13.00 (10.50/16.00), 21.00 (17.00/25.00), 23.00 (21.00/26.50) and 24.00 (20.00/29.00), respectively (p < 0.005); OS presented a statistically significant difference between MM and SMD at 6–12 months, being 100% and 62.5%, respectively (p < 0.05), (Table 4). No significant differences where shown for engraftment time or for OS among the different types of leukemias or among the different types of lymphomas (data not shown).

Neutrophil and platelet engraftment occurred more rapidly in autologous PBSCT than in allogeneic BMT. Neutrophil engraftment occurred in median on day 10.00 (09.00/11.00) and 19.00 (16.00/23.00), respectively and platelet engraftment occurred in median on day 11.00 (10.00/13.00) and 21.00 (18.00/25.00), respectively (p < 0.0001), (Table 5). In relation to the effect of CR on engraftment, neutrophil and platelet engraftment occurred more rapidly in patients that used CBV, when compared to those using FluCy, CyTBI, BuCy120, BuCy200 and Cy200, neutrophil engraftment occurring in median on day 09.00 (09.00/10.50), 12.50 (11.50/16.00), 20.00 (17.50/21.00), 19.00 (18.00/23.00), 23.50 (22.00/25.00) and 19.00 (18.00/29.00) (p < 0.05), respectively; and neutrophil engraftment was more rapid in patients using M-200 when compared to those using CyTBI, BuCy120, BuCy200 and Cy200 and FluCy than CyCy200 (p < 0.05), occurring in median on day 11.00 (10.00/11.50) for M-200. Platelet engraftment occurred in median on day 11.50 (10.00/13.00) for CBV, 15.50 (13.50/17.50) for FluCy, 20.00 (18.00/22.00) for CyTBI, 22.00 (21.00/25.00) for BuCy120 and 25.00 (24.00/26.00) for BuCy200; was more rapid in patients that used M-200 in comparison to those using FluCy, CyTBI, BuCy120 and BuCy200, occurring in median on day 11.00 (10.00/13.00) for M-200; more rapid in FluCy than in BuCy120, (p < 0.05). OS was significantly different between M-200 with CyTBI and BuCy200 in the period of 6–12 months, being 100%, 50% and 33.33% respectively (p < 0.05), (Table 6).

Discussion

Mobilized peripheral blood stem-cell transplantation (PBSCT) has largely replaced the use of bone marrow as the preferred source of hematopoietic stem-cell in autologous transplant and has been increasingly used in the allogeneic procedure. PBSCT is better than BMT with regard to hospitalization period, transplant-related mortality and main produces a more rapid hematopoietic reconstitution than that using marrow derived stem-cells, probably because of the higher content of committed hematopoietic precursor cells. In line with this, we and other authors found a more rapid hematologic recovery, with a shorter interval of time for neutrophil and platelet engraftment with PBSCT when compared with BMT. This finding in our case specific can be due that the PBSCT enhance engraftment and accelerate hematopoietic recovery and all patients with PBSCT realized autologous transplantation that is a more simple procedure when compared with BMT, because involves the use of the patient's own marrow to reestablish hematopoietic cell function.

Allogeneic HSCT has been preferentially performed in patients under the age of 60 years, being that 40–50 is considered old for this procedure, because of a higher incidence of complications in the older patients. On the other hand, autologous HSCT can be performed safely in older patients, because it is a more simple procedure and presents fewer complications. In terms of the effect of age on neutrophil and platelet engraftment, we found that engraftment was more rapid in the 50–59 year-old group. This finding could be due to the fact that the older group was also linked with some more favorable variables. For example, most of the patients in the older group underwent the more favorable autologous PBSCT (Table 3) and received M-200 and CBV, which were also associated with a faster engraftment.

In relation to the effect of disease type on engraftment, neutrophil and platelet engraftment occurred more rapidly in MM and in...
lymphomas when compared with leukemias, SMD and AA. This may also be due to association with other more favorable factors, i.e., patients with MM and Hodgkin or no-Hodgkin lymphoma, mainly underwent autologous PBSCT, with M-200 and CBV, which showed more rapid engraftment than the allogeneic HSCT with other drugs utilized in the CRs for leukemias, SMD and AA patients. The same occurred in OS, where patients with MM showed a better OS than that for SMD, probably because of its association with variables such as autologous PBSCT, and the use of M-200. Carral et al.20 showed that the leukemia group had a delayed hematopoietic recovery when compared with MM and lymphomas and suggested that residual leukemia in the patient may also contribute to delayed engraftment.

The objective of clinical research is to find chemotherapy drugs that produce minimal toxic effects on normal tissue. Today, chemotherapy is performed with a combination of drugs, because it allows maximal cellular death with tolerable toxic limits and avoids cell resistance.21 In this study, it can be observed that CBV, M-200 and FluCy produced better engraftment than the other CRs with high doses of cyclophosphamide or the presence of busulfan cytostatic drugs. The OS was also better in M-200 patients than in those using other drugs such as CyTBI and BuCy200, probably because of Cy and TBI toxicity. In the case of FluCy, Srinivasan et al.22 demonstrated that fludarabine associated to cyclophosphamide helped achieve engraftment, allowing a reduction of the traditionally high dose of cyclophosphamide. The same authors indicated that fludarabine increased engraftment rates, and showed minimal morbidity.23-26 George et al.27 observed that fludarabine improved OS in children with AA.

In summary, our study shows that the most important variable influencing engraftment in patients undergoing HSCT was the source, being that autologous PBSCT resulted in faster neutrophil and platelet engraftment. Another important variable was the CR, where CBV, M-200 and FluCy produced faster neutrophil and platelet engraftment. Other variables such as disease type and age that showed an influence were probably associated with the source and CR utilized in HSCT.

**Materials and Methods**

**Patients.** This is a retrospective study, where 25 patients who were undergoing autologous HSCT and 40 undergoing allogeneic HSCT, in the onco-hematology unit of the Hospital Universitario of the Universidade Federal de Santa Maria, Brazil, between January 2006 and December 2007, were investigated. The information about factors affecting the engraftment in HSCT patients were taken from patient files.

The present study was approved by the Human Ethical Committee of the Universidade Federal de Santa Maria, protocol number: 0152.0.243.000-06.

Table 1 shows the pre-transplant characteristics, graft source and the conditioning regimens of patients undergoing autologous and allogeneic HSCT.

**Table 2 Influence of sex on engraftment**

|               | Male (n = 38) | Female (n = 27) |
|---------------|--------------|----------------|
| No engraftment| 01           | 02             |
| Engraftment   | 37           | 25             |
| Neutrophil engraftment (day) | 13.00 (10.00/19.00) | 16.00 (11.00/19.00) |
| Platelet engraftment (day) | 16.00 (12.00/21.00) | 20.00 (13.00/23.00) |
| OS (<6 months) | 36/38 (94.74%) | 23/27 (85.18%) |
| OS (6–12 months) | 31/38 (81.58%) | 21/27 (77.78%) |

OS: overall survival; Neutrophil and platelet engraftment were expressed as median (lower/upper quartile).
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transplantation (days minus 8 to minus 5) + cyclophosphamide (60 mg/m²), given for 2 consecutive days, starting 4 days before the transplantation (days minus 4 to minus 3) for acute and

Table 3 Influence of age on engraftment

| Age Group         | No engraftment | Engraftment | Neutrophil engraftment (day) | Platelet engraftment (day) | OS (<6 months) | OS (6–12 months) |
|------------------|----------------|-------------|-------------------------------|--------------------------|----------------|------------------|
| <20 years (n = 10) | 01             | 09          | 19.00 (16.00/29.00)           | 21.00 (17.00/25.00)       | 12/12 (100%)  | 12/12 (100%)     |
| 20–29 years (n = 10) | 01             | 10          | 18.50 (11.00/24.00)           | 22.00 (13.00/25.00)       | 19/21 (90.48%) | 19/21 (90.48%)   |
| 30–39 years (n = 10) | 01             | 10          | 17.00 (13.00/19.00)           | 15.00 (10.00/23.00)       | 14/16 (87.50%) | 14/16 (87.50%)   |
| 40–49 years (n = 15) | 01             | 14          | 15.00 (11.00/21.00)           | 18.50 (17.00/25.00)       | 10/10 (100%)  | 10/10 (100%)     |
| 50–59 years (n = 16) | 01             | 15          | 11.00 (10.00/18.00)           | 13.00 (10.00/21.00)       | 15/15 (93.33%) | 15/15 (93.33%)   |
| ≥60 years (n = 04) | 04             | 04          | 11.50 (10.00/18.00)           | 15.00 (10.00/18.00)       | 4/4 (100%)    | 4/4 (100%)       |

OS: overall survival; Neutrophil and platelet engraftment were expressed as median (lower/upper quartile). aSignificantly different from MM.

Table 4 Influence of disease type on engraftment

| Disease Type | No engraftment | Engraftment | Neutrophil engraftment (day) | Platelet engraftment (day) | OS (<6 months) | OS (6–12 months) |
|--------------|----------------|-------------|-------------------------------|--------------------------|----------------|------------------|
| MM (n = 12)  | 01             | 12          | 11.00 (10.00/11.50)           | 11.00 (10.00/13.00)       | 12/12 (100%)  | 12/12 (100%)     |
| Lymphoma (n = 21) | 01             | 20          | 10.00 (09.00/15.50)           | 10.50 (10.00/16.00)       | 19/21 (90.48%) | 19/21 (90.48%)   |
| Leukemia (n = 16) | 15             | 18.00       | (16.00/20.00)                 | (17.00/25.00)             | 14/16 (87.50%) | 14/16 (87.50%)   |
| SMD (n = 08)  | 08             | 20.50       | (18.50/25.00)                 | (21.00/26.50)             | 08/08 (100%)  | 08/08 (100%)     |
| AA (n = 08)   | 07             | 22.00       | (18.00/29.00)                 | (20.00/29.00)             | 07/08 (87.50%) | 07/08 (87.50%)   |

MM, multiple myeloma; SMD, syndrome myelodysplastic; AA, aplastic anemia; OS, overall survival; Neutrophil and platelet engraftment were expressed as median (lower/upper quartile). aSignificantly different from MM.

Table 5 Influence of transplantation type and graft source on engraftment

| Transplantation Type | Allogeneic BMT (n = 40) | Autologous PBSC (n = 25) |
|----------------------|-------------------------|--------------------------|
| No engraftment       | 02                      | 01                       |
| Engraftment          | 38                      | 24                       |
| Neutrophil engraftment (day) | 19.00 (16.00/23.00) | (09.00/11.00)             |
| Platelet engraftment (day) | 21.00 (18.00/25.00) | (10.00/13.00)             |
| OS (<6 months)       | 34/40 (85.00%)          | 24/25 (96.00%)           |
| OS (6–12 months)     | 31/40 (77.50%)          | 22/25 (88.00%)           |

BMT, bone marrow transplantation; PBSCT, peripheral blood stem cell transplantation; OS, overall survival; Neutrophil and platelet engraftment were expressed as median (lower/upper quartile). aSignificantly different from Allogeneic BMT and Autologous PBSCT.

3) before the transplantation for multiple myeloma in PBSCT. CBV: cyclophosphamide (1,500 mg/m²) given for 4 consecutive days starting 6 days before the transplantation (days minus 6 to minus 3) + carmustine (BCNU—450 mg/m²) given only for 1 day before the transplantation (day minus 6) + etoposide (VP-16—250 mg/m²) given for 3 consecutive days two times a day, starting 6 days before the transplantation (days minus 6 to minus 4) or BEAM: carmustine (BCNU—300 mg/m²) given only for 1 day before the transplantation (day minus 7) + etoposide (VP-16—200 mg/m²) given for 4 consecutive days two times a day, starting 6 days before the transplantation (days minus 6 to minus 3) + cytarabine (100 mg/m²) given for 4 consecutive days two times a day, starting 6 days before the transplantation (days minus 6 to minus 3) + melphalan (140 mg/m²) given only for 1 day before the transplantation (day minus 6)  for Hodgkin and non-Hodgkin lymphomas in PBSCT. FuCy: fludarabine (30 mg/m²) + cyclophosphamide (300 mg/m²), both given for 3 consecutive days, starting 4 days before the transplantation (days minus 4 to minus 2) for Hodgkin and non-Hodgkin lymphomas in BMT. BuCy120: busulfan (1 mg/kg), given for 4 consecutive days, four times a day, starting 8 days before the transplantation (days minus 8 to minus 5) + cyclophosphamide (60 mg/m²), given for 2 consecutive days, starting 4 days before the transplantation (days minus 4 to minus 3) for acute and
chronic myeloid leukemia and syndrome myelodysplastic in BMT. BuCy200: busulfan (1 mg/kg), given for 3 consecutive days three times a day, starting 9 days before the transplantation (days minus 9 to minus 7) + cyclophosphamide (50 mg/m²), given for 4 consecutive days, starting 6 days before the transplantation (days minus 6 to minus 3) or Cy200: cyclophosphamide (40 mg/m²), given for 5 consecutive days, starting 5 days before the transplantation (days minus 5 to minus 1) for aplastic anemia in BMT. CyTBI: cyclophosphamide (60 mg/m²), given for 2 consecutive days, starting 7 days before the transplantation (days minus 7 to minus 6) + total body irradiation, made for 4 consecutive days, starting 4 days before the transplantation (days minus 4 to minus 2, three times on day and day minus 1, two times on day) for acute and chronic lymphoid leukemia in BMT. All patients had 2 rest days (without chemotherapy) before the transplantation (days minus 2 and minus 1), except FluCy and CyTBI CR patients, that had only one rest day (day minus 1 and minus 5, respectively).

**Engraftment.** Neutrophil engraftment was defined as the first of >500/mm³, while platelet engraftment was defined as first of >20,000/mm³.

**Statistical analysis.** Sex, age, disease, graft source and conditioning regimen were examined in terms of their effect on neutrophil and platelet engraftment.

Variables were compared using Kruskal-Wallis ANOVA followed by Mann-Whitney U test. Overall survival was estimated using the Fisher test.

p-value <0.05 was considered statistically significant.

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### Table 6 Influence of conditioning regimen on engraftment

|          | M-200 (n = 12) | CBV (n = 13) | BEAM (n = 05) | FluCy (n = 04) | BuCy120 (n = 19) | BuCy200 (n = 03) | Cy200 (n = 05) | CyTBI (n = 04) |
|----------|----------------|--------------|---------------|---------------|----------------|----------------|---------------|---------------|
| No engraftment | 01            |              |               |               |               |               |               |               |
| 04/04     | 01            |              |               |               |               |               |               |               |
| Neutrophil engraftment (day) | (10.00/11.50) | (10.00/11.50) | (10.00/24.00) | (11.50/16.00) | (18.00/23.00) | (22.00/25.00) | (18.00/29.00) | (17.50/21.00) |
| Platelet engraftment (day)   | (10.00/13.00) | (10.00/13.00) | (11.00/24.00) | (13.50/17.50) | (21.00/25.00) | (24.00/26.00) | (20.00/29.00) | (22.00/29.00) |
| OS (<6 months)              | 12/12 (100%)  | 12/13 (92.30%) | 04/05 (80.00%) | 04/04 (100%)  | 18/19 (94.74%) | 02/03 (66.67%) | 04/05 (80%)   | 03/04 (75.00%) |
| OS (6–12 months)            | 12/12 (100%)  | 10/13 (76.92%) | 04/05 (80.00%) | 04/04 (100%)  | 15/19 (78.95%) | 01/03 (33.33%) | 04/05 (80%)   | 02/04 (50.00%) |

M-200, melphalan 200 mg; CBV, cyclophosphamide + BCNU + etoposide; BEAM, BCNU + etoposide + arabinoside + melphalan; FluCy, fludarabine + cyclophosphamide; BuCy120, busulfan + cyclophosphamide 120 mg; BuCy200, busulfan + cyclophosphamide 200 mg; Cy200, cyclophosphamide 200 mg; CyTBI, cyclophosphamide + total body irradiation. Data were expressed as median (lower/upper quartile).

*Significantly different from M-200. #Significantly different from CBV. *Significantly different from FluCy. §Significantly different from Cy200. *OS significantly different from M-200.

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