the standard cytogenetics classification (unfavorable, intermediate, favorable). We then compared each of this subgroup with the corresponding population of patients presenting the same risk factors but owning a HLA identical sibling. In the standard risk group (n = 66), no-donor patients presented a favorable 10-year survival (74%) compared with patients with a donor (51%) (P = not significant), and allo-SCT should not be recommended. In the poor risk group (n = 131), patients without a donor presented a poor outcome (10-year OS = 17%), which was not better if a donor existed (28%) (P = .29); in these patients with a poor outcome whatever the treatment, investigational studies should be recommended. In the intermediate risk group (n = 275), allo-SCT offered a better outcome; the 10-year probabilities of relapse, nonrelapse death, OS, and LFS (no donor vs donor) were 47% versus 18% ($P < .0001$), 7% versus 17% ($P = .02$), 47% versus 63% ($P = .02$), and 45% versus 64% ($P = .001$), and the 10-year OS was 56% versus 41% ($P = .01$). Allo-SCT represent a real chance of long-term outcome and might benefit from recent advances. We conclude that long-term outcome might be achieved in some subgroups of patients and that the indication for allo-ASCT could be assessed trough a simple classification based on common parameters.

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TOTAL BODY IRRADIATION (TBI) AND G-CSF-COMBINED HIGH-DOSE CYTARABINE AS A PREPARATIVE REGIMEN FOR ALLOGENEIC HEMATOPOIETIC STEM CELL TRANSPLANTATION FOR ACUTE MYELOGENOUS LEUKEMIA (AML) AND ADVANCED MYELODYSPLASTIC SYNDROME (MDS)

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The most common reason for failure in later-stage patients after allogeneic hematopoietic stem cell transplantation (allo-HSCT) for AML and advanced MDS is leukemic relapse, demonstrating the inability of preparative regimen to completely eradicate leukemic cells. We report the outcome of allo-HSCT for myeloid malignancies using myeloablative preparative regimen including TBI and high-dose cytarabine. In addition, granulocyte colony-stimulating factor (G-CSF) was simultaneously administered with cytarabine to increase the susceptibility of leukemic cells to cytarabine. **Patients and Methods:** Patients with myeloid malignancies, including AML, AML evolving from MDS, and advanced MDS (RAEB, RAEB-t, CMML) were eligible. For conditioning, patients received TBI (12 Gy) followed by intravenous high-dose cytarabine (3g/m^2 every 12 hours for 4 consecutive days). Recombinant G-CSF (lenograstim; 5 μg/kg/day) was administered intravenously by continuous infusion for 4 days, starting 12 hours before the first dose of Ara-C and continued until the last dose of Ara-C. For the prophylaxis of acute graft-versus-host disease, cyclosporine A or tacrolimus with or without short-term methotrexate was given. **Results:** Seventy-nine patients were evaluable, with a median age of 39 years (range, 15–58 years). Their diagnoses were AML in 57, AML from MDS in 11, and MDS in 11 (RAEB in 8, RAEB-t in 1, CMML in 1). Five-year overall survival (OS), disease-free survival (DFS), and relapse rate (RR) were 79.1%, 76.3%, and 15.2% in AML in remission, and 41.9%, 34.4%, and 46.4% in AML not in remission. These rates were 43.6%, 43.6%, and 0% in AML from MDS and 71.6%, 72.7%, and 10% in advanced MDS. For AML in remission, no risk factor affecting OS and RR was observed, whereas both high-risk karyotype abnormality and high numbers of blasts in the peripheral blood negatively affected OS, and these 2 factors and absence of chronic GVHD significantly increased RR in AML not in remission. **Conclusions:** These data suggest that TBI and G-CSF combined high-dose cytarabine is a highly effective regimen for allo-HSCT for AML in remission, AML evolving from MDS, and advanced MDS with a notably low incidence of relapse.

LYMPHOMA/MULTIPLE MYELOMA

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THE USE OF AUTOLOGOUS LMP2-SPECIFIC CYTOTOXIC T LYMPHOCYTES FOR THE TREATMENT OF RELAPSED EBV+ HODGKIN'S DISEASE AND NON-HODGKIN'S LYMPHOMA

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EBV-associated Hodgkin's disease (HD) and some cases of non-Hodgkins lymphoma (NHL) show type II latency expressing the subdominant EBV antigens EBNA1, LMP1, and LMP2, which may serve as targets for immunotherapy approaches. In previous studies, we used polyclonal EBV-specific CTL in patients with relapsed EBV+ HD and saw 2 complete and 1 partial responses in 11 patients. Analyses of EBV-CTL lines showed that small populations of T cells reactive against the tumor-associated antigen LMP2 were present in most of the infused lines, with some expansion in the peripheral blood after infusion. We therefore hypothesized that CTL specifically targeting LMP2 might have greater efficacy in these patients. LMP2-CTLs were generated from 10 patients using dendritic cells and lymphoblastoid cell lines (LCLs) that had been genetically modified to overexpress LMP2 by transduction with an Ad5f35LMP2 vector. Polyclonal LMP2-CTL lines recognized 2–6 (median, 4) LMP2 epitopes, as determined using overlapping LMP2 peptide pools in ELISPOT assays. A mean of 22.8% (range, 5%-42.1%) of CD8+ T cells bound HLA-restricted LMP2 tetramers, compared with a mean of 0.11% (range, 0.01%-0.24%) of LMP2-tetramer positive CD8+ T cells found in CTLs generated with genetically unmodified LCLs from the same patients. So far, 6 patients have been treated with 2 doses of 2×10^7 CTL/m^2 2 weeks apart. No immediate toxicity was observed. In patients with identified LMP2 epitopes, measurement of IFNγ secretion by CD8+ T cells after stimulation with appropriate LMP2 peptides in ELISPOT assays showed a 4- to 25-fold increase in spot-forming cells after infusions. In contrast, frequencies of CMV and superantigen-specific T cells did not increase. Four patients without radiologic evidence of disease who received CTL as adjuvant therapy post-SCT or chemotherapy remain well up to 12 months post-CTL. Two patients with measurable disease at the time of CTL infusion had stable disease 8 weeks post-CTL. They received 2 further doses of LMP2-CTL. One patient continues with stable disease, and the other patient had a complete radiologic response. This patient had a supraclavicular lymph node resection that showed selective accumulation of LMP2 tetramer-positive cells (0.3%, compared to 0.01% in the peripheral blood) with few residual tumor cells. Immunotherapy with autologous LMP2-CTL is therefore well tolerated in patients with relapsed EBV+ HD/NHL, and infused LMP2-CTL cells can localize to the tumor and induce a clinical response.

STEM CELL BIOLOGY

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REPAIR OF CROHN'S DISEASE WITH EMBRYONIC STEM CELLS

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The primary objective of this work is to determine differentiation and repair potential of murine embryonic stem cells (ES) in murine Crohn's disease (CD) model. **Methods:** Colitis was induced in IL10−/− knock-out mice using piroxicam. The colitis in this model is patchy and progressive and leads to death unless rescue therapy is provided. Enhanced yellow fluorescent protein (EYFP)

marked murine ES cells (R1/129) and ES-CCE without marker fluorescence protein in vitro differentiation induced by TGF-β (2 ng/ml), EGF (25 ng/ml), and β-FGF (100 ng/ml) for 7 days. IL10−/− mice were injected with predifferentiated ES-YFP cells and killed at 2 months postinjection. Serial frozen sections of colon, small intestine, and thymus were made, and EYFP staining was double detected using a confocol microscope and determined by immunohistochemistry using anti-GFP antibody. **Results:** Histopathologic analysis demonstrated improvement in colon tissue. Fluorescent and confocal microscopy demonstrated presence of the ES-EYFP cells in the colon, small intestine, liver, and thymus tissues. Anti-GFP staining was also detected in these tissues. The EYFP signal was not detected in sham and control IL10−/− knock-out mice. Immune studies by ELISA demonstrated a TH1→TH2 shift suggesting immune recovery. **Conclusion:** Our results suggest that the study of differentiation and repair potential of embryonic stem cells in pathological Crohn's disease model may lead to an alternative therapeutic potential for the treatment and prevention of inflammatory bowel disease.

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HUMAN HEMATOPOIETIC STEM CELLS CAN BE EXPANDED EX VIVO USING RECOMBINANT TAT-HOXB4 PROTEIN

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In mouse bone marrow transplantation models, engineered overexpression of HOXB4 has been one of the most potent stimulator of hematopoietic stem cell (HSC) expansion identified to date. The addition of soluble recombinant TAT-HOXB4 protein was also recently reported to enable rapid in vitro expansion of murine HSCs that retain their in vivo proliferation and differentiation capacity. However, the expansion potential of TAT-HOXB4 has always been tested on murine cells, and the capacity of this recombinant protein to expand human hematopoietic stem cells has remained hypothetical. To determine the ability of recombinant TAT-HOXB4 protein to promote human HSC expansion, we performed a series of experiments using CD34+ populations isolated from healthy volunteers. The CD34+ cell populations were cultured for 4 days in ex vivo 15 medium supplemented with stem cell factor (300 ng/mL) and G-CSF (50 ng/mL) in the presence or absence of TAT-HOXB4 protein (50 nmol/L). In response to TAT-HOXB4, total numbers of mononuclear cells demonstrated a modest but distinct 2-fold increase compared with controls. TAT-HOXB4 treatment had a major proliferation enhancing effect on the more primitive cell populations such as CFU-GEMM, BFU-E, and BFU-Meg, which numbers increased 26.5- ± 1.4-fold (mean ± SD), 2.2- ± 0.7-fold, and 2.1- ± 0.2-fold, respectively, over their input values, and 19.1- ± 1.3-fold, 2.7- ± 0.7-fold, and 31- ± 3.4-fold, respectively, compared with growth factor-only controls. In response to TAT-HOXB4, the total numbers of CD34+CD38−Lin− cells increased 2.1- ± 0.7-fold above their starting numbers, compared with a 1.5- ± 0.5-fold loss of this population in control cultures. HSC numbers were enumerated at the beginning, and after a 4-day TAT-HOXB4 treatment period under the same conditions using a NOD/SCID repopulation assay. In response to 50 nM TAT-HOXB4, NOD/SCID repopulating cell (SRC) numbers increased 2-fold over their input values, compared with a 9-fold loss in control cultures without TAT-HOXB4. These results show that recombinant TAT-HOXB4 protein, even when combined with a limited number of hematopoietic growth factors, has the capacity to rapidly induce ex vivo expansion of human stem cell and progenitor cell populations. They also provide the first evidence that TAT-HOXB4 protein could be used to expand human HSCs ex vivo before autologous or allogeneic stem cell transplantation.

SUPPORTIVE CARE

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ABDOMINAL CT IN THE CLINICAL EVALUATION OF ACUTE GRAFT-VERSUS-HOST DISEASE (GVHD) OF THE GASTROINTESTINAL TRACT: DIFFUSE SMALL INTESTINE INVOLVEMENT IS ASSOCIATED WITH POOR OUTCOME

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Acute GVHD is the major treatment-related complication after allogeneic stem-cell transplantation. Acute GVHD of GI tract is a clinical diagnosis supported by histopathologic findings. The volume of diarrhea determines disease stage and prognosis; however, its assessment is inconvenient and inaccurate. Moreover, patients may present with atypical symptoms, whereas GI tract biopsies may be falsely negative due to patchy involvement or may be contraindicated in patients with severe thrombocytopenia. This study was designed to determine the CT features associated with acute GI GVHD and to evaluate its role in assessment of severity and prognosis. A total of 34 consecutive patients with symptoms suggestive of acute GI GVHD were evaluated by abdominal CT. Of these 34 patients, 18 had clinical stage I-II and 14 had clinical stage III-IV by the Glucksberg criteria; 32 patients had pathological findings on CT. The most consistent finding was thickening of the bowel wall, which was limited to the small bowel (n = 11) or large bowel (n = 5) or involved both segments (n = 16). Involvement was diffuse or segmental. Other manifestations included intestinal dilatation (n = 11), mucosal enhancement (n = 3), and gastric wall thickening (n = 7). Extraintestinal findings included mesenteric stranding (n = 22), ascites (n = 13), and biliary system abnormalities (n = 11). Ten patients had urinary excretion of orally administered gastrografin, which is not normally absorbed by an intact intestine. Diffuse thickening of the small bowel wall and/or any involvement of the large intestine were patterns associated with severe clinical presentation. Eleven and 14 of the 16 patients with clinical stage III-IV had these patterns, compared to 5 and 7 of the 18 patients with stage I-II, respectively ($P = .03$). Overall, 19 patients responded to immunesuppressive therapy. Nine patients are alive; 25 died, 13 of complications directly related to GVHD. Diffuse small bowel disease was associated with poor prognosis. Only 6 of 16 patients showing this pattern responded to therapy, compared with 13 of 18 patients without this pattern ($P = .05$). The cumulative incidence of GVHD-related death was 56% and 25%, respectively ($P = .05$). Overall survival was not significantly different in the 2 groups. In conclusion, abdominal CT may have an important role in the diagnosis of GVHD in atypical presentations and in the prognostic evaluation. Diffuse small intestinal disease is associated with poor response to therapy and GVHD-related mortality. CT findings may direct the clinician in determining the therapeutic approach.

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SAFETY AND TOLERABILITY OF ANTIBACTERIAL PROPHYLAXIS DURING THE POSTENGRAFTMENT PHASE AFTER MYELOABLATIVE HEMATOPOIETIC STEM CELL TRANSPLANTATION

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Background: Bacterial infections are frequent complications of the postengraftment phase after ablative allogeneic HSCT. This pilot study investigated the safety and tolerability of administering antibiotic (ABX) prophylaxis from when patients tolerated PO medications after engraftment (ANC > 1500/mm^3 for 2 days) through day +100. **Methods:** Twenty-three engrafted allogeneic BMT patients at OHSU, received moxifloxacin (MOXI) 400 mg po daily. Drug was started on discontinuation of any ABX used for the treatment of pre-enrollment bacterial infection or neutropenic fever. Rates of bacterial infection, bacteremia, and colonization were recorded. Comparisons were made with a cohort of 60 con-