101 BONE MARROW TRANSPLANTATION IN CONGENITAL IMMUNODEFICENCY STATES - ULM EXPERIENCE

Wolfgang Hartmann and Wilhelm Friedrich spn. by Enno Kleihauer - Department of Pediatrics, University of Ulm, D7900 Ulm, Germany

Between 1983 and 1990 we treated 53 patients for congenital combined immunodeficency by bone marrow transplantation. 50 patients had SCID including 5 with ADA deficency and 3 with Omenn syndrome, 2 were treated for bare lymphocyte syndrome, two siblings suffered from a functional T-cell defect. The overall survival was 57% with a mean follow up of 4.2 years. After HLA - identical transplantation 7/9 (77.7%) and after haploidentical transplantation 29/45 (64.4%) survived. After haploidentical transplantation with T - cell depletion and without pretransplant conditioning, GvHD occured in 1/23, after pretreatment with ATG in 1/5 and after conditioning with busulfan and cyclophosphamide in 7/17 pts.. There was a slightly higher early mortality in patients with conditioning, but no difference in long time survival. However conditioned patients had a benefit in respect to B - cell reconstitution. The occurence of maternally derived lymphocytes (40% of pts.) had no influence on survival. The outcome of all these patients seems mainly influenced by their clinical state prior to BMT.

102 BONE MARROW TRANSPLANTATION FOR CHRONIC GRANULOMATOUS DISEASE X/b+.

M.M. Manzionna, P. Di Bartolomeo #, G. Di Girolamo #, D. De Mattia, G. Torlontano #, F. Schettini.
Dipartimento di Biomedicina dell'Età Evolutiva - Clinica Pediatrica 1 Università di Bari and # Divisione di Ematologia, Cattedra di Ematologia, Ospedale Civile di Pescara, Università di Chieti - Italy.

An 8 year old boy affected by a rare form of X-linked Chronic Granulomatous Disease (CGD) and present cytochrome b underwent successful bone marrow transplantation on August 1989 in Pescara (3.4 x10⁸ nucleated cells/Kg) from his 6 year old histocompatible sister (HLA-identical and MLC negative) after a preconditioning regimen of Busulfan (3.25 mg/kg/day for 4 days) followed by Cyclophosphamide (50 mg/kg/day for 4 days). The patient was protected by LAF isolator and by our decontamination protocol. Cyclosporine was administered for 12 months and oral Acyclovir for 270 days. The clinical course was uncomplicated and the boy was engrafted promptly with PMN counts normalizing on day +20 and the complete reversal of neutrophil function defect. Cytogenetic showed complete engraftment of donor origin at 4 months after BMT. Eighteen months after BMT the boy still continues to be free of infections. In fact NBT was already positive at the nineth month for Zymosan (92%) and for PMA (91%). The O_2^- generation with PMA was 8.88 nmol/min/10⁶ cells and with fMLA was 13.01 nmol/min/10⁶ cells. Cytochrome b spectra showed peaks at 428, 530, 558 nm (7.81 pmol/10⁶ cells) and all his neutrophils stained for cytochrome b on the cell surface by using monoclonal antibodies 7D5 (98%). Earlier attempts of BMT for CGD by bone marrow from related or unrelated donors have failed either because of slow loss of the graft and gradual deterioration in neutrophil function or because of lethal infections and/or GvH disease. We conclude that as well as in CGD X/b-, previously described by us (Bone Marrow Transplant 4, 695, 1989), a complete neutrophil engraftment can be achieved also in this rare form CGD X/b+.

Supported by a grant from M.U.R.S.T..

103 CLINICAL AND GENETIC HETEROGENEITY OF RECESSIVE FORM OF HEREDITARY SPHEROCYTOSIS.

A.Iolascon,S.Perrotta,E.Miraglia del Giudice,L.Pinto and S.Cutillo
Department of Pediatrics - University of Naples (Italy)

α-spectrin synthesis exceeds ß by a factor four (Hanspal,1987). This statement suggests that α-spectrin deficiency would be evident only in the homozygous state. Agre (1982,85) described some cases of recessively inherited HS characterized by severe spectrin deficiency and life-threatening hemolysis. Marchesi (1989) founded in 9 out of 20 of these kindreds an acidic shift of αII domain. More recently Forget (1990) analyzing the genomic DNA of these subjects demonstrated an Ala-Asp (GAT-GCT) substitution at position 972 of α-sp. In order to assess clinical findings of rHS and verify the linkage with α-spectrin gene we studied 11 unrelated kindreds of italian extraction, characterized by affected propositus with hematologically and biochemically normal parents. Two of these had severe HS, whereas the remaining have a clinical picture of typical HS. We examined 3 polymorphic sites (XbaI,PvuII,MspI) founded in linkage disequilibrium with α-sp gene (Hoffman,1987)(3021-E1 cDNA probe was given by courtesy of B.G.Forget). In the informative families rHS is associated with the following haplotype X(+),P(-),M(+) in three chromosomes and with ++-, -+- and --- in three others. The haplotype +-+ is the most frequent in normal population of our country. This finding suggests the heterogeneity of molecular defect at the α-gene level. Furthermore in two cases the haplotype assignement allows to the exclusion of α-sp gene involvement. This suggests that the α-spectrin gene abnormalities couldn't cause all rHS.

104 NEWBORN ERYTHROCYTES ARE MORE RESISTENT THAN ADULT TO FREE RADICAL-MEDIATED DAMAGE CAUSED BY LIPID EMULSION.

S Kljuchnikov, O Pitkänen, K Raivio and S Andersson - 4th Children's Hospital, Univ. of Moscow, USSR; and Children's Hospital, Univ. of Helsinki, Helsinki, Finland.

Intravenous lipid emulsions containing polyunsaturated fat are susceptibe to lipid peroxidation, and may trigger cell damage. We incubated adult (A) or term neonatal (N) erythrocytes with 0.44 % Intralipid® (IL) in phosphate-buffered saline for 17 h. Hemolysis was 40.5±2.1% in A and 22.4±1.3% in N ($p<0.001$). MCV increased by 69.7% in A and 48.5% in N ($p<0.001$). Glutathione (GSH) decreased in A from 63.6±4.7 to 21.6±4.5 (66.0 %) and in N from 61.1±5.9 to 25.7±5.0 mg/dl (57.9%)(changes $p<0.001$). However, malondialdehyde and conjugated dienes were higher in N (both $p<0.001$). Desferoxiamine or sodium etidronate inhibited the effects of IL in both A and N. We conclude that despite higher lipid peroxidation in the membrane, the newborn red cell is more resistant to free radical damage than that of the adult.

105 HIGH DOSES OF HUMAN RECOMBINANT ERYTHROPOIETIN (rHuEPO) ARE EFFECTIVE IN THE TREATMENT OF ANEMIA OF PREMATURITY (AP)

Virgilio Carnielli, Rosalia Da Riol, Graziella Zacchello, Felice Cantarutti, Giovanni Montini - Department of Pediatrics, University of Padova, Via Giustiniani 3, Padova, Italy.

rHuEPO has been preaviously used for the treatment of AP with uncertain results; dosages between 40 and 300 IU/Kg/week have generally failed to reduce the need for blood transfusions in premature infants, although no major side-effects were shown. We performed a pilot study to demonstrate the safety and the efficacy of higher doses (120-300-600-1200 IU/kg/week) of rHuEPO administered to premature infants. Only doses of 1200 IU/Kg/week were shown effective to increase the reticulocyte count; we describe these data. Patients: Inclusion criteria were: gestational age (GAGE) less than 33 weeks and birth weight (BW) less than 1.75 Kg; admission to our neonatal intensive care unit. Group A: 8 premature infants (mean BW 1.38±0.26; GAGE 30.2±1.6) were given 1200 IU/Kg/week in 3 doses and 20 mg/Kg/week of iron. Group B: 11 premature infants (BW 1.34±0.26; GAGE 29.6±2.4) were used as controls. Results: no side-effects of the treatment, especially for blood pressure, white blood cell and platelet count, were shown. Main results are shown in the table.

	Number transfusions	RC transfused (ml/Kg)	Blood sampled (ml/Kg)	Mean Hct (%)	Mean retics (%)
EPO	1.0±1.7	16±29	20±23	48.1±7.0	4.6±0.3
CONTR.	3.1±2.2	48±34	27±19	41.9±3.6	1.6±0.4
p	0.019	0.023	0.21	0.048	0.0001

CONCLUSIONS: From these preliminary data high doses of rHuEPO and iron supplementation are effective in reducing the number of blood transfusions in premature infants.

106 EFFECT OF RECOMBINANT HUMAN ERYTHROPOIETIN (EPO) IN PREMATURE NEONATES: PRELIMINARY RESULTS OF A DOUBLE BLIND CONTROL TRIAL

Vasiliki Soubasi, George Kremenopoulos, Chaido Tsantali, Sotiria Mastrogianni, George Kyriakides, Dimitris Tsakiris, (spn.by Dimitris Anagnostakis) - Department of Neonatology, University of Thessaloniki, 546 22 Thessaloniki, Greece

We investigated whether preventative treatment with higher doses of EPO in the premature neonates is safe and reduces the need for transfusion. 20 infants were randomized to an EPO (n:12,BW:1176 ± 234g,GA:28,2 ± 2,2w) and a control group (n: 8, BW: 1232 ± 239g, GA: 29,3 ± 1,6w). EPO 150U/kg was given s.c. every 3rd day for 6w early from the 1st w of life and iron 3mg/kg/d from day 14.The 2 groups were similar (BW,GA, clinical status). During the 6 w of therapy with EPO, there were no differences between control and EPO groups in Ht, Hb, total leukocytes, neutrofils, HbF, transfusion requirements,mortality or morbitidy. Reticulocytes and thrombocytes were increased from the 3rd w and serum ferritin decreased from the 6th w of EPO treatment compared to control group (table).

		pre-EPO	2nd w	4th w	6th w	
Reticulocytes	EPO	12 ± 4,8	1,37 ± 1	3,3 ± 1,7	5,6 ± 1,6	$p<0,001$
	control	8,9 ± 4	1,36 ± 1,3	1,8 ± 1,9	1,25 ± 1,1	
Thrombocytes	EPO	216 ± 50	325 ± 120	369 ± 69	463 ± 97	$p<0.05$
	control	269 ± 69	262 ± 166	213 ± 116	290 ± 143	
Ferritin	EPO	196 ± 221	273 ± 80	284 ± 147	200 ± 193	$p<0.05$
	control	210 ± 67	181 ± 85	335 ± 122	547 ± 279	

In conclusion EPO administration of 300 U/kg/w appears to be safe. However, although the need for transfusion was not reduced the reticulocyte production was significantly increased.