Thymus-Dependent Lymphocytes in Human Bone Marrow

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Abstract Human bone marrow is known to contain significant numbers of bursa-dependent lymphocytes. The presence of thymus-dependent (T) cells is controversial. Bone marrow cells obtained from healthy volunteers was fractionated by density centrifugation. A lymphocyte-enriched subpopulation was shown to be reactive to alloantigens in mixed lymphocyte culture and to contain substantial numbers of T lymphocytes. The T lymphocytes were detected by cell surface markers (rosette formation with sheep RBC) and by response to the mitogens phytohemagglutinin and concanavalin A. Bone marrow T cells exhibited functional characteristics quantitatively different from peripheral blood T cells, suggesting that they may represent a particular subpopulation of T cells. The lymphocyte-enriched fraction additionally contained committed granulopoietic stem cells capable of colony formation in semisolid gel. The presence of T cells in human bone marrow is consistent with findings in other mammals and may explain the high incidence of graft versus host disease in bone marrow transplant recipients.

Introduction Peripheral blood lymphocytes can be divided into two major subpopulations: thymus-dependent (T) cells and bursa-dependent (B) cells. In most species, bone marrow lymphocytes are predominantly of the B-cell type (1-5). Recent studies using mice indicate that T cells and T-cell progenitors are also present in the bone marrow (6-11). The situation in man is unclear.

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Abbreviations used in this paper: B, bursa-dependent; B-RFC, B-rosette-forming cells; CFU-C, colony-forming cells; ConA, concanavalin A; HBSS, Hanks' balanced salt solution; PBS, phosphate-buffered saline; PHA, phytohemagglutinin; SRBC, sheep red blood cells; T, thymus-dependent; T-RFC, T-rosette-forming cells.

Several studies have reported the absence of cells with T characteristics (4, 5, 12) while other investigators found phytohemagglutinin (PHA) and alloantigen-responsive cells in human marrow (13, 14).

In addition to lymphoid cells, bone marrow contains committed granulopoietic stem cells (CFU-C) capable of colony formation in semisolid gel. These cells and their progenitor, the pluripotent hematopoietic stem cell, are morphologically similar to lymphocytes (15, 16). In murine systems, hematopoietic stem cells have been separated from lymphocytes by density centrifugation (7, 17).

In order to determine if T lymphocytes are present in human marrow, we obtained multiple small aspirates from normal volunteers. Mononuclear cells were isolated by density centrifugation and studied for T- and B-lymphocyte cell surface marker and functional characteristics and for colony-forming capacity in agar. The results indicate that substantial numbers of lymphocytes with T-cell characteristics are present in normal human bone marrow. Bone marrow T cells differ from peripheral blood T cells in their response to mitogens and alloantigens.

Methods Bone marrow cells. Informed consent was obtained from normal volunteers, ages 20 to 35 yr. Bone marrow was obtained from the posterior iliac crest by a sterile technique. 1-ml aliquots were carefully aspirated from separate sites along the posterior iliac crest and mixed with 500 U of preservative-free heparin. 60-ml samples of heparinized peripheral venous blood were obtained from marrow donors at the time of marrow aspiration.

Fractionation of bone marrow. The heparinized bone marrow was centrifuged (2,000 g at 24°C) for 2 min and the leukocyte-rich supernate was recovered. Antisera to A and/or B substance (Ortho Diagnostics, Raritan, N. J.) were added, as determined by the blood type of the donor, and the suspension was mixed gently for 5 min at 24°C to agglutinate the red cells. The suspension was centrifuged at 1,000 g at 24°C for 5 s and the nucleated cells recovered and carefully layered over a Ficoll-Hypaque gradient prepared according to the method of Boyum (18). The gradient tube was then centrifuged at 1,000 g at 24°C for 2
min. The light cell fraction (I) was collected by aspirating
the interface cells. After a preliminary centrifugation
at 3,000 g at 24°C for 1 min, the supernate was discarded
and the cells resuspended in McCoy's medium (Grand
Island Biological Co., Santa Clara, Calif.) and centrifuged
at 1,000 g at 24°C for 7 s. The supernate containing the
light cells was aspirated and saved, and the pellet was
resuspended with McCoy's and recentrifuged. This process
was repeated five times, and the supernates were pooled,
 washed once in TC-199 medium (Grand Island Biological
Co.) and resuspended to the desired concentration.

To prepare the heavy cell fraction (II), the pellet from the
Ficoll-Hypaque gradient was recovered and resuspended
in McCoy's. Additional anti-A and/or -B antisera were
added and incubated with mixing at 24°C for 3-5 min. The
suspension was centrifuged at 1,000 g at 24°C for 2 s and
the supernate recovered and centrifuged at 3,000 g at 24°C
for 1 min. The supernate was then discarded and the
pellet resuspended in McCoy's and centrifuged at 1,000 g
at 24°C for 15 s to remove contaminating light cells. The
pelleted cells were then resuspended in TC-199 to the
desired concentration. Peripheral blood lymphocytes were
prepared as described above for the light cell fraction
with heparinized peripheral blood. Cytocentrifuge slides
of the fractionated bone marrow were stained with May-
Grunwald-Giemsa and differential counts performed.

Mixed lymphocyte culture (MLC). MLC studies were
performed by using a one-way semimicro-technique (19).
Briefly, responder cells (5 × 10⁶) mixed with an equal
number of stimulator cells in 0.1 ml were incubated at 37°C
for 96 h. Tritiated thymidine (0.5 μCi; sp act 22 Ci/mmol;
Schwarz/Mann Div., Becton, Dickinson & Co., Orangeburg,
N.Y.) was added for an additional 16 h, and cultures har-
collected and the trichloroacetic acid-precipitable [3H]thymidine
was quantified by counting in a Searle scintillation counter
(Amersham/Searle Corp., Arlington Heights, Ill.). Results
are expressed as mean±SEM counts per minute of triplic-
ate cultures. Stimulation ratios were calculated by divid-
ing the mean counts per minute of stimulated cultures by
the mean of autologous control cultures (background).
Stimulation indices ≥2 are significant (19). Stimulator
cells pretreated with mitomycin are designated by the
subscript "M."

Mitogen stimulation. Mitogen stimulation studies were
performed under conditions identical to those used for the
MLC. Responder cells, 1 × 10⁶ in 0.1 ml, were mixed with
0.01 ml of a 1/10 dilution of phytohemagglutinin
M. (PHA-M; Difco Laboratories, Detroit, Mich.) or 0.01
ml of a serial dilutions of concanavalin A (ConA; Calbio-
chem, Los Angeles, Calif.) and incubated for 96 h. Cul-
tures were processed as previously described.

T-rosette-forming cells (T-RFC). Spontaneous T-RFC
were determined by a modification of the method previ-
ously described (20). 5 × 10⁶ cells (0.1 ml) in Hanks' bal-
canted salt solution (HBSS; Grand Island Biological Co.)
were mixed with 0.1 ml of a 5% suspension of sheep red
blood cells (SRBC) in a disposable glass 16 × 75-mm
survival tube. Fresh SRBC in Alsevers' solution were obtained
weekly (Rosemead Laboratories, Rosemead, Calif.), washed
twice before use, and resuspended in HBSS. 25 μl of fetal
calf serum, previously absorbed with 1 vol of SRBC, was
added and the mixture centrifuged at 400 g at 4°C for 5
min. Tubes were incubated at 24°C for 90 min, the pellet
gently resuspended, and an aliquot removed for counting.
T-RFC were determined by phase microscopy; viable cells
surrounded by three or more SRBC were scored as roset-
es. A minimum of 100 cells was counted and T-RFC reported
as a percentage.

B-rosette-forming cells (B-RFC). B-RFC were deter-
mined by rosette formation with antibody (A) - and com-
plement (C)-coated SRBC (E)—EAC. A 5% solution of
SRBC was incubated with anti-sheep-hemolysin A
(Grand Island Biological Co.) at one dilution below the
hemagglutinating titer, at 37°C for 30 min (1 vol/1 vol).
The "sensitized" SRBC were washed twice, resuspended in
HBSS, and incubated with a 10² dilution of normal
mouse serum (1 vol/1 vol) at 37°C for 30 min. These
SRBC, designated "EAC," were washed three times and
resuspended in HBSS. The cells to be tested (5 × 10⁶ in
0.1 ml) and 0.1 ml of a 0.5% suspension of EAC were
incubated on a rotating wheel (6 rpm) at 37°C for 60
min. B-RFC were determined microscopically in the same
manner as T-RFC. The assay detects cells with a C3
receptor (21). Controls using unsensitized and sensitized
SRBC did not form rosettes under these conditions.

Immunofluorescence. Immunoglobulin-bearing cells
were determined by direct staining with fluorescein-conjugated
anti-immunoglobulin (22). 5 × 10⁶ cells (0.1 ml) were
washed twice in phosphate-buffered saline (PBS) and
incubated with 0.015 ml of fluorescein-conjugated goat anti-
human immunoglobulin (Meloy Laboratories Inc., Spring-
field, Va.) at 0°C for 30 min. Cells were washed three
times in cold PBS with 0.02% NaN₃ and immunofluores-
cent-positive cells were determined by fluorescence micros-
copy (E. Leitz, Wetzler, Germany).

Marrow cell culture. Colony-forming cells (CFU-C)
were assayed by a standard double-layer agar technique
(23, 24). McCoy's 5A medium with 15% fetal calf serum
and antibiotics was used throughout. Bone marrow cells
were washed in complete medium and viable cell counts
performed in a hemocytometer with trypan blue. 2 × 10⁵
marrow cells were plated in 0.3% agar on feeder layers
of 1 × 10⁶ normal peripheral leukocytes immobilized in 0.5%
agar. Duplicate plates were incubated in a humidified
environment of 7.5% CO₂ at 37°C. Colonies composed of
40 or more cells were counted in the upper agar layer at
10 days with a dissecting microscope. Selected colonies
were "picked" with a finely drawn pipette, smeared, and
stained with Giemsa for morphological examination.

TcR-labeled RBC stain. Radiolabeled TcR studies
were kindly performed by Dr. William Figueroa. The percent
of a bone marrow aliquot which might be contributed by
dilution with peripheral blood was determined as follows.
20 ml of sterile heparinized blood was labeled with ⁵¹Cr as
described (25) and reinfected into the donor. Simultaneous
blood and bone marrow were obtained at 180 min and ali-
quots assayed for hemoglobin concentration and gamma
emission.

RESULTS

Bone marrow fractionation. Bone marrow was sepa-
rated into light (fraction I, Fig. 1) and heavy (frac-
tion II, Fig. 2) fractions by density centrifugation.

Fraction I was comprised predominantly of lymphocytes;
small numbers of immature granulocytes and mono-
cytes were present (Table I). Fraction II contained
granulocytes in all stages of differentiation; there was
minimal contamination with lymphocytes and mono-
cytes. Small numbers of nucleated RBC's and plasma
were observed in both fractions.

Cell surface markers. The results of studies of T-
and B-cell surface markers are presented in Table II.
Peripheral blood contained 2 × 10⁶ lymphocytes per ml,
**Figure 1** Fraction I cells. Light-density bone marrow cells fractionated by buoyant density centrifugation.

**Figure 2** Fraction II cells. Heavy-density bone marrow cells fractionated by buoyant density centrifugation.
70% of which were T cells; the total T-cell content of peripheral blood was therefore $1.4 \times 10^6$ per ml ($2 \times 10^6 \times 0.70$). Similar calculations for B cells indicated $0.3-0.4 \times 10^6$ B cells per ml. Identical calculations were performed for unfraccionated and fractionated bone marrow cells. Examination of Table II reveals the following (a) unfraccionated bone marrow contains sixfold more T cells than can be explained by peripheral blood contamination ($8.7 \times 1.4 \times 10^6$ per ml); (b) similar results were found evaluating bone marrow B cells ($2.0 \times 0.3 \times 10^6$ per ml); (c) there are approximately four times more T cells than B cells in bone marrow ($7.3 \times 1.7 \times 10^6$ per ml); (d) fractionation results in a 50-55% cell recovery with no selection for fraction I vs. fraction II cells or T vs. B cells (45 vs. 55%); (e) fraction I contained cells with T- and B-cell markers; (f) fraction II contained a small number of T- and B-RFC and a large number of immunofluorescence positive cells. The latter exhibited a speckled staining pattern characteristic of aggregate binding, rather than the diffuse pattern of surface immunoglobulin observed with fraction I cells; (g) 65-75% of bone marrow lymphocytes could be classified as T or B cells by using these markers.

**MLC.** Unfraccionated bone marrow cells had a high spontaneous $[^{3}H]$thymidine incorporation and were not stimulated by allogeneic cells (Table III). Less incorporation was observed in cultures with allogeneic stimulator cells. Fraction I cells had a lower spontaneous incorporation than unfraccionated cells and were stimulated by allogeneic lymphocytes ($B \times D_m$, $B \times E_{m}$ = Table IV). In most instances, fraction I cells had a lower stimulation index than lymphocytes from the same donor tested against the same stimulator cell ($A \times D_m$, $B \times E_{m}$). Fraction II cells had a high spontaneous incorporation and could not be stimulated. Fraction II cells showed decreased incorporation in cultures with allogeneic lymphocytes ($C \times D_m$, $D \times E_{m}$). Unrelated lymphocytes were stimulated by both fraction I ($D \times B_m$, $E \times B_m$) and II cells ($D \times C_m$, $E \times C_m$). These results were reproduced with cells from five donors.

To evaluate responder cell kinetics, MLC were performed with a fourfold number of responder cells. Fraction II cells were not stimulated by allogeneic cells while fraction I cells exhibited an increase in the stimulation index.

**Mitogen response.** Fractionated marrow cells were tested for response to PHA and ConA (Table V). Fraction I cells were stimulated by both mitogens and fraction II cells were not. Similar results were obtained over a range of mitogen concentrations. Doubling the
### Table III

**MLC Reactivity of Bone Marrow**

<table>
<thead>
<tr>
<th>Responder cell</th>
<th>Stimulator cell†</th>
<th>Cell Donor Types</th>
<th>AM</th>
<th>BM</th>
<th>CM</th>
<th>DM</th>
<th>EM</th>
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<tr>
<td>A</td>
<td>A. D. Lymph</td>
<td>Mean</td>
<td>383</td>
<td>796</td>
<td>3,147</td>
<td>2,486</td>
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<td></td>
<td>SEM</td>
<td>44</td>
<td>166</td>
<td>285</td>
<td>331</td>
<td>675</td>
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<td></td>
<td>SI</td>
<td>1.0</td>
<td>2.1</td>
<td>8.2</td>
<td>6.5</td>
<td>10.5</td>
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<tr>
<td>B</td>
<td>A. D. BM</td>
<td>Mean</td>
<td>6,196</td>
<td>4,887</td>
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<td>307</td>
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<td>0.4</td>
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<td>C</td>
<td>R. G. Lymph</td>
<td>Mean</td>
<td>4,969</td>
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<td>1,553</td>
<td>1,476</td>
<td>4,509</td>
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<tr>
<td></td>
<td>SEM</td>
<td>456</td>
<td>285</td>
<td>136</td>
<td>111</td>
<td>403</td>
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<tr>
<td></td>
<td>SI</td>
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<td>2.3</td>
<td>1.0</td>
<td>1.0</td>
<td>2.9</td>
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</tr>
<tr>
<td>D</td>
<td>R. G. BM</td>
<td>Mean</td>
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<td>6,233</td>
<td>5,622</td>
<td>3,508</td>
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<tr>
<td></td>
<td>SEM</td>
<td>298</td>
<td>422</td>
<td>1,971</td>
<td>397</td>
<td>445</td>
<td></td>
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<tr>
<td></td>
<td>SI</td>
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<td>0.8</td>
<td>1.1</td>
<td>1.0</td>
<td>0.6</td>
<td></td>
</tr>
<tr>
<td>E</td>
<td>UC</td>
<td></td>
<td>Lymph</td>
<td>Mean</td>
<td>9,236</td>
<td>7,828</td>
<td>8,403</td>
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<tr>
<td></td>
<td>SEM</td>
<td>508</td>
<td>1,330</td>
<td>1,340</td>
<td>1,246</td>
<td>218</td>
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<tr>
<td></td>
<td>SI</td>
<td>6.0</td>
<td>5.1</td>
<td>5.5</td>
<td>5.3</td>
<td>1.0</td>
<td></td>
</tr>
</tbody>
</table>

* See Methods for details. Results are expressed as mean ±SEM counts per minute; n = 3. SI, stimulation index. † Mitomycin pretreated. § Lymph, peripheral blood lymphocyte; BM, unfractionated bone marrow. || UC, unrelated control.

### Table IV

**MLC Reactivity of Fractionated Bone Marrow**

<table>
<thead>
<tr>
<th>Responder cell</th>
<th>Stimulator cell‡</th>
<th>Cell Donor Types</th>
<th>AM</th>
<th>BM</th>
<th>CM</th>
<th>DM</th>
<th>EM</th>
</tr>
</thead>
<tbody>
<tr>
<td>A</td>
<td>M. K. Lymph</td>
<td>Mean</td>
<td>1,263</td>
<td>1,163</td>
<td>1,234</td>
<td>10,497</td>
<td>8,836</td>
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<tr>
<td></td>
<td>SEM</td>
<td>453</td>
<td>190</td>
<td>131</td>
<td>1,298</td>
<td>1,074</td>
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</tr>
<tr>
<td></td>
<td>SI</td>
<td>1.0</td>
<td>0.9</td>
<td>1.0</td>
<td>8.3</td>
<td>7.0</td>
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<tr>
<td>B</td>
<td>M. K. I</td>
<td>Mean</td>
<td>2,320</td>
<td>1,685</td>
<td>1,962</td>
<td>9,496</td>
<td>9,420</td>
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<tr>
<td></td>
<td>SEM</td>
<td>123</td>
<td>49</td>
<td>354</td>
<td>558</td>
<td>1,958</td>
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<tr>
<td></td>
<td>SI</td>
<td>1.4</td>
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<td>1.2</td>
<td>5.6</td>
<td>5.6</td>
<td></td>
</tr>
<tr>
<td>C</td>
<td>M. K. II</td>
<td>Mean</td>
<td>11,320</td>
<td>10,135</td>
<td>11,889</td>
<td>4,703</td>
<td>4,719</td>
</tr>
<tr>
<td></td>
<td>SEM</td>
<td>1,703</td>
<td>1,774</td>
<td>453</td>
<td>346</td>
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</tr>
<tr>
<td></td>
<td>SI</td>
<td>1.0</td>
<td>0.9</td>
<td>1.0</td>
<td>0.4</td>
<td>0.4</td>
<td></td>
</tr>
<tr>
<td>D</td>
<td>UC-1</td>
<td></td>
<td>Lymph</td>
<td>Mean</td>
<td>13,978</td>
<td>13,555</td>
<td>9,901</td>
</tr>
<tr>
<td></td>
<td>SEM</td>
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<td>422</td>
<td>101</td>
<td>65</td>
<td>793</td>
<td></td>
</tr>
<tr>
<td></td>
<td>SI</td>
<td>6.8</td>
<td>6.6</td>
<td>4.8</td>
<td>1.0</td>
<td>3.0</td>
<td></td>
</tr>
<tr>
<td>E</td>
<td>UC-2 Lymph</td>
<td>Mean</td>
<td>9,135</td>
<td>9,596</td>
<td>6,695</td>
<td>7,045</td>
<td>1,373</td>
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<tr>
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<td>SEM</td>
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<td>471</td>
<td>247</td>
<td>1,099</td>
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<td>4.9</td>
<td>5.1</td>
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</table>

* See Methods for details. Results expressed as mean ±SEM counts per minute ±SEM; n = 3. SI, stimulation index. † Mitomycin pretreated. § Lymph, peripheral blood lymphocyte; I, light-density bone marrow cells; II, heavy density bone marrow cells. See Methods. || UC, unrelated control.
number of responder cells resulted in the same pattern of response to PHA and allogeneic cell stimulation. Peripheral blood lymphocytes and fraction I cells were compared for proliferative response over a range of PHA and ConA concentrations (Fig. 3). Approximately similar curves were observed but peripheral blood lymphocytes consistently proliferated to a greater degree than fraction I cells. Both populations tested contained an equivalent percentage of T cells (70 vs. 65%).

Colony-forming capacity. Unfractionated bone marrow contained between 28 and 140 CFU-C per $2 \times 10^5$ marrow cells (Table VI). Fraction I cells were enriched three- to sixfold for CFU-C while fraction II cells were markedly depleted. Morphologic examination of colonies from the three populations failed to reveal significant differences.

$^{51}$Cr-labeled RBC studies. Peripheral blood samples had a hemoglobin concentration of 13.6 g/100 ml while in bone marrow it was 14.2 g/100 ml. Gamma emissions of a 1-ml aliquot of blood and bone marrow were 327 and 288 cpm, respectively. These studies suggest that 88% of bone marrow volume is composed of peripheral blood. The remaining 12% is composed of bone marrow cells ($\leq 3\%$) and marrow “fluid” not in 180-min equilibrium with the peripheral blood.

**DISCUSSION**

Human bone marrow contains cells with characteristics of T lymphocytes. These cells demonstrated rosette formation with sheep erythrocytes, reactivity in MLC, and responsiveness to T-cell mitogens (PHA and ConA).

Bone marrow can be fractionated on the basis of cell density into MLC-responsive and nonresponsive subpopulations. The responsive population (fraction I) contains cells with morphologic, functional, and cell surface marker characteristics of lymphocytes. The fraction also contains cells with the ability to form granulocytes and monocyte-macrophage colonies in semisolid gel (CFU-C). The MLC-nonresponsive population (fraction II) contains predominantly immature and mature granulocytes; very small numbers of T-RFC are present. This fraction is depleted of cells with CFU-C.
activity, is unresponsive to mitogenic stimulation, and is able to stimulate allogeneic lymphocytes strongly. Fraction II cells exhibit a high spontaneous DNA synthetic level that was reduced in the presence of allogeneic lymphocytes. The mechanism underlying this phenomenon is unclear.

The present study differs from previous reports (4, 5, 14) in several regards: (a) We demonstrated T-RFC in bone marrow. The method of rosette testing differed from other studies. (b) We were unable to demonstrate MLC reactivity in unfraccionated bone marrow. The MLC reactivity of unfraccionated bone marrow reported by some authors (14) was low with stimulation indices < 2. (c) We found a substantial DNA synthetic response to PHA, using a different source and concentration of the mitogen. Our findings are similar to those reported in murine systems. Claman (6) demonstrated a PHA and a ConA response in murine bone marrow and El-Arini and Osoba (7) have demonstrated T cells and T-cell progenitors in light-density fractions of murine bone marrow. These investigators were able to show MLC reactivity and graft versus host disease (11) with marrow lymphocytes. Burkart and Meuwissen (13) have demonstrated PHA- and pokeweed mitogen-responsive cells in primate bone marrow.

It is important to exclude contamination of bone marrow samples with peripheral blood as the sole source of T cells. Several arguments militate against this possibility. All marrow samples were exactly 1-ml aspirates and were carefully collected. Dicke and Lowenberg showed no increase in PHA responsiveness of marrow aliquots in a range of 0.3-3 ml (26). While the percent of T cells (T-RFC) is comparable to peripheral blood T-lymphocyte levels, the response to mitogens and alloantigens was significantly different. Finally, the absolute number of T cells in bone marrow is too high to be accounted for by contamination with peripheral blood T cells. This data is additionally supported by the observations obtained with 3Cr-labeled erythrocytes.

It is not clear whether bone marrow T cells are identical to peripheral blood T cells. Bone marrow T-cell progenitors in mice appear to have decreased or absent \( \theta \)-antigen on their surface and have different response curves to PHA and ConA (27, 28). The situation in man may be comparable. The data on \( \theta \)-antigen has not been resolved but differences in responsiveness to PHA, ConA, and alloantigens were observed in this study.

The presence of T cells in human marrow may relate to the high incidence of graft versus host disease in bone marrow transplant recipients (29). As MLC-reactive cells are hypothesized to be precursors of cells responsible for mediating graft versus host disease, fractionation of bone marrow into MLC-reactive and non-reactive populations has potentially important implications. Cells with colony-forming activity segregated with MLC-reactive cells. This is not unexpected in view of the reported lymphoid morphology and density of the colony-forming cell (16, 30). There is no assay for the pluripotent stem cell in man. Nonetheless, data from animal work indicate that pluripotent stem cells, committed granulopoietic precursors (CFU-C), and bone marrow T cells and T-cell progenitors are present in light-density fractions. Even if these cells could be separated completely from bone marrow, the clinical value of doing so must be questioned. For example, if T cells were selectively removed from transplanted bone marrow, functional T cells could arise in the host from precursor lymphocytes or from the pluripotent stem cell. Our data suggest that the complete spectrum of lymphoid cell differentiation is manifest in human bone marrow.

Note added in proof: Since the preparation of the manuscript, A. S. Fauci has published data confirming several of the aspects presented in this paper (1975. Human bone marrow lymphocytes. I. Distribution of lymphocyte subpopulations in the bone marrow of normal individuals. J. Clin. Invest. 56: 98–110).

ACKNOWLEDGMENTS

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REFERENCES