European Journal of Cancer Vol. 31A, No. 11, pp. 1883–1884, 1995.

0959–8049/95 $9.50 + 0.00

Letters

0959-8049(95)00363-0

Human Stem Cell Factor Protects CD34 Positive Human Myeloid Leukaemia Cells from Chemotherapy-induced Apoptosis

H.T. Hassan and M. Freund

THE HUMAN CD34 haemopoietic cell surface antigen is a highly glycosylated 115 kDa type I integral membrane protein expressed in only 1–4% of normal adult bone marrow cells, in mainly progenitor and precursor cells [1]. The human CD34 gene has been localised to band 23 on the long arm of chromosome 1, a region containing a cluster of genes encoding haemopoietic regulatory and signalling molecules, and contains eight exons spanning 26 kb of DNA [2]. CD34 is also expressed in acute myeloid leukaemia (AML) blast cells from 40–60% of adult patients with all FAB (French–American–British) subtypes, except acute premyelocytic leukaemia (APL) (M3) [3–6], myelodysplastic cells [7], chronic myeloid leukaemia cells especially from patients with accelerated and blastic phases [8], B-acute lymphatic leukaemia cells [9] and Philadelphia positive acute lymphatic leukaemia cells [10]. Adult patients with CD34 positive (CD34+) AML cells have been shown to have low total leucocyte count, a previous history of myelodysplasia and/or chemoradiotherapy, a high frequency of chromosomal abnormalities involving chromosomes 5 or 7 [3–5] and translocation t(8;21)(q22;q22) [6], low myeloperoxidase positivity [11], low CD15 expression [3] and significantly low remission and survival rates [3, 4, 11]. Also, in patients with myelodysplasia, CD34 expression has been associated with transformation to AML with poor survival [7]. Moreover, *in vivo* transplantation studies have shown the expansion of AML cells in SCID mice to be restricted to the CD34+ population [12]. Since human recombinant stem cell factor, whose receptor is expressed in most AML cells, has been shown to stimulate the proliferation of mainly CD34+ AML cells [13], we investigated whether the poor response of patients with CD34+ myeloid leukaemia cells to chemotherapy could be due to the effect of stem cell factor.

The establishment of several human myeloid leukaemia cell lines in culture has proven to be a very important tool for studying myeloid cell proliferation and differentiation [14]. The regulatory signals required to direct haemopoietic progenitor cells toward self-renewal and differentiation along specific lineages require the development of culture systems devoid of uncharacterised components. Traditional methodology, requiring the use of either xenogeneic serum components and/or conditioned medium supplements to support these cells, has the potential to be a confounding variable to experimental results [14]. In our laboratory, a novel CD34+ human myeloid leukaemia cell line, MHH225, has been established from the bone marrow of a 60-year-old patient with AML(M7), and grown in serum-free RPMI1640 culture medium [15]. The MHH225 cell line has several chromosomal aberrations including deletion in chromosome 7, t(9;12)(q10;q10), der(21) and add(11q) [15]. The MHH225 cell line provides a unique model for studying CD34+ human myeloid leukaemia cells, and might contribute to the improvement of our knowledge of myeloid leukaemogenesis and its positive and negative regulation by human cytokines in serum-free culture. The effect of stem cell factor on apoptosis, induced by cytarabine, daunorubicin or carboplatin, three commonly used chemotherapeutic drugs in the treatment of AML, was examined in the MHH225 cells. The MHH225 cells were cultured at a concentration of 100 000 cells per ml on day 0 in 24-well tissue culture plates at 37°C in a humidified incubator for 96 h under CO_2. Stem cell factor was added at a concentration of 200 ng/ml, which has been found to be the optimum concentration for the proliferation of these CD34+ leukaemia cells. Apoptosis was determined after 96 h by measuring the expression of APO-1 (CD95) antigen in treated MHH225 cells, in both the presence and absence of human recombinant stem cell factor. Table 1 shows the significant inhibitory effect on chemotherapy-induced apoptosis in the presence of stem cell factor. Stem cell factor significantly reduced the induced apoptosis by more than 50% in all CD34+ human leukaemia cells treated by any of the three chemotherapeutic drugs. The present results suggest that the poor response of patients with CD34 positive leukaemia cells could be, at least, partially due to less chemotherapy-induced apoptosis as a result of protection by stem cell factor.

Table 1. Effect of stem cell factor on chemotherapy-induced apoptosis in CD34+ leukaemia cells

Chemotherapeutic drug (concentration)	Percentage of apoptosis in human CD34+ myeloid leukaemia cells		
	Without stem cell factor	With stem cell factor	*P*
Cytarabine (0.5 μg/ml)	79.1 ± 6.0	37.4 ± 5.1	<0.01
Daunorubicin (0.05 μg/ml)	86.2 ± 8.1	42.8 ± 4.6	<0.01
Carboplatin (0.05 μg/ml)	93.1 ± 5.7	38.0 ± 3.2	<0.01

Values represent mean ± S.D. of nine determinations from three separate experiments.

Correspondence to Dr H.T. Hassan.
Both authors are at the Department of Haematology and Oncology, University of Hannover Medical School, Konstanty-Gutschow-Straße 8, 30625 Hannover, Germany.
Received 28 Nov. 1994; accepted 30 Jun. 1995.

1. Silvestri F, Banavali S, Baccarani M, Preisler HD. The CD34 haemopoietic progenitor cell associated antigen: biology and clinical applications. *Haematologica* 1992, 77, 265–273.
2. Satterthwaite AB, Burn TC, Le Baeu MM, Tenen DG. Structure of the gene encoding CD34, a human haemopoietic stem cell antigen. *Genomics* 1992, 12, 788–794.
3. Borowitz MJ, Gockerman JP, Moore JO, *et al.* Clinicopathologic

and cytogenetic features of CD34 (My10) positive acute nonlymphocytic leukemia. *Am J Clin Pathol* 1989, **91**, 265–270.
4. Geller RB, Zahurak M, Hurwitz CA, *et al.* Prognostic significance of immunophenotype in adults with AML: the significance of the stem cell glycoprotein CD34 (My10). *Br J Haematol* 1990, **76**, 340–344.
5. Lee EJ, Yang J, Leavitt RD, *et al.* The significance of CD34 and TdT determinations in patients with untreated *de novo* acute myeloid leukaemia. *Leukemia* 1992, **6**, 1203–1209.
6. Kita K, Nakase K, Miwa M, *et al.* Phenotypical characteristics of acute myelogenous leukaemia associated with the t(8;21)(q22;q22) chromosomal abnormality: frequent expression of immature B-cell antigen CD19 together with stem cell antigen CD34. *Blood* 1992, **80**, 470–477.
7. Sullivan SA, Marsden KA, Lowenthal RM, *et al.* Circulating CD34+ cells: an adverse prognostic factor in myelodysplastic syndromes. *Am J Hematol* 1992, **39**, 96–101.
8. Banavali S, Silvestri F, Hulette B, *et al.* Expression of haemopoietic progenitor cell associated antigen CD34 in CML. *Leukemia Res* 1991, **15**, 603–608.
9. Schmitt C, Eaves CJ, Landsdorf PM, *et al.* Expression of CD34 on human B-cell precursors. *Clin Exp Immunol* 1991, **85**, 168–173.
10. Azumi E, Umemoto M, Kubo M, *et al.* CD34 antigen expression in children with Philadelphia chromosome-positive ALL. *Cancer* 1991, **67**, 1565–1569.
11. Myint H, Lucie NP. The prognosis of the CD34 antigen in AML. *Leukemia Lymphoma* 1992, **7**, 425–429.
12. Lapidot T, Sirard G, Vormoor J, *et al.* A cell initiating human AML after transplantation into SCID mice. *Nature* 1994, **367**, 645.
13. Pietsch T, Kyas U, Ludwig W-D, Welte K. Mitogenic effect of human stem cell factor alone and in combination with G-CSF, GM-CSF, and IL-3 on fresh leukemic blast cells from patients with stem cell factor receptor-positive AML. In Freund M, *et al*, eds. *Cytokines in Hemopoiesis, Oncology and Aids II*. Berlin, Springer Verlag, 1992, 171–178.
14. Hassan HT. Synergism between differentiation inducing agents: preclinical and clinical studies in myeloid leukaemia. In Rifkind R, ed. *The Pharmacology of Cell Differentiation*. Amsterdam, Elsevier, 1993, 277–294.
15. Hassan HT, Grell S, Borrmann-Danso U, Freund M. Interleukin-4 inhibits proliferation of human leukaemic megakaryoblastic cells. *Hematol Oncol* 1994, **12**, 61–66.

European Journal of Cancer Vol. 31A, No. 11, pp. 1884–1885, 1995.

0959–8049/95 $9.50 + 0.00

0959-8049(95)00309-6

Profile of T-cell Lymphomas in Ibadan, Nigeria

J.O. Thomas

MALIGNANT LYMPHOMA (ML) is one of the commonest malignancies in Ibadan, Nigeria. Generally, B-cell lymphomas are more frequent, except in HTLV-1 endemic zones of South East Asia, where T-cell lymphomas make up 40–80% of the non-Hodgkin's lymphomas (NHLs) [1–2]. Cutaneous T-cell lymphomas and *Mycosis fungoides* constitute the majority of the post-thymic T-cell neoplasms in Western countries, whereas node-based peripheral T-cell lymphomas are predominant in other regions of the world [3, 4]. Post-thymic T-cell malignancies, in contrast to T-lymphoblastic lymphomas, show significant geographical, clinicopathological and prognostic diversity [5, 6]. Recently, previously ill-defined lymphoproliferative disorders, such as lymphomatoid granulomatosis, lymphomatoid papulosis, polymorphic reticulosis and mid-line malignant reticulosis, have been established as clonal expansions of post-thymic T-cells. The spectrum of post-thymic T-cell neoplasms has, therefore, widened. In view of this, the clinicopathological features and geographical distribution of T-cell neoplasms must be thoroughly investigated.

In Ibadan, Nigeria, although NHLs are common, very few data are available on the histopathological profile of T-cell

Table 1. Characteristics of 12 cases of T-cell lymphomas seen within a 3 year period in Ibadan, Nigeria

Case no.	Age	Sex	Presentation	Histological type Updated Kiel's classification
1	10	M	Mediastinal mass with pleural infiltration and pleural effusion	Lymphoblastic
2	62	M	Generalised lymphadenopathy, marrow failure and wasting	Small cell, lymphocytic
3	40	M	Generalised lymphadenopathy	Small cell, lymphocytic
4	19	M	Multiple skin nodules involving face, limbs and trunk	*Mycosis fungoides*
5	32	F	Breast subcutaneous, ulcerative, nodule associated, right axillary lymphadenopathy	Angiocentric pleomorphic, small cell type
6	Adult†	M	Lymph node	Pleomorphic mixed medium/large cell type
7	20	M	Cervical lymph node enlargement	Pleomorphic large cell type
8	32	M	Cervical lymph-adenopathy	Pleomorphic, small cell type
9	30	F	Extradural cord compression; nodules on the scalp. Right axillary lymph-adenopathy	Lymphoblastic type
10	68	M	Right axillary lymphadenopathy. Right submandibular mass extending to right posterior triangle of the neck. Severe generalised pruritic papular rash	AILD type*
11	50	M	Large cervical, preclavicular and axillary nodes	Pleomorphic medium and large cell type
12	60	M	Multiple discrete cervical axillary and lymphadenopathy	Small cell, lymphocytic

*AILD, angio-immunoblastic lymphadenopathy-like; † Age unknown.

Correspondence to J.O. Thomas at the Department of Pathology, University College Hospital, Ibadan, Nigeria.
Received 20 Dec. 1994; accepted 31 May 1995.