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The presence of erythroid cells in the thymus gland of man

MARION D. KENDALL AND JAYANTI SINGH
Department of Anatomy, St Thomas's Hospital Medical School, London SE1 7EH

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INTRODUCTION

The thymus gland is currently regarded as a lymphoid organ, important because of its capacity to produce large numbers of lymphocytes that enter the circulation as T-lymphocytes. The organ in adults has not hitherto been regarded as a site for erythropoiesis although this capacity has been observed in fetal and child thymus glands (Albert, Wolf, Pryjma & Vazquez, 1966; Taylor & Skinner, 1976).

The occurrence of extensive cyclical erythropoiesis in the thymus glands of birds (Kendall & Ward, 1974; Ward & Kendall, 1975; Bacchus & Kendall, 1975; Kendall, 1975*a*, *b*, 1978) and rodents (Albert, Wolf & Pryjma, 1965; Albert, Wolf, Pryjma & Vazquez, 1965; Kendall, in preparation) prompted the present search for evidence of thymic erythropoiesis in man.

MATERIALS AND METHODS

The right lobe of the thymus was biopsied during open heart surgery performed on 35 patients. The patients were both male and female and the adults ranged in age from 20–60 years of age. Biopsies from the thymus gland of three children (aged 6, 7 and 12 years) undergoing corrective cardiac surgery were also included in this study. The haemoglobin levels prior to surgery were all between 12·5 and 16·0 %, except in one patient (11·9 %) who was iron-deficient. The packed cell volumes, where determined, were between 45 % and 48 %. Samples of the lobes were fixed in 10 % formaldehyde and prepared for light microscopy with haematoxylin and eosin staining. A small portion of the thymus, judged by eye to be relatively fat-free, was finely minced and fixed in ice cold 5 % glutaraldehyde in phosphate buffer (pH 7·3). After an overnight wash in ice cold buffer plus 10 % sucrose the tissues were post-fixed in 1 % osmium tetroxide in veronal acetate buffer (pH 7·4), dehydrated, embedded in Araldite and cut with glass and diamond knives. Thick sections (1 μm) from 4–6 blocks from each gland were stained with Azur II and viewed in the light microscope. Areas containing cortex or medulla were then selected for thin sectioning. Thin sections were stained with uranyl acetate and examined in AEI electron microscopes.

RESULTS

Examination of the haematoxylin and eosin stained sections of the thymic biopsies showed normal histology in all cases with no evidence of neoplasia or germinal centre formation. All the patients, including the children, had some degree of fatty involution in the thymus, but this is normal being due to age-related changes.

In the material from 12 patients the lymphoid tissue was either very scanty or

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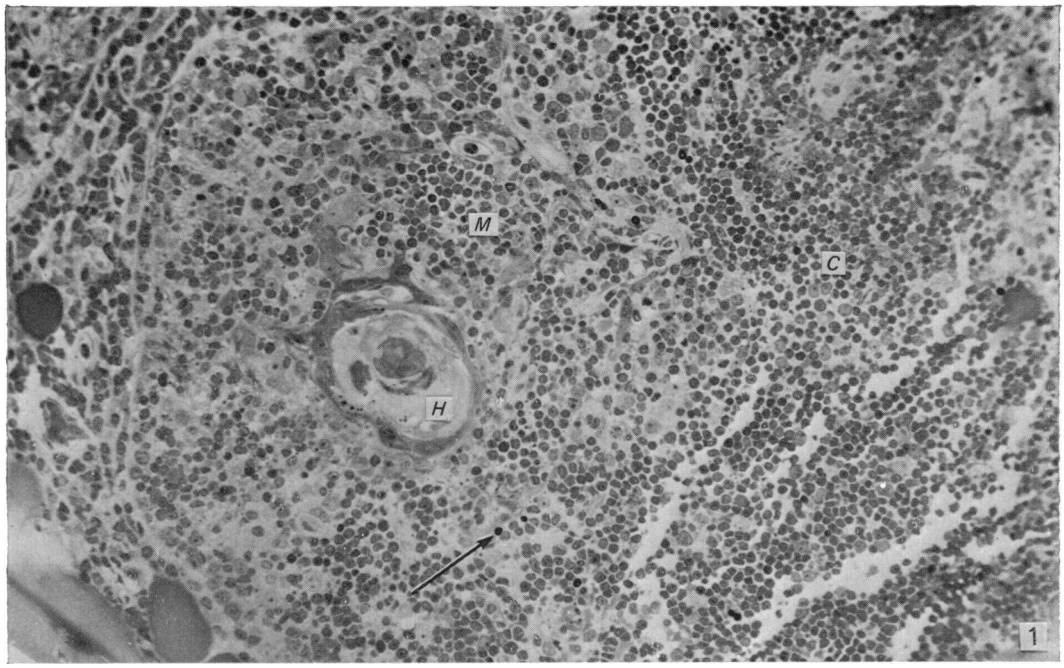

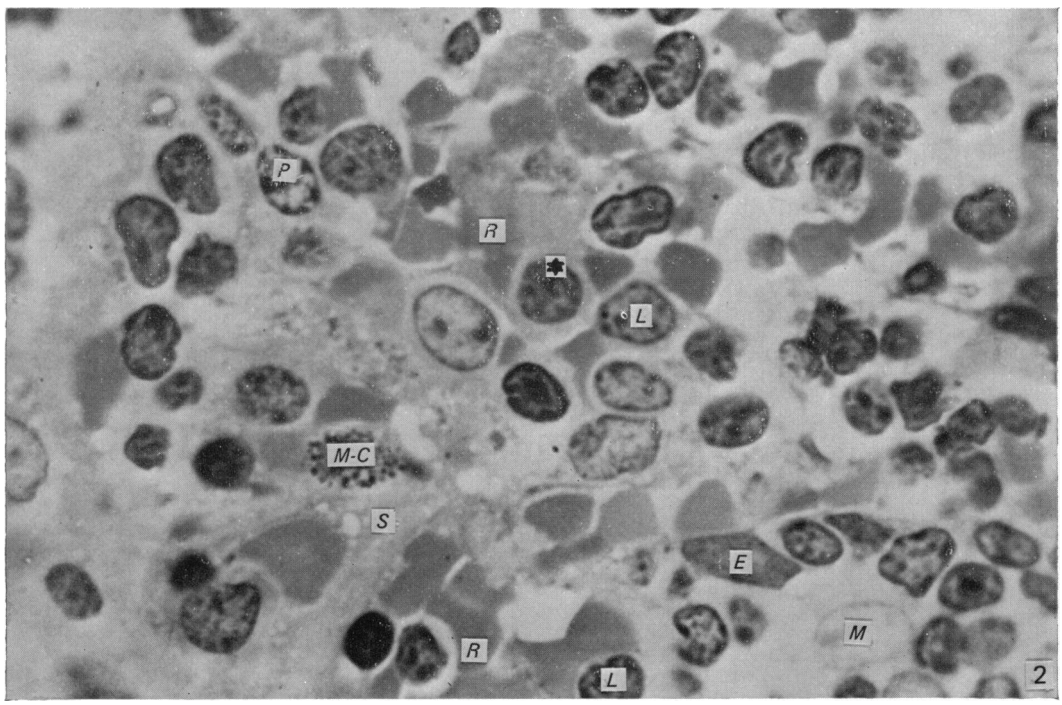

Fig. 1. The cortex (*C*) and medulla (*M*) of the thymus gland of a woman of 48 years of age undergoing surgery for aortic and mitral valve replacement. *H*, Hassall's corpuscle. The arrow indicates a cell that may be a normoblast or a pyknotic cell. × 160.

Fig. 2. An area of cortex adjacent to a septum (*S*) in the thymus gland of a girl of 6 years biopsied while undergoing corrective heart surgery. The cortex contains a large number of erythrocytes (*R*) lying between lymphoid cells (*L*). A possible early erythroid cell is marked*. Plasma cells (*P*) and a mast cell (*M-C*) lie outside the cortex, probably in the perivascular space. Epithelial-reticular cells (*E*) and macrophages (*M*) are within the cortex. × 640.

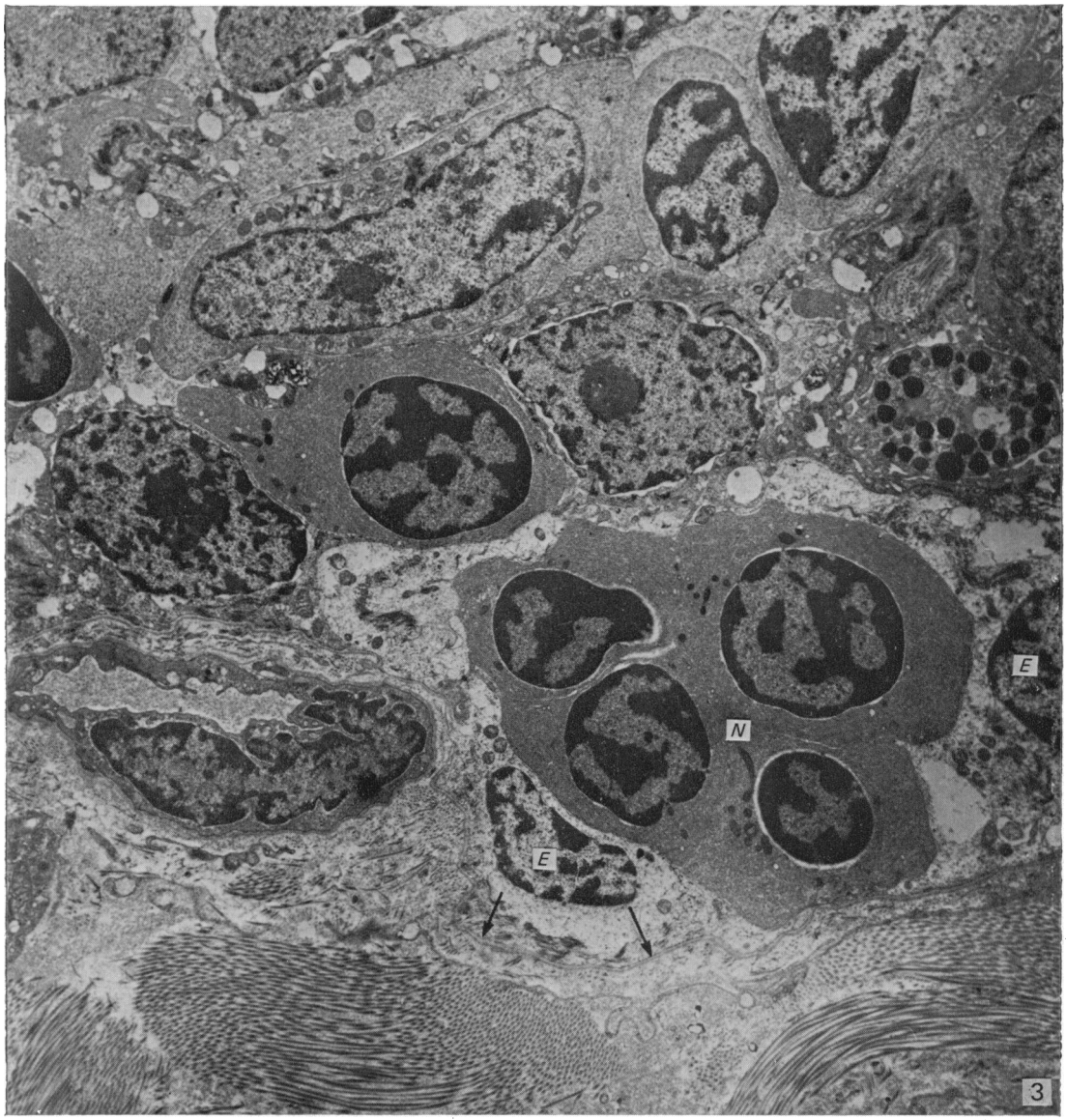

Fig. 3. The thymic cortex from a man of 48 years of age undergoing saphenous vein by-pass surgery. Arrows indicate the basal lamina of the epithelial-reticular cells (*E*) that separates the perivascular space containing a blood vessel from the cortex of the gland. Four normoblasts (*N*) are completely surrounded by epithelial-reticular cells. A mast cell with dense granules is close by. × 6000.

non-existent, and no further examinations were undertaken. Material from the remaining 23 patients all contained lymphoid tissue that could be identified as cortex or medulla or both. A light micrograph (Fig. 1) shows the typical appearance of these thymic biopsies with identifiable lymphoid cells, epithelial–reticular cells, macrophages, granulocytes, plasma cells and erythrocytes within the parenchyma of the gland (Fig. 2). Some sections contained very large numbers of mitotic figures. Pyknotic cells were looked for in all the sections, but when cells with dense nuclei

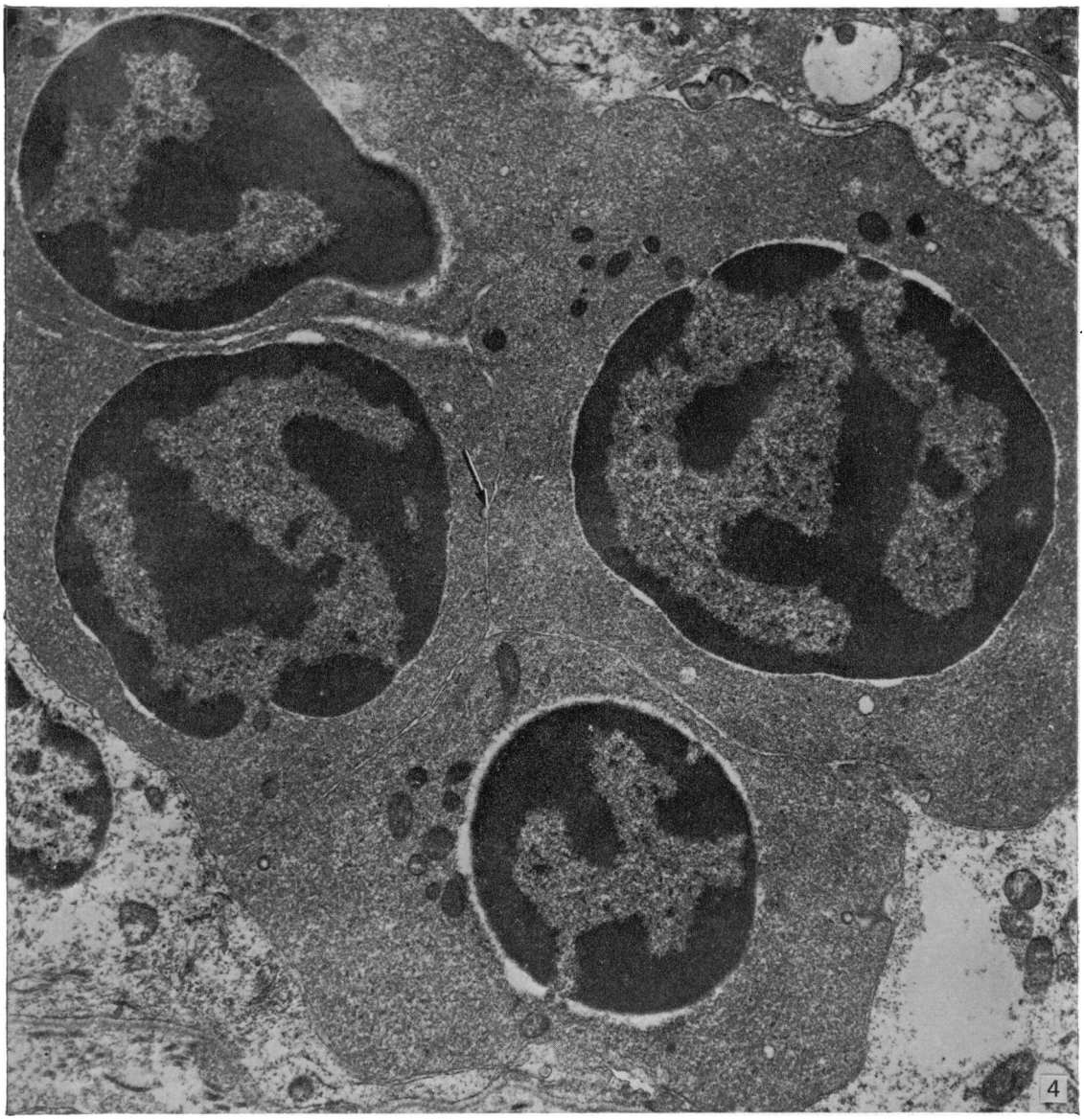

Fig. 4. An enlargement of the 4 normoblasts shown in Fig. 3. Mitochondria may still be identified within the fairly electon-dense cytoplasm. Arrow indicates a rhopheocytotic vesicle. Cytoplasm from epithelial-reticular cells completely surrounds the normoblasts on this section. ×15000.

were found it was not possible at this level of resolution to determine if they were pyknotic cells or late normoblasts. However, pyknotic nuclei were observed within macrophages in some sections. Twelve patients had large numbers of mature erythrocytes within the parenchyma of the thymus, more frequently in the cortex. These cells were quite clearly *not* contained within the blood vessels of the gland. Either these cells had escaped from the blood vessels or they had developed *in situ*.

The presence of normoblasts in the electron micrographs from two patients confirmed erythrocyte development within the thymus (Fig. 3). Such cells were

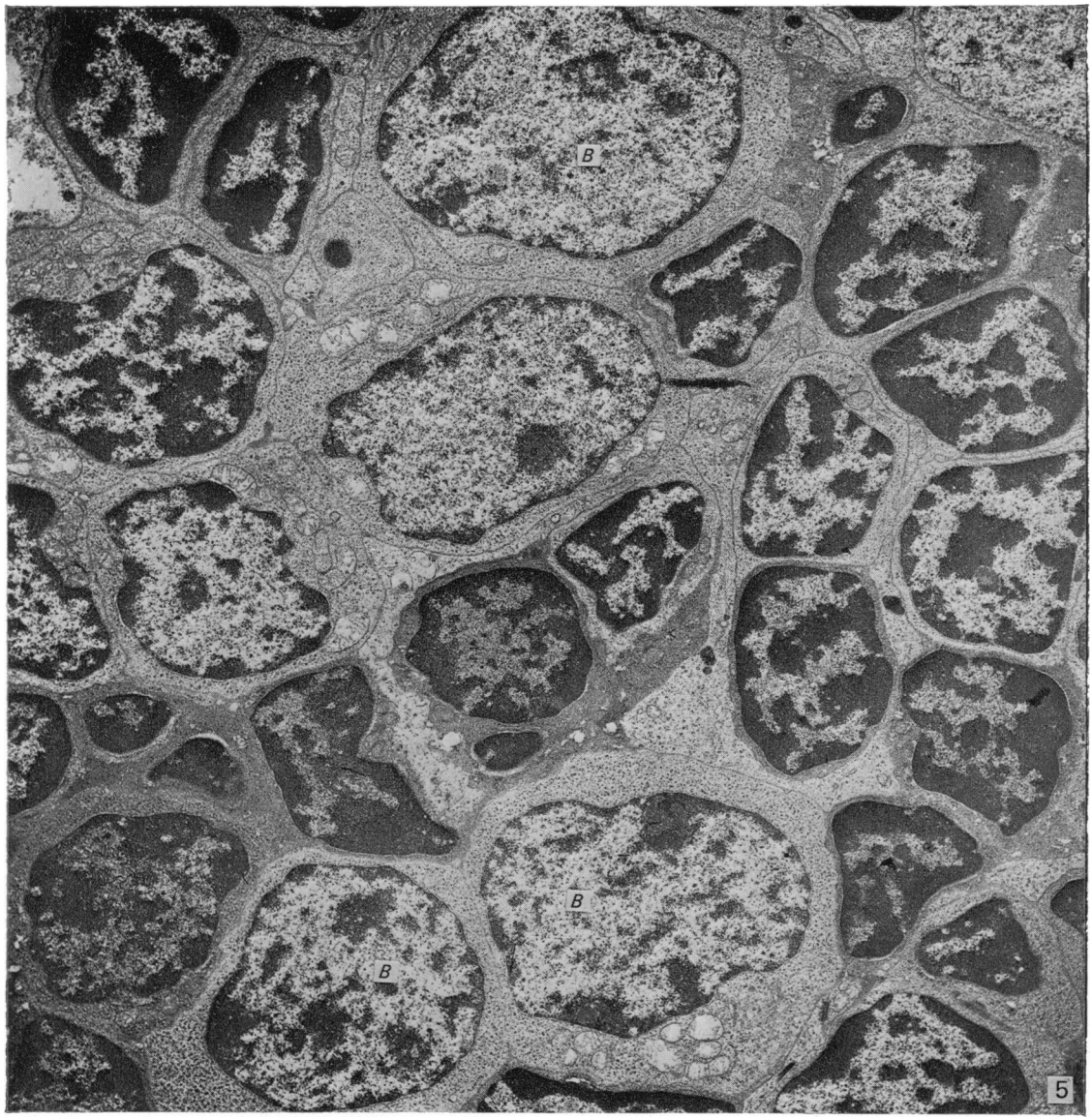

Fig. 5. Possible blast cells (*B*) in the cortex of a woman of 33 years of age undergoing corrective heart surgery. × 3000.

found in small groups, often surrounded by epithelial-reticular cells (Fig. 4). Most erythroid groups were close to septa that contained blood vessels. Of the other twelve thymus glands examined under the electron microscope, ten contained cells that were probably erythroid, but the morphology was not as distinctive as that of the cells shown in Figure 3. These probable erythroid cells were more electron-dense than the surrounding lymphocytes, and some contained rhopheocytotic vesicles and small aggregates of particulate material similar to ferritin. The cytoplasm in some cells was similar in content and extent to that of the normoblasts of Figure 3, but was usually less dense.

Many of the anucleate mature erythrocytes found apparently free within the parenchyma of the gland differed from one another with regard to the degree of electron density of their cytoplasm, and in the extent to which remnants of organelles could be seen within the cytoplasm. These observations suggest that at least some of the cells were recently formed, and could have developed within the thymus.

The lymphoid cells present ranged from typical small pachychromatic lymphocytes through a variety of larger, more leptochromatic cells to several that could be blast cells (Fig. 5). Possible proerythroblasts were identified. One series of sections contained profiles of a megakaryocyte, a cell type that, like the erythroid cell, is normally associated with the bone marrow.

DISCUSSION

This investigation has shown that a small number of erythroid cells was present in many of the thymus glands of patients undergoing open heart surgery. All stages in the erythroid series of cells could be identified once haemoglobinisation had commenced. Many larger, more primitive looking cells were observed, but it was not possible to identify them with the methods used.

Erythropoiesis in the fetal thymus gland has been reported, as has the presence of erythroid cells in the thymus glands of young children. Albert, Wolf, Pryjma & Vazquez (1966) identified erythroblasts in thymic material from 4 paediatric patients. More recently, Taylor & Skinner (1976), using histochemical and immunohistochemical methods on material obtained from 35 fetuses, 5 stillbirths, 32 neonatal deaths and 6 childhood deaths, have demonstrated the presence of cells of the erythroid series. They found it difficult to assess the proportion of erythroid cells in the thymus, but in adequately fixed material it appeared that less than 5 % of the round cells (including all lymphocytes) stained for haemoglobin, and in two cases the percentage of haemoglobin-containing cells was up to 30 %. They concluded that significant thymic erythropoiesis occurs in the mid-term fetus.

The fact that the apparently normal adult thymus may produce erythrocytes may seem surprising since the thymus for many years has been considered as a major lymphoid organ. However, there are clearly subtle interactions between T-lymphocytes and other haematopoietic cells that are only now being demonstrated. A recent review by Cline & Golde (1979) quotes four lines of evidence for T-lymphocyte interactions with early erythroid cells (burst forming units-erythroid, BFU-E) indicating that T-lymphocytes and their products affect early erythroid progenitor cells and thereby interact with erythropoietin in controlling erythropoiesis. They also include evidence for T-cells increasing the numbers of colony forming units – spleen (CFU-S), and for the effect of the stromal cells in favouring certain lines of development.

Thus in some ways it may be more pertinent to regard the thymus as a haematopoietic organ in which cell differentiation normally favours the lymphoid line (as the bone marrow tends to favour granulopoiesis and erythropoiesis), and to look for the factors in the microenvironment which influence the direction of cell development.

The demonstration of thymic erythropoiesis, and thus of an indirect *in vivo* support for T-lymphocyte interaction in the control of erythropoiesis, may be relevant to our understanding of thymoma-associated aplastic anaemia. Some patients with aplastic anaemia prove to have thymomas, and partial or complete thymectomy has given some remission and in several cases a complete cure for the

condition (Ross, Finch, Street & Streider, 1954; Jacobs, Hutter, Pool & Ley, 1959; Anon, 1959; Parry, Kilpatrick & Hardisty, 1959). Thymic involvement in the condition has previously been inexplicable.

SUMMARY

Biopsies of the right lobe of normal thymus glands without signs of neoplasia or germinal centre formation from 35 patients ranging in age from 20 to 60 years of age, and from 3 children aged 6, 7 and 12, showed on electron microscopic examination of the material from 14 patients that in 12 cases erythroid cells of all stages of development past the beginning of haemoglobinisation were present in some degree. Earlier erythroid cells could not be identified on morphological grounds with certainty, but cells which could have been lymphoblasts, proerythroblasts and stem cell were all observed. A section of a megakaryocyte was seen in one thymus. The importance of erythropoiesis within the thymus gland is briefly discussed.

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