Haematology and Blood Transfusion Vol. 29
Modern Trends in Human Leukemia VI
Edited by Neth, Gallo, Greaves, Janka

Evidence of Sternberg-Reed Cells Being Derived from Activated Lymphocytes

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A. Introduction

The nature and origin of Sternberg-Reed (SR) cells remains obscure despite numerous studies of this topic. In recent experiments we raised monoclonal antibodies against the Hodgkin's disease-derived cell line L428, with the aim of producing reagents specific for SR cells. Three monoclonal antibodies (designated Ki-1, Ki-24, and Ki-27; Stein et al. 1982, 1983) were obtained in the course of this study, and their reactivities are detailed in Table 1.

Monoclonal antibody Ki-1 differed from the other two reagents in that it detected a subpopulation of large cells, preferentially distributed around the margin of B cell follicles (Stein et al. 1982). This population of cells does not correspond to any cell type previously recognized using other monoclonal antibodies. Since the earliest site of lymph node involvement in Hodgkin's disease is the perifollicular region (as has been recognized previously from conventional histology and can also be shown by immunolabelling with Ki-1 antibody), we proposed that this population of Ki-positive cells in normal lymphoid tissue may represent the physiological counterpart of SR cells. However, it is not clear whether the subpopulation represents a novel cell lineage or simply a differentiation stage within one or more cell lineages. To study this question further, the expression of Ki-1 antigen, and also of the antigens detected by antibodies Ki-24 and Ki-27, was investigated in a wide range of lymphoid tissue samples, including fetal material and peripheral blood lymphocytes stimulated by a variety of mitogenic agents.

B. Material and Methods

I. Cases

Fresh unfixed biopsies of lymphoma and other tissues were obtained from the Hospital of the University of Kiel Medical School and the John Radcliffe Hospital, Oxford, England.

II. Phytohemagglutinin (PHA)-Stimulated and Virus-Transformed Human Peripheral Blood Cells

Cells (1×10^6 per ml) were cultured in the presence of 0.25 μg PHA/ml. Cells were harvested after 72 h and centrifuged onto glass slides. HTLV II transformed peripheral blood cells were a gift from Dr. I. Y. Chen. These cells had been transformed by co-cultivation with irradiated Mo-T cells, as described elsewhere (Chen et al. 1983). EBV transformed blood cells were prepared by in vitro infection as recently described (Moss et al. 1978).

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Table 1. Reactivity of three monoclonal antibodies (Ki-1, Ki-24 and Ki-27) raised against the Hodgkin's disease-derived cell line L 428

Antibody	Normal lymphoid tissue	L 428 cell line	Sternberg-Reed cells
Ki-1	Scattered large perifollicular cells	+	+
Ki-24	None, or a very few scattered cells	+	+
Ki-27	Vessels, sinus lining cells, and no or only occasional lymphoid cells	+	+

III. Immunolabelling of Sections and Cytocentrifuged Cells

Immunolabelling was performed using either the alkaline phosphatase: antialkaline phosphatase (APAAP) method (Cordell et al. 1984) or the three-stage immunoperoxidase method (Stein et al. 1982).

C. Results and Discussion

The results obtained are summarized in Tables 2–5. The non-reactivity of Ki-1 antibody with fetal liver, fetal and adult bone marrow and fetal and postnatal thymus makes it unlikely that Ki-1-positive cells found in normal tissue represent precursors of T, B or monocyte/macrophage origin. This conclusion is supported by the fact that the other two SR-cell-associated antigens (Ki-24 and Ki-27) were also absent from these tissues.

Each of the three SR-cell-associated antigens (Ki-1, Ki-24 and Ki-27) could be induced on peripheral blood lymphocytes by exposure to PHA or by infection with HTLV or EBV (Table 3), although the proportion of positive cells differed for each antigen. Expression of each antigen was consistently accompanied by the appearance of the activation-associated antigens, i.e. the IL2 receptor (detected by anti-Tac, TÜ69 and ACT1).

These findings suggest the possibility that SR cells may represent activated T lymphocytes. To explore this possibility we analysed SR cells in situ by staining tissue sections for T cell and B cell antigens and activation-associated antigens using a newly developed highly sensitive immunalkaline phosphatase method (the APAAP technique). This analysis revealed that at least some of the SR cells in the majority of non-lymphocyte-predominant types of Hodgkin's disease express a variety of T cell antigens (T1, T3, T4 and T11). It was also possible to demonstrate strong staining for IL2 receptor in the majority of cases. However, a surprising finding was that a varying proportion of SR cells in many of these cases expressed B cell antigens, usually associated with T cell antigens but occasionally alone (Table 4).

These observations provide evidence that SR cells may indeed be activated T lymphocytes. However the expression of B cell antigens by SR cells in some cases requires explanation. One possible hypothesis is that the anti-B cell antibodies are only specific for this cell lineage when resting lymphoid cells are analysed, and this specificity may be lost when lymphocytes undergo transformation. Hence activated T cells may aberrantly express apparently B cell-specific markers.

The expression of B cell-associated antigens in some SR cells (and also the absence of T cell-associated antigens from cases of lymphocyte-predominant Hodgkin's disease) prompted us to investigate the possible B cell nature of SR cells in Hodgkin's disease further. For this purpose we analysed the expression of J chain, since this molecule has been shown to be a reliable marker for cells of the B lineage and numerous studies in the past have failed to demonstrate its expression in T lymphoid cells. As shown in Table 5 and Fig. 1, SR cells in all cases of

Table 2. Antibody Ki-1 reactivity in tissue containing myeloid and/or lymphoid precursor cells

Tissue	Ki-1 positive
Fetal tissue	
Liver (2)[a]	None
Bone marrow (2)	None
Thymus cortex (2)	None
Postnatal tissue	
Liver (6)	None
Bone marrow (6)	None
Thymus cortex	None

[a] Number of samples are given in parenthesis

Hodgkin's disease of nodular sclerosing, mixed cellularity and lymphocyte-depleted types were negative for J chain. However, SR cells in the majority of cases of lymphocyte-predominant disease of nodular subtype were J chain positive. This is in keeping with a previous report in the literature (Poppema 1980), in which J chain expression by SR cells was described in a single case of lymphocyte-predominant Hodgkin's disease.

Our working hypothesis (Table 6) based upon these results is that Hodgkin's disease represents the neoplastic proliferation of

Table 3. Antigen profile of peripheral blood lymphocytes following stimulation or transformation with PHA without or with IL2, HTLV or EBV

Stimulating transforming agent	Ki-27	Ki-24	Ki-1	T3	Slg	To15 B 4	Tac	TU69
None	0	0	0	82	12	15	0	0
PHA	3	6	15	93	10	14	97	98
PHA plus IL2	15	80	20	100	0	NA	100	100
HTLV	0	98	97	95	0	0	96	97
EBV	0	100	100	0	100	100	10	15

Table 4. Patterns of antibody reactivity of Sternberg-Reed cells of Hodgkin's disease established in 35 cases

Ki-1	HLA-DR	T3/T11	B4/To15	Tac	TU69
+	+	–	–	+/–	+/–
+	+	+	–	+/–	+/–
+	+	+	+	+/–	+/–
+	+	–	+	+/–	+/–

Table 5. Reactivity of Sternberg-Reed cells of Hodgkin's disease for J chain and granulocyte antigen detected by 3C4 or C3D-1

Histological type	No. of cases	J chain	Granulocyte antigen
Nodular sclerosis	9	0	9
Mixed cellularity	8	0	6
Lymphocyte depletion	6	0	4
Lymphocyte predominance (nodular subtype)	29	22	0
		0	5

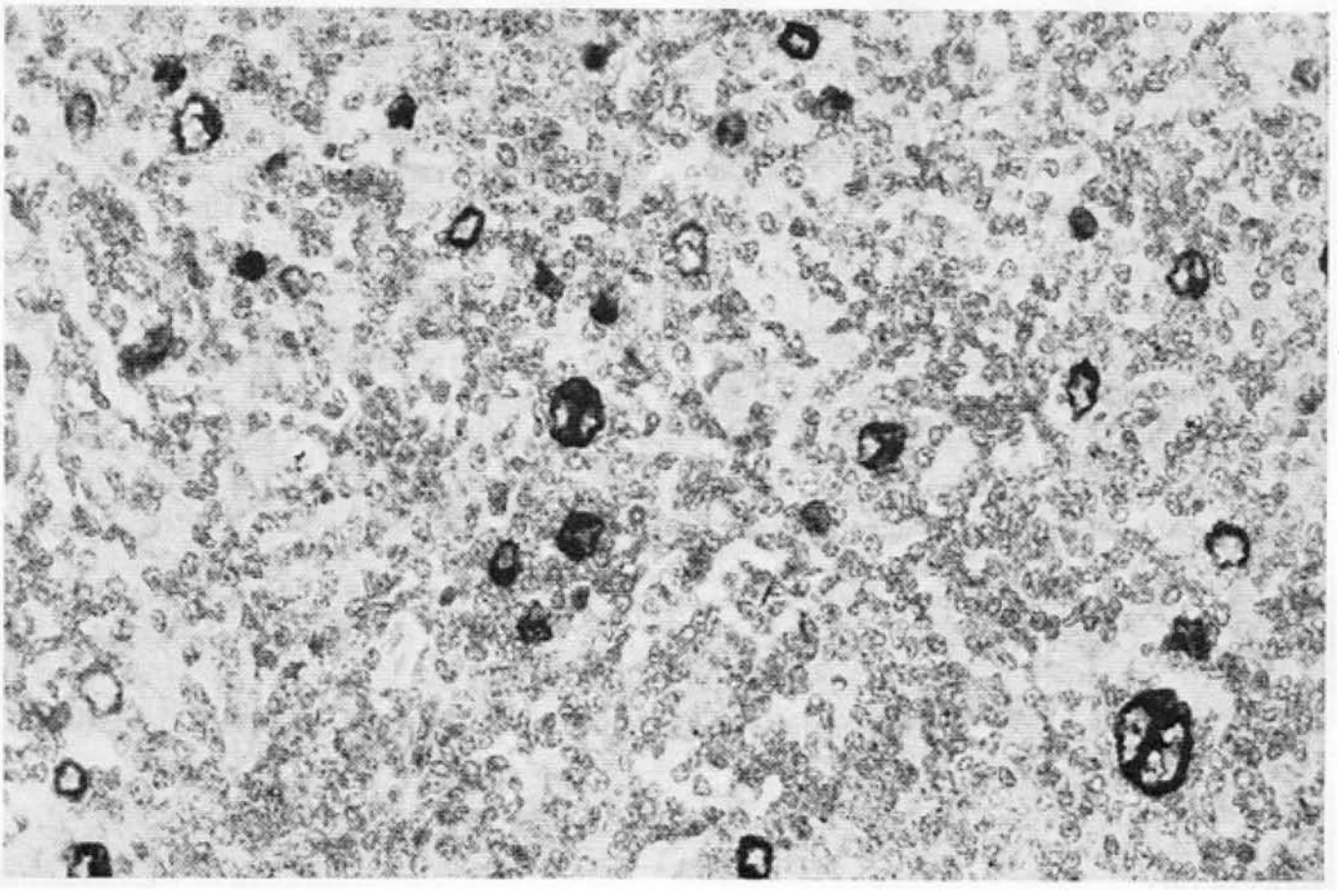

Fig. 1. Hodgkin's disease, lymphocyte predominance, nodular subtype immunostained with an anti-J-chain antiserum. The Sternberg-Reed cells are strongly positive (APAAP)

activated lymphoid cells, but that the histological type of the disease may be related to whether these cells are of T cell or B cell origin. Neoplasms of activated T cells show the histological appearances of nodular sclerosing, mixed cellularity and lymphocyte-depleted Hodgkin's disease. In contrast, the more rarely encountered neoplasms of activated B cell give rise to lymphocyte-predominant disease.

Table 6. Putative origin of Sternberg-Reed cells in Hodgkin's Disease

Origin	Histological appearance
Activated T cell	Nodular
	Mixed cellularity
	Lymphocyte depletion
Activated B cell	Lymphocyte predominance, nodular subtype (J chain positive)

References

Chen ISY, Quan SG, Golde DW (1983) Human T-cell leukemia virus type II transforms normal human lymphocytes. Proc Natl Acad Sci USA 80:7006–7009

Cordell JL, Falini B, Erber WN, Ghosh AK, Abdulaziz Z, MacDonald ST, Pulford KAF, Stein H, Mason DY (1984) Immunoenzymatic labeling of monoclonal antibodies using immune complexes of alkaline phosphatase and monoclonal anti-alkaline phosphatase (APAAP complexes). J Histochem Cytochem 32:219–229

Moss DJ, Rickinson AB, Pope JH (1978) Long-term T cell-mediated immunity to Epstein-Barr virus in man. I: Complete regression of virus transformation in culture of seropositive donor leukocytes. Int J Cancer 22:662–668

Poppema S (1980) The diversity of immunohistological staining pattern of Sternberg-Rees cells. J Histochem Cytochem 28:788–791

Stein H, Gerdes J, Schwab U, Lemke H, Mason DY, Ziegler A, Schienle W, Diel V (1982) Identification of Hodgkin and Sternberg-Reed cells as a unique cell type derived from a newly detected small cell population. Int J Cancer 30:445–459

Stein H, Gerdes J, Schwab U, Lemke H, Diel V, Mason DY, Bartels H, Ziegler A (1983) Evidence for the detection of the normal counterpart of Hodgkin and Sternberg-Reed cells. Hematological Oncology 1:21–29