

# Peripheral Blood Stem Cells: Transplantation and Beyond

*L.B. To*

## **Blood Stem Cell Autotransplantation**

Recent data from the International Bone Marrow Transplant Registry (IBMTR) and the European Blood and Marrow Transplant Group (EBMT) show that the number of autologous transplants since 1990 has exceeded that of allogeneic transplants. According to EBMT data, the number of autologous transplants to treat lymphoma and breast cancer in Europe has increased fivefold between 1990 and 1994, with the percentage of autologous transplants using blood cells rising from 15% to 75%. Although the use of blood cells for transplant began in the mid-1980s (with the exception of sporadic case reports), it is now the predominant source of cells for hemopoietic rescue.

Early murine hemopoietic transplantation studies were mostly performed with bone marrow (BM) rather than blood cells for two reasons: there are inherent methodological limitations of adequate and repeated samplings of blood cells in mice, and Micklem et al<sup>(1)</sup> showed that steady-state blood cells were inferior to bone marrow as a source of marrow repopulating cells. The failure of engraftment in early case reports of steady-state blood cell transplants in humans<sup>(2,3)</sup> also reinforced the emphasis on BM as the source of hemopoietic rescue. These data overshadowed the more encouraging reports in larger animal models.<sup>(4)</sup>

Preference for BM over blood cells was reinforced by the clinical results of autologous transplantation using steady-state blood cells that showed that their hemopoietic reconstitutive capacity is no better than BM.<sup>(5)</sup> Hence, the use of mobilised blood cells for transplantation was initially viewed with a healthy skepticism. It was not until successive reports of clinical studies had demonstrated the phenomenon of blood cell mobilisation and the hemopoietic reconstitution advantage of mobilised blood cells<sup>(6-11)</sup> that systematic experimental studies commenced. Since then, these studies have demonstrated beyond doubt the equivalence if not superiority of mobilised blood cells to BM for hemopoietic reconstitution.

## **Blood Cell Mobilisation**

Progenitor cells are present in low quantities in blood during steady state hemopoiesis postnatally with the exception of myeloproliferative states.

### ***Myelosuppressive Chemotherapy***

Myelosuppressive chemotherapy was the first clinically useful blood cell mobilization protocol. During the recovery phase following myelosuppressive chemotherapy, 50-fold or greater increases in PB CFU-GM occur. High-dose cyclophosphamide is the most commonly reported protocol<sup>(8,11,12)</sup> because it is active against most tumours and can be given justifiably even in diseases when conventional treatment is not sufficiently myelosuppressive.

The main limitations of chemotherapy mobilisation are neutropenic sepsis, bleeding diathesis and the unpredictability of the timing of apheresis.<sup>(13)</sup> With the advent of hemopoietic growth factors, it is no longer necessary to use myelosuppressive chemotherapy alone for mobilisation.

### ***Myelosuppressive Chemotherapy Plus Hemopoietic Growth Factors***

The largest series studying the effect of G-CSF in enhancing chemotherapy mobilisation was reported by Schwartzberg.<sup>(14)</sup> Both dose escalation and addition of G-CSF were associated with higher CD34+ cell yield. In an earlier study the same authors reported a shortening of neutropenia and a reduction of hospitalisation. The dose of G-CSF used with chemotherapy is in the range of 3-6 mg/kg/day. It has become an increasingly common practise to use a 300 mg ampoule of G-CSF per day instead of adhering to a strict per kg formula.<sup>(15)</sup> A recent report described patients who received G-CSF from day +5<sup>(16)</sup> with still adequate CD34+ cell yield. Hence, a delayed start of G-CSF post mobilisation may reduce cost without loss of efficacy.

GM-CSF, IL-3 and PIXY321 have also been used to enhance mobilisation with chemotherapy. Gianni et al has shown that starting GM-CSF five days after chemotherapy was just as effective as starting on day 1, but starting on 7 or 10 days may be less effective.<sup>(12)</sup>

Many different myelosuppressive chemotherapy protocols have been used in conjunction with cytokines for mobilisation. The results of the protocols do not differ significantly; therefore, the use of blood cell mobilisation as part of therapy is more important than which protocol is used. An additional impression is that adequate mobilisation may occur even with chemotherapy regimen that are only mildly myelotoxic, such as cyclophosphamide 1-2 gm/m<sup>2</sup>.

### ***Hemopoietic Growth Factors Alone***

G-CSF has been shown to increase the level of PB progenitor cells in cancer patients.<sup>(17)</sup> Dose escalation beyond 10-16 mg/kg/day does not appear to further enhance mobilisation. In human G-CSF-mobilised PB CD34+ cells, subsets of CD38-, HLA-DR- and CD33- cells can be readily identified that are capable of generating CFU-GM in liquid culture for three weeks or more.<sup>(18)</sup>

Sheridan et al reported on 14 patients with poor prognosis non-myeloid malignancy who were transplanted with both the cryopreserved G-CSF mobilised blood cells and autologous BM cryopreserved prior to mobilisation.<sup>(19)</sup> They all received G-CSF post transplant. Compared to two historical groups transplanted with autologous BM with and without posttransplant G-CSF, there was a more rapid platelet reconstitution with a resultant reduction in platelet transfusion requirement. Reports from many centres have confirmed that G-CSF mobilised blood cells produced more rapid neutrophil and platelet reconstitution.

Basser et al<sup>(20)</sup> described G-CSF mobilisation in patients who had not received any prior chemotherapy. Three leukaphereses yielded  $115 \times 10^4$  CFU-GM/kg BW, range 23-274  $\times 10^4$ /kg BW. Hence, the progenitor yield appeared to be higher in patients whose BM has not been damaged by chemotherapy.

The mobilisation effect of SCF on humans when used alone has not been published. The use of SCF/G-CSF for mobilisation followed the demonstration of synergy of such a combination in experimental animals. In patients with breast cancer, concomitant SCF enhances blood cell mobilisation of G-CSF.<sup>(21)</sup>

Socinski et al described an 18-fold increase in PB CFU-GM after GM-CSF (Lancet 1988).<sup>(22)</sup> A later report described a more modest 4- to 9-fold increase in PB CFU-GM. Peters et al<sup>(23)</sup> also suggested that GM-CSF is less efficacious than G-CSF in mobilisation. IL3 has little activity in mobilisation.<sup>(24)</sup> Vadhan-Raj et al<sup>(25)</sup> reported only modest mobilisation activity of PIXY321 in 24 patients with sarcoma prior to chemotherapy.

Brasel et al<sup>(26)</sup> reported preliminary data from a murine study comparing the progenitor cell mobilisation effect of Flt3 ligand. Flt3 ligand produced an 83-fold increase of PB CFU-GM, but Flt3 ligand + G-CSF gave a 2193 fold increase. Synergism with GM-CSF was minimal.

Erythropoietin, macrophage inflammatory protein-1a and IL-6 have been shown to have very modest activity in progenitor cell mobilisation.<sup>(27-29)</sup> IL-1 mobilises CFU-GM and CFU-S12 after 4-8 hours and these PB cells have long-term hemopoietic reconstitutive capacity.<sup>(30)</sup> The same group also demonstrated similar activity with IL-8.<sup>(31)</sup>

The mobilisation potential of thrombopoietin has not been reported.

### **Factors Affecting Yield**

In progenitor cell mobilisation following myelosuppressive chemotherapy, the dose of chemotherapy, the severity of myelosuppression and the rate of recovery of leukocyte count all correlate positively with progenitor cell yield following chemotherapy.<sup>(11,13,14)</sup>

The addition of G-CSF or GM-CSF to myelosuppressive chemotherapy appears to enhance progenitor cell yield.<sup>(14)</sup> The amount of previous chemoradiotherapy and the degree of bone marrow involvement are significant determinants of progenitor cell yield.<sup>(15,32)</sup> Experimental data suggest that chemoradiotherapy damages stem cells.

The importance of hemopoietic reserve in progenitor cell mobilisation underscores how critical it is to incorporate mobilisation as part of planned treatment rather than as part of salvage for resistant disease.<sup>(32)</sup>

### **Timing of Collection**

In chemotherapy, mobilisation aphereses are usually performed when the leukocyte count starts to rise above  $1 \times 10^9/L$ .<sup>(11)</sup> In G-CSF, mobilisation aphereses are usually performed on days 5, 6 and 7 of G-CSF administration.<sup>(19)</sup> In chemotherapy + G-CSF/GM-CSF mobilisation, most groups start apheresis when the leukocyte count is  $2-5 \times 10^9/L$  and some groups recommend not starting until the leukocyte count is greater than  $10 \times 10^9/L$ .<sup>(15)</sup> Nowadays CD34+ cell levels should be used because it is a more direct measure of progenitor cells. Most groups use  $20-40 \times 10^6/L$  as the minimum starting level. The yield should also be defined as CD34+ cells.

There has been considerable interest in collecting sufficient progenitor cells for rescue from a single apheresis. Several groups are now using large volume apheresis to maximise progenitor cell yield,<sup>(33)</sup> although the patient's tolerance can be a problem.

### **Target and Thresholds**

A target progenitor cell yield from leukapheresis is important because the number of progenitor cells re-infused correlates with the rate of hemopoietic reconstitution, which influences the safety and costs of transplants. The target yield then determines when and how many leukaphereses will be performed. A highly significant correlation between CFU-GM and CD34+ cells has been confirmed by numerous reports so data based on either assay are valid.

A CFU-GM cell dose effect was first suggested in 1986<sup>(9)</sup> when analysis of the limited data available then suggested that a minimum of  $30\text{-}50 \times 10^4/\text{kg BW}$  is required for rapid hemopoietic reconstitution. In G-CSF mobilised blood cell transplants, Sheridan et al reported that patients receiving  $> 30 \times 10^4$  CFU-GM/kg BW recovered faster than those receiving less.<sup>(34)</sup> With more data now available,  $15\text{-}20 \times 10^4$  CFU-GM/kg or  $1\text{-}2 \times 10^6$  CD34+ cells/kg is generally agreed as the minimum threshold below which rapid hemopoietic reconstitution may not occur.<sup>(10,35)</sup>

A progenitor cell dose above the minimum threshold is generally associated with increasingly rapid hemopoietic reconstitution.<sup>(10)</sup> However there seems to be an upper threshold effect at  $50 \times 10^4$  CFU-GM/kg or  $5\text{-}8 \times 10^6$  CD34+ cells/kg above which further increases in cell dose may not substantially further hasten recovery.<sup>(36)</sup>

### **Tumour Control**

Since the benefit of mobilised blood cell transplants lies in shorter hospitalisations, fewer blood products, less antibiotic usage and probably lower procedure-related mortality, these do not by themselves lead to a different tumour control outcome than bone marrow transplants. However, the increased safety allows older patients and more diseases to be treated with high dose therapy. An even more important development is the use of multiple cycles of high dose therapy and blood cell rescue to deliver a higher total dose and dose rate.<sup>(20,37)</sup>

### **Malignant Contamination**

We know little about the possible significance of cells or molecular markers that are indicative of the target neoplasm in hemopoietic cells used for autologous transplantation. Whether they represent tumour stem cell or effete end cells is not easy to determine. The notable exception is in acute myeloid leukemia and neuroblastoma, where gene marking studies have shown that the autologous cells infused contributed to relapse.<sup>(38)</sup> Nonetheless, not all acute myeloid leukemia patients receiving autologous transplants relapse; thus, whether leukemic contamination occurs in all patients is not yet known. In acute lymphoid leukemia, persistence of the leukemia-associated molecular marker indicates a higher risk of relapse.<sup>(39)</sup> The presence of molecular markers of

lymphoma and myeloma has also been described.<sup>(40-43)</sup> The detection of epithelial cells in BM and mobilised blood cells of patients with breast and ovarian cancer has been reported by several groups.<sup>(44,45)</sup> Malignant contamination in mobilised blood cells appears to be less frequent than BM.<sup>(44,46)</sup>

What remains unanswered is whether the tumour stem cell exhibits the same phenotype as cells identified by these methods. Furthermore, whether all viable cells that bear the same immuno- and molecular phenotype are capable of metastasis is yet to be determined.

## **Paediatric Patients**

The most comprehensive series of reports of paediatric blood cell harvesting and transplants comes from Takaue et al.<sup>(47)</sup> The main areas distinguishing this paediatric population are special requirements for vascular access and leukapheresis, high progenitor yields and risks of 'stem cell exhaustion'.<sup>(48)</sup> The authors suggest that the mobilisable pool is a major component of the total body pool in small children during hemopoietic recovery.

## **Applications in Non-Malignant Diseases**

Multiple sclerosis, auto-immune diseases and paroxysmal nocturnal hemoglobinuria have all been targeted for autotransplantation using mobilised blood cells.<sup>(49,50)</sup> Mobilised blood cells have also been proposed as the source of CD34+ cells for gene therapy.<sup>(51)</sup> Specific diseases or therapeutic indications that have been targeted include methotrexate resistance,<sup>(52)</sup> chronic granulomatous disease<sup>(53)</sup> and Gaucher disease.<sup>(54)</sup>

## **Summary**

Mobilised blood cells are now used not only for single hemopoietic rescue following high dose therapy, but also to provide sufficient progenitor cells for multiple rescues, ex vivo processing and gene therapy. G-CSF alone or a combination of chemotherapy and hemopoietic growth factors such as G-CSF and GM-CSF are the two mobilisation protocols most commonly used. Hemopoietic reserve affects progenitor cell yield, so mobilisation protocols should incorporate mobilisation early in the treatment process. Adequate standardisation of stem cell measurement should allow reliable quantitation of hemopoietic reconstitutive capacity. Whenever possible, data on malignant contamination should be collected and collated with treatment outcome.

## **Acknowledgment**

The author acknowledges the secretarial assistance of Mrs Ann Haylock.

## **References**

1. Micklem HS, Anderson N, Ross R: Limited potential of circulating hemopoietic stem cells. *Nature* 256: 41-43, 1975.
2. Hershko C, Ho WG, Gale RP, Cline MJ: Cure of aplastic anaemia in paroxysmal nocturnal hemoglobinuria by marrow transfusion from identical twin: failure of peripheral leucocyte transfusion to correct marrow aplasia. *Lancet* 1: 945-947, 1979.
3. Abrams RA, Glaubiger D, Appelbaum FR, Deisseroth AB: Result of attempted hematopoietic reconstitution using isologous, peripheral blood mononuclear cells: A case report. *Blood* 56: 516, 1980.
4. Abrams RA, McCormack K, Bowles C, Deisseroth AB: Cyclophosphamide treatment expands the circulating hematopoietic stem cell pool in dogs. *J Clin Invest* 67: 1392, 1981.
5. Kessinger A, Armitage JO, Landmark JD, Smith DM, Weisenburger DD: Autologous peripheral hematopoietic stem cell transplantation restores hematopoietic function following marrow ablative therapy. *Blood* 71: 723-727, 1988.
6. To LB, Haylock DN, Kimber RJ, Juttner CA: High levels of circulating hemopoietic stem cells in very early remission from acute non-lymphoblastic leukaemia; their collection and cryopreservation. *Br J Haemat* 58: 399-410, 1984.
7. Juttner CA, To LB, Haylock DN, Branford A, Kimber RJ: Circulating autologous stem cells collected in very early remission from acute non-lymphoblastic leukaemia produce prompt but incomplete hemopoietic reconstitution after high dose melphalan or supralethal chemoradiotherapy. *Br J Haematol* 61: 739-745, 1985.
8. Korbling M, Dorken B, Ho AD, Pezzuto A, Hunstein W, Fliedner TM: Autologous transplantation of blood derived hemopoietic stem cells after myeloablative therapy in a patient with Burkitt's lymphoma. *Blood* 67: 629-632, 1986.
9. To LB, Dyson PG, Juttner CA: Cell-dose effect in circulating stem-cell autografting. *Lancet* 2: 404-405, 1986.
10. Reiffers L, Leverger G, Marit G et al: Hematopoietic reconstitution after autologous blood stem cell transplantation. In: Gale RP, Champlin RE, eds. *Bone Marrow Transplantation: Current Controversies*. Proceedings of Sandoz-UCLA Symposium. New York: Alan R Liss, p. 313, 1988.
11. To LB, Haylock DN, Thorp D, Dyson PG, Branford AL, Ho JQK, Dart GW, Roberts MM, Horvath N, Bardy P, Russell JA, Miller JL, Kimber RJ, Juttner CA: The optimisation of collection of peripheral blood stem cells for autotransplantation in acute myeloid leukaemia. *Bone Marrow Transplantation* 4: 41-47, 1989.
12. Gianni AM, Siena S, Bregni M et al: Granulocyte-macrophage colony-stimulating factor to harvest circulating hemopoietic stem cells for autotransplantation. *Lancet* 2: 580-585, 1989.
13. Rowlings PA, Rawling CA, To LB, Bayly JL, Juttner CA: A comparison of peripheral blood stem cell mobilization after chemotherapy with cyclophosphamide as a single agent in doses of 4 g/m<sup>2</sup> in patients with advanced cancer. *Aust N Z J Med* 22: 600-604, 1992.
14. Schwartzberg LS: Peripheral blood stem cell mobilization in the out-patient setting. In: EW Wunder EW, Henon PR (Eds): *Peripheral blood stem cell autografts*. *J Hematother* 177-184, 1993.

15. Haas R, Mohle R, Fruhauf S et al: Patient characteristics associated with successful mobilizing and autografting of peripheral blood progenitor cells in malignant lymphoma. *Blood* 83: 3787-3794, 1994.
16. Haynes A, Hunter A, McQuaker G, Anderson S, Bienz N, Russell NH: Engraftment characteristics of peripheral blood stem cells mobilised with cyclophosphamide and the delayed addition of G-CSF. *Bone Marrow Transplant* 16: 359-363, 1995.
17. Duhrsen U, Villeval J-L, Boyd J, Kannourakis G, Morstyn G, Metcalf D: Effects of recombinant human granulocyte colony-stimulating factor on hematopoietic progenitor cells in cancer patients. *Blood* 72: 2074-2081, 1988.
18. To LB, Haylock DN, Dowse T, Simmons PJ, Trimboli S, Ashman LK, Juttner CA: A comparative study of the phenotype and proliferative capacity of peripheral blood (PB) CD34+ cells mobilized by four different protocols and those of steady-phase PB and bone marrow CD34+ cells. *Blood* 84: 2930-2939, 1994.
19. Sheridan W, Begley CG, Juttner CA et al: Effect of peripheral blood progenitor cells mobilized by filgrastim (G-CSF) on platelet recovery after high dose chemotherapy. *Lancet* 339: 640-664, 1992.
20. Basser RL, To LB, Begley CG, Juttner CA, Maher DW, Szwe J, Cebon J, Collins J, Russell I, Fox RM, Sheridan WP, Green MD: Adjuvant treatment of women with high risk breast cancer using multiple cycles of high-dose chemotherapy supported by filgrastim (G-CSF)-mobilised peripheral blood progenitor cells. *Clin Cancer Res* 1: 715-721, 1995.
21. Basser R, Begley, Mansfield R, To LB, Juttner C, Maher D, Fox R, Cebon J, Szer J, Grigg A, Clark K, Marty J, Menchaca D, Thompson B, Russell I, Collins J, Green M: Mobilization of PBPC by priming with stem cell factor (SCF) before filgrastim compared to concurrent administration. *Blood* 86(Suppl 1): 2736 687a, 1995.
22. Socinski MA, Elias A, Schnipper L, Cannistra SA, Antman KH, Griffin JD. Granulocyte-macrophage colony stimulating factor expands the circulating haemopoietic progenitor cell compartment in man. *Lancet* 1(8596): 1194-1195, 1988.
23. Peters WP, Rosner G, Ross M, Vredenburgh J, Meisenberg B, Gilbert C, Kurtzberg J: Comparative effects of granulocyte-macrophage colony stimulating factor (GM-CSF) and granulocyte colony-stimulating factor (G-CSF) on priming peripheral blood progenitor cells for use with autologous bone marrow after high-dose chemotherapy. *Blood* 81: 1709-1719, 1993.
24. Vose JM, Kessinger A, Bierman PJ, Sharp G, Garrison L, Armitage JO: The use of rhIL-3 for mobilization of peripheral blood stem cells in previously treated patients with lymphoid malignancies. *Int J Cell Cloning* 10(Suppl 1): 62-64, 1992.
25. Vadham-Raj S, Broxmeyer HE, Andreeff M, Bandres JC, Buescher S, Benjamin RS, Papadopoulos NE, Burgess A, Patel S, Plager C, Hittelman WN, McAlister I, Garrison L, Williams DE: In vivo biologic effects of PIXY 321, a synthetic hybrid protein of recombinant human granulocyte-macrophage colony-stimulating factor and interleukin-3 in cancer patients with normal hematopoiesis: A phase I study. *Blood* 86: 2098-2105, 1995.
26. Basel K, McKenna HJ, Charrier K, Morrissey P, Williams DE, Lyman SD: Synergistic effects in vivo of flt3 ligand with GM-CSF in mobilization of colony forming cells in mice. *Blood* 86(Suppl 1): 499a, 1995.

27. Ganser A, Bergmenn M, Völkers B, Grützmacher P, Scigalla P, Hoelzer D: In vivo effects of recombinant human erythropoietin on circulating human hematopoietic progenitor cells. *Exp Hematol* 17: 433-435, 1989.
28. Lord BI, Woolford LB, Wood LM, Czaplowski LG, McCourt M, Hunter MG, Edwards RM: Mobilization of early hematopoietic progenitor cells with BB-10010: A genetically engineered variant of human macrophage inflammatory protein -1a. *Blood* 85: 3412-3415, 1995.
29. Pettengell R, Woll PJ, Chang J, Coutinho L, Crowther D, Testa NG: Effects of erythropoietin on mobilisation of haemopoietic progenitor cells. *Bone Marrow Transplant* 14: 125-130, 1994.
30. Fibbe WE, Hamilton MS, Laterveer LL et al: Sustained engraftment of mice transplanted with IL-1 primed blood-derived stem cells. *J Immunol* 148: 417-421, 1992.
31. Laterveer L, Lindley IJD, Heemskerk DPM, Camps JAJ, Pauwels EKJ, Willemze R, Fibbe WE: Rapid mobilization of hematopoietic progenitor cells in rhesus monkeys by a single intravenous injection of interleukin-8. *Blood* 87: 781-788, 1996.
32. Dreger P, Klöss M, Petersen B, Haferlach T, Löffler H, Loeffler M, Schmitz N: Autologous progenitor cell transplantation: prior exposure to stem cell-toxic drugs determines yield and engraftment of peripheral blood progenitor cell but not of bone marrow grafts. *Blood* 86: 3970-3978, 1995.
33. Hillyer CD: Large volume leukapheresis to maximize peripheral blood stem cell collection: *J Hematother* 2(4): 529-532, 1993.
34. Sheridan WP, Begley CG, To LB, Grigg A, Szer J, Maher D, Green MD, Rowlings PA, McGrath KM, Cebon J, Dyson P, Watson D, Bayly J, deLuca E, Tomita D, Hoffman E, Morstyn, Juttner CA: Phase II study of autologous filgrastim (G-CSF) mobilized peripheral blood progenitor cells to restore hemopoiesis after high-dose chemotherapy for lymphoid malignancies. *Bone Marrow Transplant* 14: 105-111, 1994.
35. Bender JG, To LB, Willism S, Schwartzberg LS: Defining a therapeutic dose of peripheral blood stem cells. *J Hematother* 1: 329, 1992.
36. Weaver CH, Hazelton B, Birch R, Palmer P, Allen C, Schwartzberg L, West W: An analysis of engraftment kinetics as a function of the CD34+ content of peripheral blood progenitor cell collections in 692 patients after the administration of myeloablative chemotherapy. *Blood* 86: 3961, 1995.
37. Bezwoda WR, Seymour L, Dansey RD: High-dose chemotherapy with hematopoietic rescue as primary treatment for metastatic breast cancer: A randomized trial. *J Clin Oncol* 13: 2483-2489, 1995.
38. Brenner MK, Rill DR, Moen RC, Krance RA, Mirro J, Anderson WF, Ihle JN: Gene-marking to trace origin of relapse after autologous bone marrow transplantation. *Lancet* 341: 85-86, 1993.
39. Brisco MJ, Condon J, Hughes E, Neoh SH, Sykes PJ, Seshadri R, Toogood I, Waters K, Tauro G, Ekert H: Outcome prediction in childhood acute lymphoblastic leukaemia by molecular quantification of residual disease at the end of induction. *Lancet* 2: 343, 196-200, 1994.

40. Sharp JG, Kessinger A, Vaughan WP et al: Detection and clinical significance of minimal tumor cell contamination of peripheral stem cell harvests. *Intl J Cell Cloning* 10: 92-94, 1992.
41. Craig JL, Langlands K, Parker AC, Anthony RS: Molecular detection of tumor contamination in peripheral blood stem cell harvests. *Exp Hematol* 22: 898-902, 1994.
42. Brugger W, Bross KJ, Glatt M, Weber F, Mertelsmann R, Kanz L: Mobilization of tumour cells and hematopoietic progenitor cells into peripheral blood of patients with solid tumours. *Blood* 83: 636-640, 1994.
43. Henry JM, Sykes PJ, Brisco MH, To LB, Juttner CA, Morley AA: Comparison of myeloma cell contamination of bone marrow and peripheral blood stem cell harvests. *Br J Haematol*, in press.
44. Ross AA, Cooper BW, Lazarus HM et al: Detection and viability of tumour cells in peripheral blood stem cell collections from breast cancer patients using immunocytochemical and clonogenic assay techniques. *Blood* 82: 2605-2610, 1993.
45. Simpson SJ, Vachula M, Kennedy MJ, Kaizer H, Coon JS, Williams S, Van Epps D: Detection of tumor cells in the bone marrow, peripheral blood, and apheresis products of Ghalie breast cancer patients using flow cytometry. *Exp Hematol* 23: 1062-1068, 1995.
46. Passos-Coelho JL, Ross AA, Moss TJ, Davis JM, Huelskamp A-M, Noga SJ, Davidson NE, Kennedy MJ: Absence of breast cancer cells in a single-day peripheral blood progenitor cell collection after priming with cyclophosphamide and granulocyte-macrophage colony-stimulating factor. *Blood* 85: 1138-1143, 1995.
47. Takaue Y, Kawano Y, Abe T, Okamoto Y, Suzue T, Shimizu T, Saito S, Sato J, Makimoto A, Nakagawa R: Collection and transplantation of peripheral blood stem cells in very small children weighting 20kg or less. *Blood* 86: 372-380, 1995.
48. Takaue Y, Watanabe T, Kawano Y, Koyama T, Huq AHM, Ninomiya T, Kuroda Y: Sustained cytopenia in small children after leukapheresis for collection of peripheral blood stem cells. *Vox Sang* 57: 168, 1989.
49. Prince GM, Nguyen M, Lazarus HM, Brodsky RA, Terstappen LWMM, Medof ME: Peripheral blood harvest of unaffected CD34+ CD38- hematopoietic precursors in paroxysmal nocturnal hemoglobinuria. *Blood* 86: 3381-3386, 1995.
50. Burt RK, Burns W, Hess A: Bone marrow transplantation for multiple sclerosis. *Bone Marrow Transplant* 16:1-6, 1995.
51. Dunbar CD, Cottler-Fox M, O'Shaughnessy JA, Doren S, Carter C, Berenson R, Brown S, Moen RC, Greenblatt J, Stewart FM, Leitman SF, Wilson WH, Cowan K, Young NS, Nienhuis AW: Retrovirally marked CD34- enriched peripheral blood and bone marrow cells contribute to long-term engraftment after autologous transplantation. *Blood* 85: 3048-3057, 1995.
52. Flasshove M, Banerjee D, Mineishi S, Li M-X, Bertino JR, Moore MAS: Ex vivo expansion and selection of human CD34+ peripheral blood progenitor cells I of a mutated dihydrofolate reductase cDNA via retroviral gene transfer. *Blood* 85: 556-574, 1995.
53. Malech HL, Sekhsaria S, Whiting-Theobald, Linton GF, Vowells LF, Miller JA, Holland SM, Leitman SF, Carter CS, Read EJ, Butz R, Wannebo C, Fleisher TA,

- Deans RJ, Spratt SK, Maack CA, Rokovich JA, Cohen LK, Maples Gallin JI: Development of a phase I clinical trial of gene therapy for chronic granulomatous disease. *Gene Therapy/Transfer Blood* 85:suppl 1, 295a, 1995.
54. Nimgaonkar M, Mierski J, Beeler M, Kemp A, Lancia J, Mannion-Henderson J, Mohny T, Bahnson A, Rice E, Ball ED, Barranger JA: Cytokine mobilization of peripheral blood stem cells in patients with Gaucher disease with a view to gene therapy. *Exp Hematol* 23: 1633-1641, 1995.