evaluation, the patients population was divided in 2 groups: <60 years old (197 pts) and >= 60 years old (61 pts). Patients characteristics are the following: Median age was 51 and 63 years old, disease status at time of transplantation was 59 (30%) and 10 (16%) in CR for both groups, treatment related mortality was 3% for both groups. At 4 years the probability of event-free survival was 33% and 35% (p= NS) and the overall survival was 52% and 44% respectively (p=NS).

These results shows that age >= 60 years is not an adverse prognostic factor for survival and toxicity for patients with multiple myeloma who receive an autologous transplantation.

Treatment of massive plasmacytoma with high-dose melphalan and autologous peripheral blood stem cell transplantation followed by radical radiotherapy

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(Nottingham, UK)

Massive plasmacytomas of the chest wall (>10cm in diameter) are associated with a high incidence of treatment failure following local radiotherapy alone or VAD followed by local radiotherapy. We report 3 patients with massive plasmacytoma who were treated with initial VAD chemotherapy consolidated by high dose melphalan (HDM, 200 mg/m2), autologous PBSCT and radical radiotherapy (30 Gy). Among these patients aged (50-3 years) presented with chest pain. CT of the chest wall revealed a massive plasmacytoma with a maximum diameter of 11, 12 and 13.5cm respectively with associated rib destruction. Bone marrow involvement was demonstrated in one case only with a low-level bone marrow plasma cell infiltrate (7%). Initial treatment with 4 cycles of VAD was used to debulk the tumour followed by PBSCT consolidation with cyclophosphamide (3g/m2) and G-CSF. In one patient the initial response to VAD was poor and this patient proceeded to radiotherapy (30 GY in 10 fractions) prior to HDM. The other 2 patients received HDM following VAD chemotherapy and finally radiotherapy (30-40Gy) to the residual mass. All patients responded and 2 remain in complete remission at 2.5 and 3.5 years although 1 patient has a large residual calcified mass. The remaining patient achieved CR but relapsed at 8 months as a generalised immunoblastic lymphoma. As previous studies have shown a high risk of treatment failure in massive plasmacytomas this approach offers the potential for long-term disease free survival.

Intensive sequential therapy comprehensive of autologous stem cell transplant in multiple myeloma: a single center experience

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Between January 1994 and April 2001, 26 patients with a median age of 51 years and untreated multiple myeloma were enrolled in an intensive sequential therapy consisting of 2 courses of VMD regimen (VCR, mitoxantrone and dexamethasone for 4 days) followed of three consecutive non-cross-resistant regimens: a) cyclophosphamide (CY) 7 g/mq followed by G-CSF 5 mcg/Kg per day and leukapheresis upon recovery from white blood cell nadir; b) EDHAP with G-CSF and beam protocol with G-CSF was used as the third regimen. Peripheral blood stem cells were collected after each regimen. The high dose therapy with Melphalan 140 mg/mq +/- fractioned TBI 1200 cGy followed by infusion of PBSCT collections was subsequently offered to our pts. In few patients, who previously received radiation therapy the preparative regimen was Busulfan 16 mg/Kg and melphalan 60 mg/Kg. The selection criteria included the presence of symptoms, age <= 60 years, adequate cardiopulmonary function, no HLA sibling donor. Renal dysfunction or evidence of amyloidosis at diagnosis were not a contraindication. Twenty pts had stage III and 6 pts stage II. The median CD34+ cells collected was 15x106 /Kg (range 2.2-46). Five pts achieved complete remission and the others pts achieved partial remission before transplant. The median CD34+ cell dose infused at the time of autografting was 6.5x106/Kg (range 2.2 -21.2 ). The neutrophil (>500 micron/L) and the platelets recovery (>20.000 micron/L) occurred a median of 12 days and 16 days after ASCT, respectively. The interval between of induction therapy and ASCT was 5.5 months. No treatment-related deaths were observed. At a median of 3 months, 14 pts received IFN therapy as maintenance. To date,15 pts are alive, of whom 7 in complete remission. Six relapsed pts underwent a second autografting. At a median of 4.5 years, the overall survival is 57%.

Thalidomide overcomes VAD chemoresistance and allows PBSCT collection in 3 multiple myeloma patients

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Objective: Thalidomide (Thal) showed a 30-50% of partial responses when it was used as salvage treatment in several series of resistant or relapsing multiple myeloma (MM) patients, who had been heavily pretreated in the majority of the cases. We investigated the activity of Thal in 3 newly diagnosed MM patients unresponsive to VAD induction therapy and included in a program of mobilization of PBSCT and high-dose treatment. Methods: Three previously untreated stage III A MM patients of respectively 50, 59 and 63 years old , who showed a resistant or progressive disease after 2 VAD cycles, started on 2 further VAD in association with 200 mg Thal, then were treated with Thal alone at escalating doses to the maximum tolerated dose until they showed the greatest response. At this time PBSCT were collected and an ASCT was sheduled. Results: The maximum tolerated dose was 100 mg for patient n.1 and 400 mg for patient n.2 and n.3. Side-effects were WHO grade III peripheral neurotoxicity for patient n.1 and WHO grade II slippus for the other 2 cases. Thal was administered for a median of 120 days (116-139) before PBSCT collection. CD34+ collected were respectively 4, 8.3 and 13.5 x 106/Kg. Response to Thal is presented in the following table.

| Patient | CD34+ | PC% | CD34+ | PC% | CD34+ | PC% |
|---------|-------|-----|-------|-----|-------|-----|
| n.1     | 10    | 90  | 40    | 90  | 20    | 90  |
| n.2     | 40    | 90  | 10    | 90  | 10    | 90  |
| n.3     | 31    | 90  | 50    | 90  | 22    | 90  |

Conclusions: Thal obtained a reduction > 50% of M-component (MC) and plasma cell (PC) marrow infiltration in 3 newly diagnosed MM patients who had a primary resistance to VAD and allowed to mobilize a number of CD34+ PBSCT adequate for an autotransplantation.

5. Autoimmune Disease

P471

Total body irradiation (TBI) and high-dose chemotherapy combined with T-cell depleted marrow grafts in multiple sclerosis (MS)

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Ablation of the immune system followed by autologous stem cell rescue is currently explored as therapy for patients with multiple sclerosis (MS). We used rigorously depletion of T-cells in the recipient by employing antithymocyte globulin (ATG) and TBI as
the preparative components, as well as using marrow grafts (rather than using peripheral blood) and CD34 selection. Patients with lastly relapsing progressive form of MS and with a score between 5 and 7 on the expanded disability scale (EDSS) were included. We report on 8 successive patients who were followed for 9-32 months post transplant (median: 18.5 mo). Time to neutrophil recovery varied from 12 to 38 days (median: 27 days). Platelet recovery was observed between 13 to 31 days (median: 25 days). Significant toxicity (WHO grade 2 or more) during the first three months after stem cell transplantation (SCT) included mucositis (gr 2, n=6), hepatic dysfunction (gr 2, n=1, gr 3, n=3), infections like central venous catheter infections, herpes simplex lesions and one clostridium difficile colitis (gr 2, n=2; gr 3, n=3; gr 4, n=1). Skin toxicity (exanthema gr 2, n=4; gr 3, n=1), and neurologic problems like spasms (gr 2, n=3; gr 3, n=1) have also been observed. In 5 of 8 patients who were all IgG-Epstein-Barr virus (EBV) positive prior to transplant, EBV reactivation was quantitatively monitored in the patients plasma samples using a validated real-time PCR test at weekly intervals. After transplantation, all 5 patients showed at least one, sometimes repeated EBV-PCR reactivations. In 3 patients, EBV-DNA was detected at high levels (peak values: 6,250 - 240,000 copies/mL). In patients with EBV reactivation and plasma EBV-DNA levels exceeding 1,000 copies/mL, no signs of EBV-related lymphoproliferative disease (EBV-LPD) were found.

Effects of this form of therapy were evaluated by EDSS and follow-up MRI scans of the cerebrum. Evaluation of the EDSS scores at current follow-up indicates progression of disability in 4, stabilisation in 2, and improvement in 2 patients. MRI scans showed a significant decrease in gadolinium positive lesions following the SCT procedure in all patients.

Thus, autologous SCT with rigorous T-cell depletition might be beneficial for MS patients with acceptable toxicity. EBV reactivation should be monitored carefully in this setting.

**P472**

**Autologous stem cell transplantation for refractory myasthenia gravis**

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Objectives: Severe refractory autoimmune disease can be treated with high dose immunosuppression followed by autologous blood stem cell transplantation (ASCT). We report on a 50 years old female patient who has been suffering from myasthenia gravis (MG) since January 1992. Despite a thymectomy in 1993 her clinical syndrome persisted. Following the year before HDIT the patient showed a worsening of the disease despite the high-dose immunoglobulins. HSC mobilization required 2 apheresis procedures to collect and store >5x10^6 CD34+ cells/kg. The conditioning regimen has been well tolerated and caused only g.i. side effects < grade II. No life-threatening, relapse related toxicity occurred. Mobility improved slightly in the post-transplant period. Engraftment was rapid and the ANC was <500/µl for 9 days, platelet counts < 20000/µl for 7 days. With follow-up of 24 months there has been no evidence of disease progression. The patient experienced a sustained clinical improvement with a decrease of motor disability. Motor nerve conduction studies measured 24 months after treatment showed improvement. Conclusions: With follow-up of 24 months, clinically important improvement was observed in one patient with CIDP after HDIT using cyclophosphamide with autologous hematopoietic stem cell rescue. HDIT should be considered in CIDP patients with progressive disease under conventional treatments.

**P473**

**Treatment of chronic inflammatory demyelinating polyradiculoneuropathy (CIDP) with high-dose immunosuppressive therapy (HDIT) using cyclophosphamide with autologous hematopoietic stem cell rescue**

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Background: Chronic inflammatory demyelinating polyradiculoneuropathy (CIDP) is considered to be an autoimmune disorder. Current treatment regimens leave 4% to 30% of patients with CIDP with moderate or severe disability. Conventional dose immunosuppressive therapy has produced very limited success in halting disease progression in treatment-resistant patients. It has been proposed that high-dose immunosuppressive therapy using cyclophosphamide with autologous hematopoietic stem cell rescue may be an effective treatment for severe autoimmune diseases.

Methods: We have designed a study to test in patients with severe autoimmune disease the toxicity and efficacy of a myeloablative regimen of cyclophosphamide 50 mg/kg x 4. Autologous hematopoietic stem cells were mobilized with cyclophosphamide 4 g/m2 and G-CSF 5 µg/kg. A total of >5x10^6 CD34+ cells/kg were stored before proceeding to conditioning therapy.

Case report: The 36-year-old male patient suffered from classic progressive CIDP starting on 1988 with progressive weakness of the upper and lower limbs. Previous treatment included corticosteroids, intravenous immunoglobulins and interferon-beta 1a with the partial control of the progression of the disease. During the year before HDIT the patient showed a worsening of the disease despite the high-dose immunoglobulins. HSC mobilization required 2 apheresis procedures to collect and store >5x10^6 CD34+ cells/kg. The conditioning regimen has been well tolerated and caused only g.i. side effects < grade II. No life-threatening, relapse related toxicity occurred. Mobility improved slightly in the post-transplant period. Engraftment was rapid and the ANC was <500/µl for 9 days, platelet counts < 20000/µl for 7 days. With follow-up of 24 months there has been no evidence of disease progression. The patient experienced a sustained clinical improvement with a decrease of motor disability. Motor nerve conduction studies measured 24 months after treatment showed improvement. Conclusions: With follow-up of 24 months, clinically important improvement was observed in one patient with CIDP after HDIT using cyclophosphamide with autologous hematopoietic stem cell rescue. HDIT should be considered in CIDP patients with progressive disease under conventional treatments.
matching donor was performed in June 2001, after conditioning with fludarabine 180 mg/m², cyclophosphamide 60 mg/kg, ATG 40 mg/kg. Neutrophil recovery was seen two weeks after transplantation, she had acute GVHD grade 1, and she was treated with the standard treatment of CMV after two months. Due to the persistence of thrombocytopenia, anemia, and mixed chimerism she was given one donor lymphocyte infusion after four months and her T, B and myeloid cells are 100% donor-derived one month later. She is in good condition five months post-transplant and does not need transfusions of RBC or platelets, although the platelet count is still subnormal. The cardiolipin antibody has disappeared and the bleeding time is normal.

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Successful allogeneic bone marrow transplant (BMT) in patient with AlpS-like disease
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The deceased fucntion of Fas or mutations of Fas gene leads to a clinical picture characterized by non-malignant lymphoproliferation with autoimmunity manifestations. Two successful allogeneic BMT have been reported in patients affected by Fas deficit so far. We describe the third case cured from ALPS-like-pattern by allogeneic BMT. The 12 years old patient was referred to our Center with severe thrombocytopenia, marked respiratory insufficiency due to pulmonary fibrosis and severe side effects of prolonged corticosteroid therapy. She was splenectomized 2 years ago for massive splenomegaly. The firstborn brother presented diffuse lymphadenopathy, hepatosplenomegaly and pancytopenia at 6 years of age, previously expired at 13 years of age with severe thrombocytopenia and autoimmune disorders. Analysis of Fas function was normal in the mother, slightly decreased in the father and strongly decreased in the sister. Expansion of double-negative T cells was absent. Sequencing of the Fas gene did not detect any mutation. The family history and the Fas function analysis suggested a genetically mediated autoimmune disease involving the Fas function. As the only HLA identical sister was suspected to be affected with the same disorder, the HLA phenotypically identical mother (matched for HLA-A,-B,-C, DRB1-B4, DQA1-B1 loci and mismatched for 1 allele on locus DPB1) was selected as the bone marrow donor. The patient was prepared to BMT with BU 14 + CY 200, CsA and sMTX were given as GVHD prophylaxis. Allogeneic engraftment was documented on day +20. She developed grade II skin aGVHD, resolved with steroid therapy. AT = ALPS-like pattern by allogeneic BMT the clinical conditions were good with 100% of donor engraftment, withouth signs of cGVHD and with normal blood count. The immunosuppressive therapy was completely stopped at 1 year after BMT. Fourteen months after BMT she is in good clinical conditions and cured of her genetically inherited disease. In conclusion this case provides confirmation that the only radical treatment for patients with ALPS or ALPS-like diseases is represented by allogeneic BMT.

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Improvement of inflammatory bowel disease after allogeneic stem cell transplantation
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After allogeneic marrow transplantation remissions of several autoimmune diseases were described. We retrospectively analysed the course of coexistent inflammatory bowel disease (IBD) in 6 patients who underwent allogeneic stem cell transplantation for acute (n=1) or chronic (n=5) myeloid leukemia. Between July 1994 and November 2000, four patients with Crohn’s disease and two patients with idiopathic ulcerative colitis and AML or CML were transplanted with bone marrow (n=4) or peripheral blood stem cells (n=2) from a HLA-identical (n=4) and HLA-partial identical (n=1) sibling donor or unrelated HLA-matched donor (n=1). Before transplant, three patients received an immunosuppressive medication for the IBD. Two of them received low doses of steroid and 5-ASA, another patient received 5-ASA alone. The patients was found to have an active IBD at transplantation. After a mean follow up period of 57 months after transplantation all patients are alive, in complete remission of CML or AML and without recurrence of IBD. Three patients suffered from temporary cytogenetic relapse of CML but did not develop any signs of intestinal inflammation again. One patient only revealed clinical signs of his idiopathic ulcerative colitis 11 relapses after transplantation. Moreover, no persisting morbidity of the gut did not prove a relapse of ulcerative colitis and diarrhea diminished under supportive therapy. This patient remained free of any signs of IBD up to date with a mean follow up of 39 after bone marrow transplantation. Five of six patients remained free of IBD over a follow up period of 107 months (mean 57 months). These observations may imply that an underlying host immune dysregulation might play a central role in the perpetuation of IBD. It may be corrected by allogeneic hematopoietic stem cell transplantation.

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P-ANCA associated vasculitis (M. Wegener): successful HLA-identical BMT
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P-ANCA associated vasculitis is a rare but fulminant disease in paediatric patients. The introduction of immunosuppressive treatment with e.g. cyclophosphamide and prednisolone has dramatically increased the remission rate. However, the rate of relapses after transplantation and disease related mortality is still high. We describe an 8 year old girl with a generalised form of Wegener's Granulomatosis with eye, renal, heart and lung involvement requiring mechanical ventilation due to pulmonary haemorrhage and ARDS. Other clinical manifestations were thrombocytopenia and increasing hepatosplenomegaly with intrahepatic cholestasis. The treatment consisted of prednisolone, cyclophosphamide and one course of methotrexate. After first signs of remission, disease progression and toxicity related problems BMT from the HLA-identical brother was considered. The girl underwent allogeneic BMT 19 months after diagnosis. To reduce toxicity, the conditioning regimen consisted of fludarabine, 2 Gy TBI and 2.5 Gy TLI. GvHD and rejection prophylaxis consisted of CsA and MMF, and 3.6 x10⁹ CD34 cells and 6.4 x10⁶ CD3 cells/kg of the recipient were transfused. G-CSF was given from day +20 till day +30 and myeloid engraftment with ANC>0.5 G/L was achieved on day +78. Chimerism analysis were performed on FACs-sorted cells by FISH and showed mixed engraftment with only mild restriction of life quality. Based on this experience, we conclude that allogeneic BMT with reduced intensity conditioning regimen may be a curative treatment option for refractory autoimmune disease.
Autologous stem cell transplantation (ASCT) in four children with refractory JCA and SLE

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ASCT has been proposed as a new therapeutic option for patients with severe autoimmune disease refractory to conventional treatment. Here, we report three children with a severe form of systemic JCA and one patient with severe systemic lupus erythematoses treated with ASCT in a phase I study. Patients: Three patients (age: 5, 9, 14 yrs) who developed severe systemic JCA with high spiking fever, rashes, hepatomegaly, polyarthritism, morning stiffness, ESR > 100 mm/h, CRP > 100 mg/l were refractory to NSAIDs, steroids, MTX, IVIG, CsA and cyclophosphamide. This patient acquired on day + 45 EBV infection with LPD which was treated successfully with ganciclovir, cidofovir and rituximab. Stem cell harvest: After a priming dose of cyclophosphamide (2 g/m²) and mobilization with G-CSF (10 μg/kg/day) peripheral blood stem cells were collected using a of a Cobe separator. Using a CliniMACS device, CD34-positive selection was performed yielding a final CD34+ cell amount of 4.2 – 6.5 x 10^6/kg contaminated with zero to 3.2 x 10^4/kg CD34+ lymphocytes, respectively. Stem cells were stored in liquid nitrogen. Conditioning regimen: Fludarabine (30 mg/m²): days –7 and –6; cyclophosphamide (50 mg/kg): days –5 to –2; ATG (5 -10 mg/kg): days –6 to –2; methylprednisolone (1g/m²): days –4 to –2. On day 0, the frozen CD34+ cells were thawed and infused. Results: Rapid engraftment of neutrophils > 1.0 GPT/l: days +10 to +13; platelets > 20 GPT/l: days +6 to +14; lymphocytes > 1.0 GPT/l: days +46 to + 66, respectively. Patients were discharged from hospital prior to day +24 to + 53, respectively, and remained free from active JCA and SLE with no immunosuppressive medication for 9, 10, 20 and 20 months, respectively.

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Autologous stem cell transplantation for refractory autoimmune cytopenias

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Autologous Stem Cell Transplantation (ASCT) has been explored as a treatment option in severe uncontrolled autoimmune disease. We report on outcome in 17 patients with hematologic autoimmune cytopenias reported to the autoimmune disease working party of the EBMT by 13 centers. Patients had idiopathic thrombocytopenic purpura (10), pure red cell aplasia (4), autoimmune hemolytic anemia (2), and Evan’s syndrome (1). Median age was 31 (4-45) years, 8 were female. Median disease duration prior to transplant was 9 (3-18) months. Stem cells were from blood (14) or from marrow (2). Peripheral stem cells were mobilized using either growth factors alone (7) or G-CSF in combination with cyclophosphamide (6). Pretransplant conditioning regimens included cyclophosphamide alone (n=3), cyclophosphamide with other drugs or ATG, (n=9), Melphalan (2), or were Fludarabine based (2). Stem cells were either unpurged (n=4) or purged of immune cells using varying methods of depletion (n=12). Median follow-up of surviving patients is 30 (5-53) months. Three patients died within 100 days posttransplant, 2 of hemorrhage and infectious complications, and 1 with progressive hemolysis. Nine patients showed a response to treatment, 5 complete remissions (3 ITP, 1 Evans, 1 PRCA) sustained in 4 patients. Of note, one of these complete remissions occurred after mobilization only. ASCT may induce a sustained remission in a fraction of patients with severe autoimmune cytopenias of long duration.

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Total alopecia cured by allogeneic PBSCT. Proof of principle of autoimmune pathogenesis?

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A 40 year old male patient with CML in first chronic phase, diagnosed 8 months previously suffered from total alopecia (alopecia areata generalis) that had occurred suddenly 11 years and 2 months earlier. The alopecia involved all body hair including eyebrows and eyelashes. To treat the CML he underwent conditioning with 120 mg/kg of cyclophosphamide and 12 Gy of fractionated TBI and received an allogeneic peripheral stem cell transplant from a HLA-identical brother, 7 years older. Graft-versus-host disease prophylaxis was with cyclosporine and short course methotrexate. Hemopoietic reconstitution was uneventful. On day 40 hair started to grow on his upper lip, and chin. Eyebrows and eyelashes grew back and on day 55 hair started to grow on the top of his head. He is currently (d+90) in remission of the CML, without manifestations of graft-versus-host disease.

The course of events is best explained by alopecia areata generalis being an autoimmune disorder cured by a graft versus autoinmunity effect generated by replacing the immunohemopoetic system through allogeneic hemopoetic stem cell transplantation.

Additional abstracts to this topic

The dynamic of immunological reconstitution in multiple sclerosis patients after autologous peripheral stem cells transplantation

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Introduction: Peripheral stem cells transplantation (PSCT) is a new perspective method in the treatment of severe autoimmune diseases. PSCT allows to achieve a stabilization in 85% patients during 5 or more years.

Goals: The determination of the peculiarities of immunological reconstitution in MS patients after high dose immunosuppressive therapy (HDIT) with autologous peripheral stem cells support.

Materials & methods: The immune monitoring (IM) was carried out in 5 MS patients after HDIT with autologous stem cells support. In four cases the PSCT were made according to EBMT protocol. One patient underwent nonmyeloablative HDIT with CSF support (Granocyte). There were four females and one male, median age was 38/–14 y.o. The EDSS scale varied from 2,0 to 8.

The following parameters: CD3, CD4, CD8, CD4/CD8, CD16, CD56 were investigated by flow cytometry with “Becton Dickinson” FacsCan. The measurements were made before PSCT; on D+30, D+100, D+180, D+360.

Results: Clinical improvement in two of five patients result in decrease of EDSS level by 0,5 and 1,5 respectively. In three other patients the stabilization of MS was observed. In patients with positive clinical results the significant decrease of initially increased autoimmune activity markers (CD4) was found out with future slight increase and stabilization on normal level. Relative increase of protective subpopulations (CD8, CD16) was determined as well. Such immunological profile was preserved during all the period of MS remission (follow up period varied from 3 months to 2 years).

Conclusions: Along with clinical improvement immunological changes pointing to decrease of autoimmune processes activity in
MS patients after HDIT with peripheral stem cells support are observed.

6. Donor Issues

P481

Mobilization and harvest of peripheral blood stem cells from 128 healthy donors. A single center experience

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Background: Between January 1995 and August 2001, mobilisation and harvest of peripheral blood stem cells for allogeneic transplantation was performed in 92 related and 36 unrelated healthy donors. Four donors were harvested on two separate occasions after renewed G-CSF mobilisation.

Method: Retrospective analysis of 128 medical reports.

Results: Median age was 40 years (range 14-71), median weight was 75 kg (range 50-130) and the male/female ratio was 1.5 (76/52). In a majority of donors (96%, 123/128) antecubital veins were used as access to the circulation. In 4% of donors antecubital veins were unsuitable, and a femoral catheter was therefore employed.

In each apheresis procedure, two blood volumes (Cobe Spectra: median 10.1 L, range 6.8-14.6 L) or 10.0L of whole blood (CS3000) were processed. Median 2 (range 1-3) apheresis procedures were performed/donation. During G-CSF mobilisation, 79% (101/128) donors reported mild to moderate side effects. After G-CSF mobilisation, median WBC was 42.9 x 10⁹/L (range 17.8-78.3 x 10⁹/L). During apheresis, 59% (76/128) of the donors reported mild side effects such as typical citrate mediated circumoral paresthesias and 16% (21/128) experienced other types of moderate symptoms. After the first apheresis 9% (9/99) of the donors had a serum potassium level below 3.0 mmol/L (min 2.7 mmol/L).

During 1999 there was a change of cell separator from Baxter CS3000 to Gambro CobeSpectra (AutoPBSC program) with the intent to minimize platelet loss and to gain better control of the citrate administration. Thrombocytopenia (defined as platelet count <100 x 10⁹/L) occurred in 44% (17/39) of the donors harvested with CS3000 and in 5% (3/66) of the donors harvested with CobeSpectra. Moderate citrate related side effects was reported in 49% (29/59) of the donors harvested with CS3000 and in 66% (45/68) of the donors harvested with CobeSpectra. There was no difference between the two cell separators regarding yield of CD34⁺ cells: median 5.48 and 5.59 x 10⁶/kg body weight of the donor for CS3000 and CobeSpectra respectively.

Conclusion: No serious side effects were reported during G-CSF mobilisation or apheresis. A lower frequency of thrombocytopenia (p=0.001) was seen after apheresis with Cobe Spectra AutoPBSC than with CS3000 with no difference regarding yield of CD34⁺ cells.

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Peripheral blood stem cells (Pbsc) mobilization and collection in healthy donors receiving granulocyte colony-stimulating factor (G-CSf) - A single-center experience about safety aspects

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From May 1995 to November 2001, a total of 48 healthy peripheral blood stem cell donors (5 of them underwent 2 mobilizations), 27 male and 21 female, median age 46 years (16-63), were mobilized by a mean dose of G-CSF 9.7+2.2 mg/kg/day. A prophylaxis with paracetamol and hirudin low-dose was administered to prevent side effects from G-CSF. A total of 133 aphereses were performed with Fresenius cell separator using the AS 104 or AS 204 systems. No donor needed CVC. A median of 2 procedures (range 1-4) and of 13.5 L (range 9-18) were performed. WBC increased to a maximum of 80.0 x10⁹/L (mean 47.8+11.6). After the aphereses, platelet count decreased to a minimum of 42.0 x10⁹/L (mean 104.4+40.6) and returning to baseline values at median day 6 (range 2-35) from the last procedure. Reinfusion of autologous platelet-rich plasma was necessary in 23/133 procedures (17.2%) because of donor platelet-count <80 x 10⁹/L. Mean number of MNC and CD34⁺ cells harvested were 939.3+344.6x10⁶/kg and 12.5+5.7x10⁶/kg, respectively. The target dose of 5.0 x10⁶/kg CD34⁺ cells was obtained in 58.5% of cases after a single aphereses. Target CD34⁺ cells was reached in all cases. The most frequent G-CSF related side-effect was bone pain (8/53 WHO grade 1, 10/53 WHO grade 2 and 1/53 WHO grade 3). Paraesthesia occurred in 36/133 (27%) aphereses (32 mild, 4 severe). 3 donors developed headache, 1 lipothyma and 2 a mild splenomegaly (>2cm).1 donor showed a mild increase in ALT levels. After a median follow-up of 19.7 months (1-73.6), no donor developed long-term adverse effects. In conclusion, G-CSF mobilization and harvesting of allogeneic PBPC was feasible and well tolerated in all donors without major short-term or long-term adverse effects.

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Safety issues of hematopoietic progenitor cell (HPC) collection from healthy donors: transient thrombocytopenia in older age and multiple aphereses

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We retropectedly analyzed possible correlations between CD34⁺ cell count in HPC collections from healthy individuals with the following variables: age, sex, body weight, medical history, bone marrow (BM) cellularity, medications, dose/duration of rhG-CSF administration, number of aphereses, peripheral blood (PB) CD34⁺ cell numbers and WBC count. We also present complications and side effects of rhG-CSF administration and apheresis in this cohort. Included in the analysis were 65 donors with a median age of 29 years (10-76); there were 57 PB and 8 BM harvests. Donors were subjected to detailed laboratory examinations, including: complete blood count, serum biochemistry, tests for hepatitis B and C viruses, HIV and herpesviruses, electrocardiogram, chest x-ray and assessment of BM smear. rhG-CSF was administered at a median dose of 10 mg/kg/day for a median of 6 days (2-12). The median number of aphereses was 3 (2-6). The median CD34⁺ cell count was 5.32 x 10⁶/kg (0.18-18.1); significantly (p=0.04) more CD34⁺ cells were collected with increasing number of aphereses. CD34⁺ cell mobilization pattern was the same, regardless of age; however, in older donors more aphereses were required for adequate harvests. On multivariate analysis, the number of aphereses was the only variable significantly associated with CD34⁺ cell count (p=0.018). Generally, the collection procedure was well tolerated, the main complications being reversible thrombocytopenia (41.5%), hypocalcemia (12.3%), bone pain (1.5%), decrease in hemoglobin level (26.1%), liver aminotransferase elevation (7.6%) and headache (3.0%). On univariate analysis, thrombocytopenia was associated with increasing number of aphereses (p=0.03), while on multivariate analysis with more advanced age (p=0.012). With a median follow-up of 33 months (9-59), there were no late side effects attributable to rhG-CSF. Nevertheless, firm conclusions about potential long-term toxicities of this method can not be drawn at this moment.