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Conditioning regimens

Pharmacodynamics of tandem high-dose melphalan with peripheral blood stem cell transplantation in children with neuroblastoma and medulloblastoma

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Summary:

Repeated high-dose (HD) chemotherapy with peripheral blood stem cell (PBSC) transplantation is a new modality aimed at increasing both the dose and its intensity in the treatment of chemosensitive tumours. The aim of this study was to evaluate the tolerance, pharmacokinetics (PK) and pharmacodynamics (PD) of HD single-agent melphalan administered over two consecutive courses (C1 and C2) in children. Twenty-one patients (10 girls) with a median age of 4.1 years (range 8 months–14 years) were entered into this study. Five had metastatic neuroblastoma (NB) and 16 a cerebral primitive neuroectodermal tumour (PNET). Melphalan was given at a dose of 100 mg/m^2 every 21 days. PBSCs were infused at a median number of 2.98×10^6 CD34$^+$ cells/kg. Forty courses, ie 21 C1 and 19 C2, were administered. Both courses were well tolerated. The median duration of ANC $<500/\mu$l was 7 and 6 days after C1 and C2, respectively. Platelet recovery (not mandatory to continue the HD strategy) was achieved in 52% of courses. GI toxicity was mild to moderate. The melphalan AUC ranged from 177 to 475 μg·min/ml (no difference between C1 and C2). Prolonged neutropenia was associated with a young age ($P < 0.001$) and a low amount of CFU-GM ($P = 0.002$). A long time to platelet recovery was associated with a high AUC ($P = 0.004$) and a young age ($P = 0.02$). Grade 1 or 2 GI toxicity was associated with a high AUC ($P = 0.015$). Partial remission was observed in 11/14 patients with measurable cerebral PNET. In conclusion, tandem HD melphalan is feasible and safe in children, and achieved a high response rate in cerebral PNET. The observed PK–PD relationships may help us design PK-guided outpatient treatment. *Bone Marrow Transplantation* (2001) **27,** 471–477.

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Melphalan is a bi-functional alkylating agent with demonstrated activity in several malignant diseases in children.[1] High-dose melphalan is currently used as a single agent or in combination with other anticancer drugs before autologous hematopoietic stem cell transplantation in the treatment of metastatic neuroblastoma, rhabdomyosarcoma and Ewing's sarcoma. High-dose melphalan, in combination with total body irradiation and chemotherapy before allogeneic bone marrow transplantation, is used in poor-risk acute lymphocytic leukaemia. In these regimens, high-dose melphalan is usually administered at doses ranging from 140 to 200 mg/m^2. In addition, recent phase II studies have shown that i.v. melphalan, at the conventional dose (45 mg/m^2 every 4 weeks), is an active drug in cerebral primitive neuroectodermal tumours (PNETs) in children, including medulloblastomas.[2]

Haematopoietic growth factors and haematopoietic stem cells collected from peripheral blood after mobilisation by chemotherapy and/or growth factors have rendered possible sequential administration of high-dose chemotherapy courses. Repeated high-dose chemotherapy is a novel way to further increase both the dose and dose density of agents used to treat chemosensitive tumours.[3] These new dose-intensive strategies are being extensively evaluated in breast cancer patients.[4,5] High-dose melphalan is particularly attractive in this regard. Clinical safety after a single high dose of melphalan is well defined: dose escalation beyond 200 mg/m^2 is limited by mucosal toxicity, ie mucositis and diarrhoea. In addition, high-dose melphalan could be administered on an outpatient basis.[5,6]

Plasma pharmacokinetics of high-dose melphalan have been evaluated in adults and children.[1] The melphalan clearance rate is highly variable with a 10-fold interpatient variation but no difference has so far been reported between adults and children.[7,8] This wide variability in the clearance rate also signifies wide variability in systemic exposure when a fixed dose is administered. Pharmacokinetically

guided dose adjustment could reduce this interpatient variability and improve tolerance and efficacy of high-dose melphalan. However, minimal information has been obtained to date on the pharmacodynamics of high-dose melphalan.

The demonstrated activity of melphalan in childhood cancers provides a solid rationale for repeated administration of high-dose melphalan. We have evaluated the tolerance and activity of tandem high-dose melphalan given as a single agent before autologous peripheral blood stem cell (PBSC) transplantation, as part of sequential high-dose chemotherapy protocols in metastatic neuroblastoma and cerebral PNETs. The objective was to administer the maximum tolerated dose of melphalan (200 mg/m^2) split into two consecutive courses, ie 100 mg/m^2 every 21 days. The aim of this study was to analyse the pharmacokinetics and pharmacodynamics of high-dose melphalan when administered sequentially as a single agent in children with malignant disease in order to define the level of systemic exposure that could be targeted in a prospective pharmacologically guided dose-adjustment strategy.

Materials and methods

Patients

Twenty-one patients were entered in this single-institution study. They were 11 males and 10 females with a median age of 4.1 years (range, 0.7–14.2 years). Sixteen patients had a brain PNET that was metastatic in 14 of them. The primary tumour was located in the cerebellum (13 patients), in the brain stem and in the pineal gland. One patient had diffuse meningitis without a primary tumour. Five patients had a stage 4 abdominal neuroblastoma with bone marrow metastases. Nineteen patients had measurable disease at the time of treatment and were evaluable by MRI, MIBG scan or CT scan. Two patients had non-measurable disease: one patient was in CR of metastatic medulloblastoma and one had diffuse meningeal disease. All tumours were histologically proven. Parents had given their consent.

Treatment

Melphalan (Alkeran; Laboratoires Welcome, Issy-les-Moulineaux, France) was administered intravenously via a central indwelling catheter at a dose of 100 mg/m^2 in NaCl 0.9% for two consecutive courses with a planned interval of 21 days between courses. According to the protocol and the manufacturer's recommendations, melphalan was administered as a rapid infusion (2–5 min) in the first five patients. As immediate tolerance was poor with hot flushes and grogginess, the duration of the infusion was extended to 15 min in the next 16 patients, whereupon, immediate tolerance was excellent. Hydration (3 l/m^2/day) was started 24 h prior to melphalan administration. Hydration was well tolerated in brain tumour patients providing a stepwise increase in fluid volumes over 3 days.

Sequential high-dose melphalan was administered as part of intensive chemotherapy protocols. Patients with stage 4 neuroblastoma were initially treated with standard conventional chemotherapy comprising etoposide/cisplatin and etoposide/high-dose cyclophosphamide/doxorubicin. Patients with high-risk cerebral PNET, ie metastatic tumour or a tumour residue, were initially treated with etoposide/carboplatin over two consecutive courses. After two courses of sequential high-dose melphalan, a third course of high-dose combined chemotherapy with autologous PBSC transplantation was planned with busulfan/melphalan in patients with neuroblastoma and busulfan/thiotepa in patients with cerebral PNET.[9] An absolute neutrophil count (ANC) above 500/μl and resolution of extra-haematological toxicity were required before starting the second and third courses of high-dose chemotherapy. Conversely, platelet recovery was not mandatory.

PBSC harvesting

Mobilised PBSC were collected by one to three leukapheresis after conventional chemotherapy combined with G-CSF (Neupogen, Amgen, France) (5 μg/kg/day) in 17 patients or at steady state after G-CSF administered at a dose of 10 μg/kg/day $\times$ 5 in four patients. The number of CD34$^+$ cells collected ranged from 3.3 to 59.5 $\times$ 10^6 cells/kg (median, 10.2 $\times$ 10^6 CD34$^+$/kg). Bone marrow was harvested in one patient with a low CD34$^+$ cell count in PBSC. Peripheral blood stem cells were cryopreserved in 10% dimethyl sulfoxide in liquid nitrogen.

PBSC transplant and transfusion procedure

The number of CFU-GM and C34$^+$ cells in each graft was quantified by *in vitro* culture and FACS analysis, respectively. The cryopreserved PBSCs were thawed and infused rapidly through a central venous catheter on day 0, ie 24 h after administration of melphalan. All patients received G-CSF (5 μg/kg/day) from the first day when the ANC was below 500/μl until it exceeded 500/μl for 2 consecutive days. All blood products were leukocyte-free and irradiated to a dose of 25 Gy. CMV-negative products were given to patients with CMV-negative serology. Packed red blood cells were transfused as needed to maintain the haemoglobin above 70 g/l. Platelets from a unique donor were transfused to maintain the platelet count above 20 000/μl and 50 000/μl in neuroblastoma and brain tumour patients, respectively.

Supportive care

All children were nursed in isolated laminar airflow single rooms until the ANC was above 500/μl for 2 consecutive days. Episodes of fever and documented infections were treated with appropriate antibiotic therapy. Mouth washes with 1.4% bicarbonate and chlorhexidin were systematically administered six times a day.

Evaluation of toxicity

Complete blood counts were obtained thrice weekly. Renal function, serum electrolytes and hepatic enzymes were evaluated twice a week. Neutrophil recovery was defined as a neutrophil count above 500/μl without G-CSF support. Platelet recovery was defined as a platelet count exceeding

50 000/μl independent of platelet transfusions. Extra-haematological toxicity was graded according to Bearman's grading system.[10]

Evaluation of tumour response

Tumour response in patients with brain tumours was evaluated after two courses by MRI scan, including spinal evaluation. Complete response (CR) was defined as a complete resolution of all measurable tumours. Partial response (PR) was a decrease greater than 50% in the product of the perpendicular tumour diameters as compared to baseline tumour imaging. Stable disease (SD) was defined as a decrease in tumour volume less than 50% or an increase in tumour volume less than 25%. Progressive disease (PD) was defined as an increase in tumour measurement greater than 25%. In patients with stage 4 neuroblastoma, response was evaluated according to the revised international criteria.[11] Complete response was defined as complete resolution of the primary tumour and metastatic lesions with normal catecholamines. Partial response was defined as a decrease in the primary tumour of greater than 50% and a decrease in the number of positive metastatic sites greater than 50%. All CT and MRI scans were reviewed by a single radiologist and all mibg scans were reviewed by a single physician.

Pharmacokinetics

We have previously established a limited sampling strategy derived from a Bayesian pharmacokinetic model of i.v. melphalan administered as a 2 to 5 min infusion.[12] According to this model, total blood samples are drawn before the infusion and after the end of the infusion at 5, 23 and 90 min. When the duration of the infusion was extended to 15 min due to poor immediate tolerance, complete pharmacokinetic sampling was performed and total blood samples were obtained before the infusion and at 5, 23, 60, 90, 120, 180 min after the end of the infusion. Samples were immediately placed on ice. Plasma was separated by centrifugation at 2000 *g* for 10 min at 0–4°C and frozen at –20°C until analysis. Melphalan was assayed using a high-pressure liquid chromatography assay, as previously described.[13]

The pharmacokinetic analysis was performed with the APIS program.[14] The plasma concentration-versus-time curve followed a bi-exponential decay during all courses. For patients with full blood sampling, pharmacokinetic parameters were obtained on an individual basis with maximum likelihood estimation (MLE) using a two-compartment model according to the following equations:

$$(0 < t < T) : Y = \sum_{i=1}^{2} A_i \, (1 - e^{-a_i t})/a_i T$$

$$(t \geqslant T): Y = \sum_{i=1}^{2} A_i \, (1 - e^{-a_i t}) \, e^{-a_i (t-T)}/a_i T$$

where Y is the plasma melphalan concentration, coefficients A_i and exponents ai ($i = 1,2$) are subject-specific coefficients and T is the duration of the infusion.

For patients with limited sampling, pharmacokinetic parameters were obtained using Bayesian estimation. The accuracy of Bayesian estimates for clearance and the AUC were first evaluated in patients with full sampling by comparing the pharmacokinetic parameters obtained using an individual analysis and a Bayesian estimation. The predicted AUC performance was then analysed according to the suggestions of Sheiner and Beal[15] by computing bias and precision between individual and Bayesian estimates. Once the accuracy of the AUC Bayesian calculation had been validated in this population, the AUC could be computed in patients with only three blood samples using the Bayesian approach.

Pharmacokinetic parameters after courses 1 and 2 were compared using a Wilcoxon non parametric test.

Pharmacodynamics

We assessed the effects of age (in years), the number of transplanted PBSCs, namely $CD34^+$ cells/kg or CFUGM/kg, and systemic exposure to melphalan measured by the AUC (in μg·min/ml) on toxicity that was evaluated after each of the two courses. Evaluation of toxicity was based on the duration of neutropenia (in days), time to platelet recovery (in days) and GI toxicity (yes/no). Time to platelet recovery was censored if thrombocytopenia persisted when the next chemotherapy course was due to be started; in these cases, we only knew that the time to platelet recovery was greater than the number of days between the two courses. GI toxicity was recorded as the presence or absence of grade 1 or 2 diarrhoea and/or mucositis.

As correlation was possible between toxicity observed after course 1 and that observed after course 2, we used regression models, which take this correlation into account in the pharmacodynamic analysis. Marginal generalised linear models and associated generalised estimating equations[16] were used for the duration of neutropenia and GI toxicity. A marginal proportional hazard model was used for the censored time to platelet recovery.[17] A robust Wald test was used to test the effect of each characteristic (age, the number of PBSCs, the melphalan AUC) on toxicity. Statistical analysis was performed using Splus, version 3.2 (MathSoft, Seattle, WA, USA) and a generalised estimating equation library (http://lib.stat.cmu.edu/S/gee).

Results

Tolerance

Twenty-one patients were treated: 19 had two courses and two patients received only one course of high-dose melphalan because of tumour progression. There were no dose reductions in any patients. The second course was administered 21 to 34 days after the first one (median 24 days). No attempts were made to shorten the interval between the two courses. Haematological toxicity is detailed in Table 1. Regarding PBSC transplants, more than 2×10^6 $CD34^+$ cells/kg were transfused during 70% of cycles. Both courses were clinically well tolerated. Neutropenia lasted less than 7 days in 67% of courses. Platelet recovery had been achieved in 21 of 40 courses (52%) at the time of the

Table 1 Haematological toxicity

	Course 1	*Course 2*
No. of patients	21	19
No. of $CD34^+$ cells transfused (10^6/kg) median (range)	2.7 (1.0–7.9)	3.7 (1.2–27.4)
No. of CFU-GM transfused (10^4/kg) median (range)	41.0 (14.0–99)	50.7 (9.8–142.0)
Days with ANC $<500/\mu$l median (range)	7 (5–14)	6 (4–10)
Days with platelets $<50\,000/\mu$l		
evaluable/non-evaluable[a]	11/10	10/9
median (range) among evaluable	10 (2–17)	13 (5–19)
No. of platelet transfusions median (range)	3 (1–17)	5 (2–12)
No. of RBC transfusions median (range)	1 (0–5)	1 (1–3)
No. of cycles with		
fever	15	12
ANC $<500/\mu$l of >7 days	7	6

[a]A platelet count above 50 000/μl was not mandatory to continue the treatment.

next high-dose chemotherapy course. Of the 19 patients who received two courses, seven exhibited platelet recovery after both courses, platelet recovery did not occur after either course in 10, and only two patients had a different recovery pattern between the two courses.

No infection was documented and no fever occurred in 13 of 40 courses. None of the patients experienced life-threatening toxicity. Gastro-intestinal toxicity was mild to moderate. According to Bearman's grading system, grade 2 mucositis requiring narcotic analgesia occurred in four of 40 courses (10%) and grade 2 diarrhoea in three of 40 courses (7.5%). An erythematous rash was observed in 10 courses and resolved rapidly. One patient developed melanodermia.

Tumour response

Nineteen of the 21 patients were evaluable for tumour response. Out of the five patients with stage 4 neuroblastoma, one was in partial remission on the metastatic sites after two courses, three had stable disease and one experienced disease progression after one course. Out of 14 patients with measurable cerebral PNET, 11 partial remissions were observed after two courses. Three tumour progressions occurred, one of them after the first course in a patient with a non-metastatic brain stem PNET.

Pharmacokinetics

Pharmacokinetic sampling was carried out in 37 out of 40 courses: 18 during course 1 and 19 during course 2. Plasma pharmacokinetics were evaluated after both courses in 16 patients. The analysis was performed in 14 courses using a Bayesian estimation and in 23 courses (12 in course 1, 11 in course 2) using MLE. The accuracy of the Bayesian estimation was verified using the 23 courses with full sampling available. No statistically significant difference was observed between MLE and Bayesian estimates of the AUC (Wilcoxon test). No bias was observed (confidence interval: (−1.5%; 0.6%)) and precision was satisfactory (2.8%).

The mean total body clearance rate was 305 and 346 ml/min/m^2 after course 1 and 2, respectively (Table 2). Interpatient variability was relatively low with coefficients of variation of 18.5% and 25% for course 1 and 2, respectively. Overall, systemic exposure to melphalan ranged from 177 to 475 μg·min/ml, ie a 2.5-fold variation. The median value was 313 μg·min/ml with 25% and 75% percentiles of 271 and 359 μg·min/ml, respectively. No significant difference was observed between courses 1 and 2, as studied by nonparametric (Wilcoxon test) analysis of paired data in 16 patients (Figure 1).

Pharmacodynamics

Myelosuppression and GI toxicity were the main adverse side-effects related to high-dose melphalan in our study, as in the literature.

Using a marginal linear model, age and CFU-GM were shown to influence the duration of neutropenia significantly on both univariate ($P < 0.001$ and $P = 0.002$, respectively) and multivariate analyses ($P < 0.001$ for both of them). Prolonged neutropenia was associated with a low number of transfused CFU-GM and a young age. No relationship was observed when PBSCs were recorded as $CD34^+$ cells ($P = 0.12$). Systemic exposure to melphalan did not influence granulocyte recovery ($P = 0.37$).

A marginal proportional hazard model was used to study the relationship between time to platelet recovery (possibly censored) and patient characteristics. AUC and age influenced the time to platelet recovery significantly in both univariate ($P = 0.004$ and $P = 0.02$, respectively) and multivariate analyses ($P = 0.004$ and $P = 0.03$, respectively). A long time to platelet recovery was associated with a high AUC and a young age. Figure 2 represents the time to platelet recovery as a function of the AUC during both courses. The number of transfused PBSCs did not influence the time to platelet recovery.

Finally, the relationship between GI toxicity and AUC was studied using a marginal logistic model. The AUC contributed significantly to the occurrence of GI toxicity ($P = 0.015$). The occurrence of grade 1 or 2 GI toxicity was associated with high systemic exposure to melphalan, but age did not impact on this adverse effect ($P = 0.07$).

Table 2 Pharmacokinetic parameters of high-dose melphalan

	Course 1	*Course 2*
No. of courses	21	19
No. of courses with PK sampling	18	19
Clearance (ml/min/m^2)		
mean (s.d.)	305 (57)	346 (88)
(range)	(211–404)	(210–565)
AUC (μg.min/ml)		
mean (s.d.)	341 (64)	300 (66)
(range)	(248–475)	(177–451)

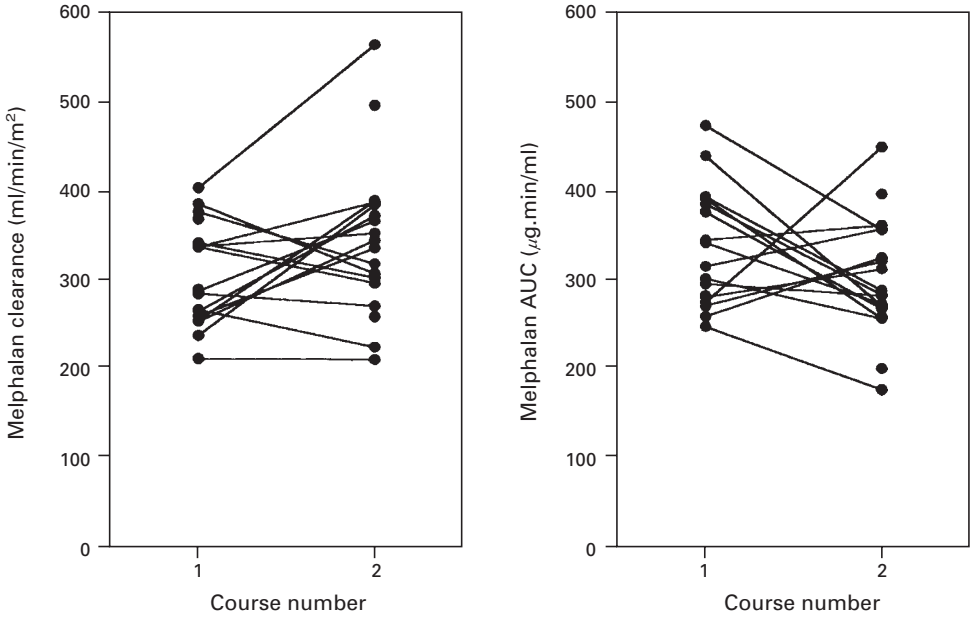

Figure 1 Intra-individual variability of melphalan clearance and AUC. Each solid line joins data obtained from the same patient after course 1 and course 2.

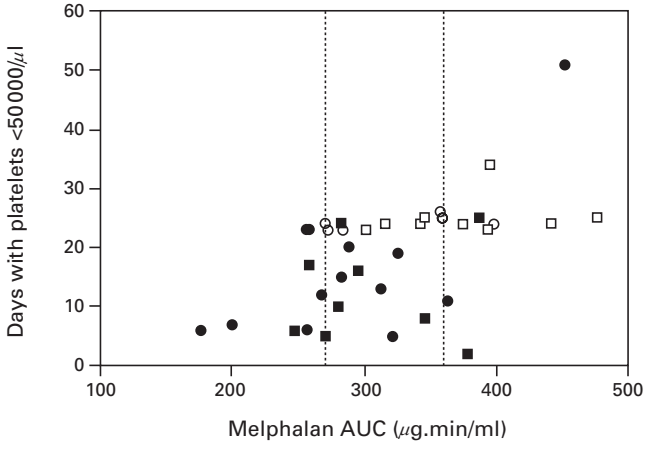

Figure 2 Influence of AUC on time to platelet recovery after course 1 (square symbols) and course 2 (circle symbols). Filled symbols represent courses with platelet recovery before the start of next treatment. Open symbols represent courses without platelet recovery before the start of next treatment. In these cases, data were censored on the first day of next treatment. The vertical dashed lines represent the 25% (left) and 75% (right) percentiles of the AUC in the entire population, ie 271 and 359 μg·min/ml, respectively.

Discussion

Our first objective was to evaluate the feasibility and safety of two cycles of high-dose melphalan with PBSC transplantation separated by a 21-day interval in children with malignant solid tumours. Overall, this therapy appears to be safe with a short duration of neutropenia (67% of cycles with less than 7 days of grade 4 neutropenia), mild to moderate extra-haematological toxicity and no life-threatening toxicity. In the present study, platelet recovery, although not required to continue the treatment, was achieved in only 52% of courses before the start of the next course. Rules are very strict regarding platelet transfusion in patients with brain tumours. In our study, the median number of transfusions was relatively low, with a median of three and five after each course. Mobilised peripheral blood stem cells are more effective than bone marrow stem cells for achieving haematological recovery after high-dose chemotherapy regimens, and especially the recovery of platelets.[18] The myelosuppression observed is comparable to (or even lower than) that reported after higher doses of single-agent melphalan with PBSC support.[4,5,19] In a double dose-density program in patients with metastatic breast cancer, Ayash *et al*[5] reported a median of 6 days (range 4–17 days) to ANC recovery above 500/μl and of 4 days (range 0–31$^+$) to platelet recovery above 20 000/μl after 140–180 mg/m^2. In this study, platelet recovery was not mandatory for the second transplant. In four of 67 patients, platelet recovery was not achieved at the time of the second intensification which occurred 27 to 37 days after the first intensification.

Following a dose of 100 mg/m^2, melphalan-induced extra-haematological toxicity was mild to moderate with no difference between courses 1 and 2, suggesting the absence of a cumulative dose-effect. No GI toxicity was observed in 57% of courses and the incidence of grade 2 diarrhoea or stomatitis did not exceed 10%. With higher doses of melphalan, the incidence of grade 3 stomatitis has been reported to range from 19% at a dose of 140–180 mg/m^2,[5] to 31% at 200 mg/m^2.[6] The tandem HD-melphalan regimen could be considered for outpatient treatment, as already demonstrated for higher doses of melphalan in multiple myeloma.[6] Platelet recovery was the overriding concern although there were no major immediate consequences (no haemorrhage and platelet transfusion requirements were limited). The influence of repeated high-dose regimens on bone marrow reserves and long-term bone marrow function will need to be evaluated, especially in patients in whom platelet recovery is not achieved between courses.

After high-dose chemotherapy, the kinetics of bone marrow reconstitution are related to the amount and quality of transplanted hematopoietic stem cells, provided that: (1) no cytotoxic drug and/or plasma metabolite concentration is present at the time of transplantation; and that (2) high-dose chemotherapy or previous treatments have not modified the bone marrow stroma to the extent that stem cell homing would be jeopardised. Out of 336 autotransplant procedures after high-dose melphalan in multiple myeloma patients, the median time to a platelet count exceeding 50 000/μl was significantly shorter (12 days) in patients who received more than 2×10^6 CD34$^+$ cells/kg than in patients who received less than 2×10^6 CD34$^+$ cells/kg (49 days).[6] In our study, more than 2×10^6 CD34$^+$/kg cells were transplanted in the great majority of cycles and duration of neutropenia did not correlate with the number of CD34$^+$ cells transplanted. A significant positive correlation was observed for the amount of transfused CFU-GM. This suggests that the amount of clonogenic progenitors already committed to granulocyte differentiation is a better predictor of the duration of neutropenia when rich PBSC grafts are used.

The number of transplanted PBSCs had no impact on platelet recovery. It has been suggested that early bone marrow recovery after PBSC transplantation is due to the presence of progenitors already committed to haematopoietic differentiation. If such is the case, platelet recovery would be better correlated with the number of clonogenic progenitors already committed to platelet differentiation, ie CFU-Meg, rather than the number of CD34$^+$ cells when rich grafts are used. Recently, Maharaj *et al*[20] reported that a low number of megakaryocytic progenitors was significantly associated with delayed platelet engraftment in patients who received a rich autologous PBSC transplant ($>10^7$ CD34$^+$/kg).[20] In addition, PBSC were collected in most of the patients after etoposide–carboplatinum, a combined regimen known to induce thrombocytopenia. The effect of this chemotherapy regimen on the amount and quality of mobilised CFU-Meg is unknown. Measurement of CFU-Meg, which is not routinely performed, may further improve PBSC transplantation after high-dose chemotherapy. Delayed platelet recovery was significantly correlated with high systemic exposure to melphalan. As these high AUCs were not associated with prolonged elimination half-lives (data not shown), we can rule out the existence of cytotoxic plasma melphalan levels at the time of PBSC transplantation (ie 24 h). As exposure to melphalan impacted on platelet recovery, self bone marrow may be more instrumental in platelet reconstitution than is the graft. The likelihood is even more plausible since unlike busulfan, melphalan is not a myeloablative drug. After a dose of 70–100 mg/m^2 without marrow rescue in patients with leukaemia, spontaneous bone marrow recovery occurred after a median of 29 and 28 days of neutropenia and thrombocytopenia.[21] We hypothesise that after a dose of 100 mg/m^2 of melphalan, platelet recovery is probably achieved through the dual intervention of grafted cells and self bone marrow, and is influenced by both exposure to the drug and the number of CFU-Meg in the graft.

Age was found to influence haematological recovery significantly, as demonstrated by neutrophil and platelet counts. Prolonged and intensive chemotherapy before PBSC collection is known to alter the quantity and quality of collected stem cells[22] but the effect of age has never been reported. These results suggest that the quality of PBSCs obtained after mobilisation by conventional chemotherapy and G-CSF treatment may be different in young and older patients.

Passage of melphalan into the central nervous system is low. Melphalan is detectable in some patients with a CSF to plasma concentration ratio up to 10%.[7] The blood–brain barrier is the main obstacle for melphalan penetration in normal brain. Drug distribution into the brain depends on active transport via the high-affinity neutral amino acid transport system through the blood-brain barrier.[23] However, the blood–brain barrier is largely altered in primary brain tumours and brain metastases. Despite a low brain distribution, melphalan has shown antitumour activity in some brain tumours, mainly medulloblastoma and other PNETs. Chamberlain *et al*[24] have reported the activity of oral melphalan (40 mg/m^2 over 5 days every 4 to 6 weeks) in 37 patients with malignant primary brain tumours. One partial remission lasting 14 months was observed in one out of five patients with a medulloblastoma. Friedman *et al*[2] reported the activity of i.v. melphalan (45 mg/m^2 every 4 weeks without hematopoietic growth factors) in 16 children with a CNS PNET, ie medulloblastoma and pineoblastoma. After two courses, one complete and two partial remissions were observed out of 12 patients with a relapsed tumour. In addition, three out of four patients with a newly diagnosed tumour experienced a partial response. The overall response rate was 37%. Myelosuppression was the main toxicity with 75% of patients experiencing anaemia, neutropenia, and thrombocytopenia. We have explored a higher dose of melphalan with PBSC support. Eleven out of 14 patients with a measurable brain PNET showed tumour reduction greater than 50% after two courses of 100 mg/m^2. The overall response rate was 79% (95% confidence interval = 57–100%). No relationship between systemic exposure to melphalan and tumour response could be documented. Altogether, these results suggest that melphalan is worth considering in high-dose chemotherapy programs for the treatment of medulloblastoma in children.

Dose-intense chemotherapy programs are designed to increase tumour cell kill by shortening the interval between courses of conventional or high-dose chemotherapy. We have shown that the incidence of GI toxicity and delayed platelet recovery were significantly associated with high systemic exposure to melphalan. Individual dose adjustment may reduce inter patient variability in systemic exposure observed after a fixed dose and help control toxicity to normal tissues. Dose-intense therapy may be improved by sticking to the 21-day schedule. As no threshold could be defined, we propose targeting the AUC within a window ranging from 271 to 359 μg·min/ml, ie the 25% and 75% percentiles, as compared to the 177–556 μg·min/ml range observed. This would reduce interpatient variability from 2.82 to 1.33. Successful targeting of this AUC window demands ± 15% accuracy of the method used to estimate melphalan clearance. We have already shown that a melphalan AUC was correctly targeted by using a test dose, with a measured error below 15% in 12 out of 14 cycles.[25]

Tandem high-dose melphalan is safe and feasible in children and could be further developed as an outpatient treatment. Individual dose adjustment based on observed pharmacodynamic relationships may further improve this dose-intense therapy by adhering to the 21-day schedule. Long-term platelet recovery remains to be evaluated prospectively.

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