The Morphologic Responses of the Lymphoid System to Homografts

III. Electron Microscopy Study

By Janine André-Schwartz

The rejection of a skin homograft by the rabbit is accompanied by the proliferation of large, basophilic cells (hemocytoblasts) in its lymphoid system. These cells first appear in the lymph node contiguous to the homograft; later, they are found in regional and distant lymphatic structures, but they are always most prominent in the lymph node draining the homograft. The administration of purine antimetabolites (6-mercaptopurine or thioguanine) inhibits the proliferation of hemocytoblasts and retards rejection of the graft. Those animals that reject the graft during treatment with antimetabolites develop an extreme proliferation of hemocytoblasts.

This paper deals with an electron microscopic study of this reaction. During the first set homograft response, large, round cells containing numerous cytoplasmic ribosomes and very little endoplasmic reticulum were observed. On the other hand, many of the cells of the second set response were typical plasma cells, rich in endoplasmic reticulum. This feature was not evident by light microscopy. When 6-mercaptopurine was administered with a first set homograft, the large, ribosome-containing cells were lacking. However, in the animal rejecting its homograft despite the administration of 6-mercaptopurine, numerous large, extremely primitive cells were seen.

Materials and Methods

Animals

Twenty-four outbred rabbits were used in these experiments. They weighed between 2.5 to 3 Kg.; no selection by sex was made. The animals were housed in the open air and fed water and Purina rabbit chow ad lib.

Technic of Skin Grafting

Pairs of rabbits selected at random were anesthetized with intravenous Nembutal, 20 mg. per Kg. After shaving the dorsal surface of the ear, a full thickness square of skin was excised and, after removing the subcutaneous fat, the graft was transplanted to a
corresponding graft-bed of the opposite member of the pair and sutured in place with 4-0 silk. Each graft was inspected daily for signs of rejection.

Solutions of 6-MP were prepared daily by dissolving 100 mg. of the powdered anti-metabolite* in 1 ml. of I N NaOH; the desired concentration for injection (10 mg./ml.) was made by appropriate dilution of the concentrated solution with physiologic saline. All injections of drugs were made subcutaneously.

*Generously supplied by the Laboratoires Diamant, Paris, France.
The following groups of rabbits were studied: six control animals that were not grafted; four animals that received one skin homograft. These were sacrificed on the 7th postoperative day, the peak of the homograft rejection in the contiguous lymph node. Four animals were sensitized by the sequential application of two skin homografts from the same donor, the second graft being applied approximately 1 week after the rejection of the first one. The rabbits were sacrificed on the 4th postoperative day, the peak of the second set reaction in the contiguous lymph node. Eight animals received a skin homograft and were begun on 6-MP, 10 mg./Kg. on the day of grafting. They were given the 6-MP daily and were sacrificed on the 18th or 20th day following application of the skin homografts. Two animals were given 10 mg./Kg./day of 6-MP and were not grafted.

**Processing of Tissues**

One hour prior to sacrifice, 0.5 ml. of Victoria Blue dye was injected subcutaneously at the base of the ear bearing the homograft, in order to facilitate identification of the cervical lymph nodes. The animals were killed with intravenous Nembutal and the most proximal lymph node was identified and removed.

**Fixation**

The tissues were divided into two samples: one was fixed for 20 minutes in 5 per cent acrolein formaldehydes at 4 C. After washing for 1 hour in Millonig buffer, it was post-fixed with 2 per cent osmium tetroxide buffered according to Millonig, and embedded in epon after dehydration in graded alcohols. The other sample was fixed directly in 2 per cent osmium tetroxide buffered according to Millonig, and embedded in epon. The tissues were cut with glass knives on a Porter Blum microtome. Section 0.5 µ thick (thick sections) were stained with azur blue and methylene blue. Ultra-thin sections were stained with either a 2 per cent aqueous solution of uranyl acetate for 20 minutes, or with lead, according to Karnovsky's method. The sections were examined with a Siemens Elmiskop I (80 kv., objective aperture 50 µ).

**RESULTS**

**Normal Controls**

The appearance of the normal lymph node fixed with aldehydes was similar to previous descriptions of osmium-fixed lymph nodes. However, details of the nuclear chromatin and cytoplasmic structures appeared more clearly in the aldehyde-fixed tissues. Figures 1A and 2 show a normal lymph node with the peculiar appearance, due to the aldehyde fixation, of the chromatin clumped along the nuclear membrane.

**First Set Reaction**

An intense proliferation of hemocytoblasts was the principle feature of this reaction. These were present, not only in the enlarged germinal centers and follicles, but extended as well into the medullary portions of the node

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Fig. 1.—Light microscopy. **A. Control, normal lymph node:** Mature lymphocytes (1) together with large light cells, reticulum cells (2) and flattened endothelial cells (3); fibroblasts (4) in the center. X900. **B. First set reaction (draining lymph node):** Proliferation of large round or ovoid cells with light nucleus and cytoplasm, large nucleoli, the “hemocytoblast” (1); a macrophage is seen (2). X900. **C. Second set reaction (draining lymph node):** Hemocytoblasts (1) and plasma cells (2); a capillary (3) is seen with two large cells, one in the wall (endothelial cell) and one in the lumen. X1000. **D. Animals rejecting their homograft when treated with 6-MP:** Few hemocytoblasts (1); many large round cells with nucleoli are constituents of the wall of a capillary; some small lymphocytes (2) between the large cells; some red blood cells in the lumen. X900.
Fig. 2.—Control: normal lymph node. Ultra-thin section. Uniform lymphocytic population in this field (1); chromatin clumped along the nuclear membrane showing typical aspect after aldehyde prefixation. X7500.
Fig. 3.—First set reaction (draining lymph node). Ultra-thin section. Presence of hemocytoblasts (1) among the lymphocytic population (2); lymphoblast (3). X5000.
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Table 1

<table>
<thead>
<tr>
<th>Hemocytoblast Type</th>
<th>Type I</th>
<th>Type II</th>
<th>Type III</th>
<th>Type IV</th>
</tr>
</thead>
<tbody>
<tr>
<td>Nucleus/Cytoplasm Ribosomes</td>
<td>6–8 μm/10–12 μm</td>
<td>5–7 μm/12–14 μm</td>
<td>5–7 μm/12–16 μm</td>
<td>3–4 μm/10–12 μm</td>
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<tr>
<td>Ribosomes very numerous rosettes</td>
<td>numerous rosettes</td>
<td>numerous; free in rosettes and on ergastoplasmic lamellae</td>
<td>few</td>
<td>numerous; free in rosettes and on ergastoplasmic lamellae</td>
</tr>
<tr>
<td>Ergastoplasmic lamellae</td>
<td>absent or very rare</td>
<td>rare</td>
<td>few definite; some part of the lamellae being granular</td>
<td>numerous</td>
</tr>
<tr>
<td>Mitochondria</td>
<td>rare</td>
<td>few</td>
<td>increased in number and size</td>
<td>well-developed</td>
</tr>
<tr>
<td>Golgi apparatus</td>
<td>small</td>
<td>small</td>
<td>slightly larger</td>
<td>enlarged</td>
</tr>
</tbody>
</table>

Macrophages containing debris were seen in higher numbers than in the controls.

Electron microscopic observations (fig. 3) showed two major features: an intense proliferation of large, round cells (hemocytoblasts) and increased numbers of mature reticulum cells. The large, round cells were 10–12 μm in diameter. They had a distinctive round shape which was a characteristic feature not found in the surrounding cells. Although similar to each other in appearance, related types of hemocytoblasts could be distinguished (table 1).

(a) Type I (fig. 4): The nucleus was large and spherical and aggregates of chromatin occurred along its membrane. The nucleolus was hypertrophied and was often attached to the nuclear membrane. Multiple nucleoli were occasionally seen. The nucleolonema was clearly visible. Some large, electron-dense granules, similar to the perichromatin granules described by Watson,14 were also seen in the nucleus. The cytoplasm was packed with free ribosomes grouped in rosettes; each rosette was composed of four to six ribosomes. One to three dense bodies were usually seen in the peripheral zone of the cytoplasm. Ergastoplasmic lamellae were either absent or minute. Mitochondria were small and oval and the Golgi apparatus consisted of a few small vesicles. The cell membrane was imbricated with those of neighboring cells (arrow, fig. 4).

(b) Type II: Very similar to Type I, but somewhat larger. The nucleus was slightly irregular in shape. The ribosomal rosettes in the cytoplasm were somewhat less numerous. The ergastoplasm was still very reduced. The Golgi apparatus was larger and the cell membrane exhibited the same features as before.

(c) Type III (fig. 5A): This cell had the basic features of Types I and II, but was slightly larger and elongated. The nucleus was also similar, but the nucleo-cytoplasmic ratio was decreased. The nucleolus was still very large and the chromatin was more irregularly scattered. A few distinct ergastoplasmic lamellae could now be clearly seen. Dense bodies were seen and the number and size of mitochondria were increased. The two main features of this cell were the well-developed Golgi apparatus with its numerous vesicles and the constant presence of one or more large cytoplasmic vacuoles.

(d) Type IV (fig. 5B): This cell was characterized by the presence of...
free cytoplasmic ribosomes together with a distinct ergastoplasm. The membranes of that structure were occasionally granular and first appeared close to the mitochondria. The mitochondria were numerous and large. The Golgi apparatus was well-developed with vesicles and flattened sacs. The large cytoplasmic vacuoles were apparently the result of active pinocytosis. The features of the nucleus were unchanged.

In addition to hemocytoblasts, many reticulum cells were present (fig. 1B). Detailed descriptions of this cell type have been given elsewhere. These cells had a well-developed cytoplasm which was in contact with collagen fibers in some areas. Their nuclei contained a medium sized nucleolus and an irregularly distributed chromatin. A moderate number of free ribosomes were seen throughout the cytoplasm. The ergastoplasmic membranes, smooth or granular, were poorly developed. The Golgi apparatus was of medium size and the mitochondria were elongated with easily seen cristae. The cell membrane had elongated protrusions which interdigitated with the surrounding cells.

Some of these cells were typical macrophages (fig. 1B) and contained phagocytized material which was electron-dense, irregularly shaped, and surrounded by a limiting membrane; smaller dense bodies with ferritin-like granules were also present together with occasional myelin figures. Undigested cellular debris was visible in the macrophages but it was impossible to recognize the origin of the ingested cell. The cells of this line were clearly more numerous than in a non-stimulated lymph node.

The lymphocytic population of these lymph nodes was, in general, similar to that previously described. Lymphoblasts and small lymphocytes were seen in moderate number and many medium sized lymphocytes were present. Their ergastoplasmic lamellae were reduced to a few flattened sacs and tubules with smooth or poorly granular membranes. The ribosomes were always seen irregularly distributed throughout the cytoplasm. Mitochondria were numerous and most often located at one side of the cell; they were also found in the indentation of the nucleus. The Golgi apparatus was poorly developed; very often, a diplosome was located near the center of this area. The nucleus contained densely packed chromatin; a medium sized nucleolus was often seen.

Second Set Reaction

By light microscopy, the sections were hypercellular and large secondary germinal centers were present. Numerous large, round, intensely basophilic cells were seen. Some of these cells were slightly smaller than those seen during the first set reaction and, although they often possessed a definite acroplasm, they did not have the distinctive morphologic features of plasma cells (fig. 1C).

Under the electron microscope, in addition to hemocytoblasts similar to those described during the first set reaction, a new type of cell with a highly developed endoplasmic reticulum was seen (fig. 6). This cell obviously be-
Fig. 4.—See legend, facing page.
MORPHOLOGIC RESPONSES TO HOMOGRAFTS

longed to the plasma cell line. Furthermore, all the transitional stages from plasmablasts to mature plasma cells were seen. Most of the cells of this line were young plasma cells and they were usually found in close association with highly vacuolated reticulum cells. The youngest recognizable plasma cells had a central, round nucleus and a few flattened ergastoplasmic lamellae in their cytoplasm. In the more advanced stage of their development, these cells had ergastoplasmic lamellae distributed throughout the cytoplasm except for the large Golgi area. The cisternae were often swollen with material somewhat less dense than the background cytoplasm. Most of the ribosomes were attached to the endoplasmic membranes. Mitochondria appeared ovoid, of variable size, and generally large with clearly visible cristae. The Golgi apparatus was distinctly seen and even prominent at times. Microbodies were frequently seen. The nucleolus was medium sized. In addition, mature plasma cells with classical morphologic features—large swollen cisternae containing cloudy material of medium contrast and eccentric nuclei, which rarely contained nucleoli—were seen. Russell bodies were not observed.

The peripheral membranes of both young and mature plasma cells were extensively interdigitated with the membranes of adjacent reticulum cells and showed the picture of active pinocytosis corresponding to the descriptions by Thiery of lymph nodes from hyperimmunized animals. The small and large lymphocytes had the same features as during the first set reaction. Eosinophilic proliferation was not seen during the first or second set reactions.

Non-grafted Animals Treated with 6-Mercaptopurine

The results in these animals were similar to those previously reported. On light microscopy, the tissues appeared shrunken in some areas and in others, a marked hyperemia was present. The cellular population was reduced; many large, mature reticulum cells were seen. On electron microscopic examination, the cellular appearance was very similar to that seen in animals retaining their homograft when given 6-mercaptopurine.

Animals Treated with 6-Mercaptopurine and Retaining Their Homograft

No proliferation of hemocytoblasts was seen in these animals. On light microscopy, the sections showed hypocellularity, shrunken tissues and marked hyperemia. Capillaries were prominent and were sometimes packed with erythrocytes. Reticulum cells were increased and many macrophages were present.

Fig. 4.—Hemocytoblast (greater enlargement of fig. 3). Ultra-thin section. Large round nucleus with prominent nucleolus (n) and evenly distributed chromatin along the nuclear membrane; finely granular interchromatinic area. In the cytoplasm, numerous rosettes of free ribosomes are evenly scattered (r); very few mitochondria (m); no Golgi apparatus is visible in this picture. \( \rightarrow \) = cytoplasmic protrusions imbricated with neighboring cells. X15,000. Inset in upper left details rosettes of ribosomes. X60,000.
Fig. 5.—See legend, facing page.
On electron microscopic examination, the large, round cells seen during the first set reactions were absent. No primitive, undifferentiated mesenchymal cells were seen (fig. 7); it was indeed difficult to find these cells even in a normal lymph node. Small lymphocytes with a rather dark cytoplasm and no visible nucleolus were prominent. Reticular cells, often packed with cellular debris very similar to ferritin-like particles, were seen, as well as formations reminiscent of lysosomes. The capillaries were prominent. Collagen fibrils were more easily seen in these lymph nodes than in those from untreated animals.

**Animals Rejecting Their Homografts despite the Administration of 6-Mercaptopurine**

By light microscopy, sections disclosed shrunken, but hypercellular tissue. There was striking hyperemia and many capillaries were seen. Large, round cells were present in the walls of these vessels. At times they formed groups in the vicinity of these capillaries. Small lymphocytes, which appeared to be migrating through the walls of these capillaries, were also found. Debris was seen in some of the cells lining these vessels, suggesting a phagocytic activity (fig. 1D).

On electron microscopy, the main feature was the proliferation of two types of cells: (a) large, round cells with an enlarged nucleus containing one or more hypertrophied nucleoli; the nuclear chromatin was clumped along its membrane (fig. 8A). Numerous free cytoplasmic ribosomes that did not form rosettes were present. Abnormal vacuoles were seen together with very dense, large mitochondria grouped in one area of the cell. Ergastoplasmic lamellae were either absent or very reduced and, when present, were smooth. (b) Unusual reticulum cells with an irregular nucleus, a medium sized nucleolus and very clear interchromatinic area (fig. 8B). The cytoplasm of these cells was very dense and contained a moderate number of free ribosomes. Although endoplasmic lamellae were numerous, they were devoid of ribosomes and appeared to form vesicles or bundles of tiny fibrils. The Golgi area was enlarged. Structural defects were frequently visible in the dense, elongated mitochondria. Increased numbers of macrophages packed with debris were also seen.

**DISCUSSION**

The large, round cells we observed in these experiments have distinctive features that permit their differentiation from the primitive mesenchymatous

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Fig. 5.—Hemocytoblasts. Ultra-thin section. A. Type III: Same features of the nucleus as previously seen. In the cytoplasm: rosettes of free ribosomes, definite granular ergastoplasmic lamellae (e) and numerous mitochondria (m). X10,000. B. Type IV: Large ovoid nucleus with the same features as previously described except for a somewhat less prominent nucleolus. In the cytoplasm: rosettes of free ribosomes, numerous granular ergastoplasmic lamellae (e), large mitochondria (m) and a very prominent Golgi apparatus (g). X10,000.
Fig. 6.—Second set reaction (draining lymph node). Ultra-thin section. Presence of a typical hemocytoblast (1) and of plasma cells (2) among the lymphocytes (3); tangential section of a capillary (4). X5000.
Fig. 7.—Animal retaining its homograft when treated with 6-MP. Ultra-thin section. Uniform lymphocytic population (1); dense appearance of the lymphocytes; several capillaries are visible (2); collagen fibrils (3). X5000.
Fig. 8.—See legend, facing page.
MORPHOLOGIC RESPONSES TO HOMOGRAFTS

127
cells seen in normal lymph nodes (table 1). Similar cells arise in vitro and in vivo after many kinds of antigenic stimulation. We have used the word "hemocytoblast" for them. They have been called by others: "transitional cells," "lymphoblasts," "lymphogonia," "large lymphoid cells," "acute splenic tumor cells," "germinoblasts" and "immunoblasts." We have used the word "hemocytoblast" in the sense given it by the Prague Conference of 1960.

The features that distinguish these cells from undifferentiated mesenchymal cells are their large, round nuclei, their single or multiple hypertrophied nucleoli and their very numerous free ribosomes which, in our observations, were always grouped in rosettes. Policard believes that the ribosomes first appear free and then become grouped in rosettes, but we did not observe this. Since much of the cytoplasmic RNA exists as ribosomes, it is probable that antibodies are synthesized on them. Indeed, Feldman showed the presence of antibody to sheep red cells in the ribosomal fraction of lymph node homogenates derived from hyperimmunized rabbits. The presence in hemocytoblasts of ribosomes arranged in clusters or rosettes is consistent with the recently obtained evidence that protein synthesis occurs on polyribosomes, which are thought to represent groups of ribosomes bound together by strands of messenger RNA.

Medium sized lymphocytes containing free ribosomes which were not grouped in rosettes, numerous mitochondria, distinct Golgi apparatus and centrioles, were very numerous during the first set reaction. We did not see mitotic figures among them. By morphologic criteria, these cells could certainly be considered "activated," suggesting their possible role in transplantation immunity as has been postulated by many authors. It has been claimed that direct transformation of lymphocytes into large pyroninophilic cells can take place. However, even in a "purified" pool of lymphocytes, one cannot exclude the presence of a small but very active contingent of undifferentiated cells which may transform into immunologically competent cells. Nevertheless, Gowan's studies strongly suggest that the small lymphocyte is essential for initiation of the primary immune response.

Macrophage activity was very evident in the present studies. A moderate but definitely increased number of macrophages containing debris was present during the first and second set reaction. Phagocytosis in the germinal

Fig. 8.—Animals rejecting their homografts in spite of injections of 6-MP. Ultra-thin section. A. Large, round cell with an enlarged nucleus containing one or more hypertrophied nucleoli; chromatin clumped along the nuclear membrane; in the cytoplasm, numerous free ribosomes which do not form rosettes; abnormal vacuoles; large very dense mitochondrias with well-visible cristae, grouped in one area of the cell. X20,000. B. Unusual reticular cell, irregular nucleus, very light interchromatinic area; very dense cytoplasm with a moderate number of free ribosomes, many vesicles bundles of tiny fibrils and enlarged Golgi area; a few very dense and elongated mitochondrias, some of which show structural defects at a higher magnification. X20,000.
centers of normal and antigenically stimulated lymph nodes has been noted for many years\textsuperscript{7-13,49,50} and some authors have considered the debris to represent digested lymphocytes.\textsuperscript{51} We were unable to identify the exact nature of this material. The role of macrophages in the immune response is not entirely clear. They might contact antigens directly, transform them, and perhaps pass them on to either other macrophages or to some other antibody forming cells.\textsuperscript{10,52,53} Alternatively, macrophages might engulf cells injured by absorption or ingestion of antigens.\textsuperscript{10,53} Burnet\textsuperscript{59} has suggested that macrophages might take up the excess of antigen which otherwise inhibits antibody-producing cells. Stetson\textsuperscript{55} proposed that macrophages store most of the antigen until its release is triggered by a “booster” injection. These ideas are supported by the observation that mice immunized with horse ferritin store antigen-antibody complexes as “phagosomes” in phagocytic reticulum cells.\textsuperscript{56} Recently, Fishman\textsuperscript{57} has shown by in vitro experiments that macrophages ingest antigen and release a stimulus for antibody formation which is probably some form of RNA.

A striking increase in macrophages was seen in the 6-mercaptopurine “tolerant” and “resistant” animals. In these animals the macrophages were even more packed with debris. It is possible that under normal circumstances, enzyme-rich phagocytic reticulum cells take up cellular debris originating from cells lysed by contact with antigens.\textsuperscript{53} Treatment with 6-MP might result in many fragile cells which could be ingested by reticulum cells. On the other hand, it is also possible that the strikingly increased number of macrophages packed with debris was due to impairment of some enzyme-dependent process within the macrophages.

Only a few capillaries were seen in our material; several of them were found in an animal rejecting its graft despite administration of 6-mercaptopurine. The finding, in control animals, of typical endothelial cells in intimate association with large, round primitive cells in the walls of these vessels, was in line with the observations of many authors.\textsuperscript{7-13,58,59} We did not see mitosis in the cells lining the walls of such activated capillaries.\textsuperscript{55} These vessels may participate in molding lymphoid tissues into avenues of migration for small lymphocytes. Policard\textsuperscript{8} proposed that antigens might be transported through the vascular wall by means of either small lymphocytes or endothelial cells. If small lymphocytes can initiate antibody production, but represent a heterogeneous population in the sense envisaged by Burnet,\textsuperscript{54} then their re-circulation via the blood stream or the thoracic duct\textsuperscript{60,61} should allow relatively small numbers of “committed” cells a better chance to seed the lymphatic system.

Of particular interest was the occurrence of cells of the plasma cell line during the second set homograft reaction. These cells were seen unequivocally only with the electron microscope. The hemocytoblasts seen during the first set reaction and the young plasma cells seen during the second set reaction were morphologically related, because discrete but continuously developing ergastoplasmic lamellae were seen throughout the development of these cells. These findings are of note in view of the evidence that circulating antibodies are formed during the homograft reaction.\textsuperscript{62-64}
Young plasma cells present around reticulum cells which contained numerous vacuoles and showing signs of intense pinocytosis were frequently seen. Many workers have studied the possible formation of plasma cells from primitive reticulum cells. Our observations strongly suggest that the plasma cell is derived from the large, round primitive cell, the hemocytoblast. However, it cannot be stated from these studies if the hemocytoblast itself originates from undifferentiated mesenchymatous cells, adult reticulum cells or from lymphocytes.

Since the draining node is morphologically normal 25 days after the application of a skin homograft, most of the hemocytoblasts seen in the first set reaction probably enter a quiescent phase. The principal function of these cells in this phase could be the retention of immunologic memory. A further antigenic stimulus to the dormant hemocytoblasts might then trigger their final development into immunologically active cells. It is likely, however, that together with these two types, various other cells are involved in the immune response. Lymphocytes of all types, reticulum cells (including histiocytes, monocytes and fixed phagocytes), and unclassifiable intermediate and primitive cells may participate directly or indirectly. However, in contrast to experiments employing defined antigens, eosinophilia was not seen in our material.

The general features of the chemical suppression of immunity by 6-mercaptopurine have been described elsewhere. Since then, there has been an increasing interest in this drug in respect to transplantation immunity. The present results confirm our previous work. Most of the animals, when given the drug, retained their homograft. There was a complete absence of hemocytoblasts under these conditions and numerous pycnotic lymphocytes and macrophages packed with debris were found. In one animal that rejected its graft despite the administration of 6-mercaptopurine, there was an intense growth of atypical and unusual hemocytoblasts and a striking number of highly atypical reticulum cells which we have never seen before. They seem to correspond to the cells we termed “6-MP-resistant hemocytoblasts” in our previous study.

Our observations indicate the unity of the immunologic reaction at the cellular level. Whether the antigen is a purified protein, or bacteria, or a skin homograft, there is a stimulus for the proliferation of hemocytoblasts, which eventually develop into plasma cells. The rate of this development and the ancillary participation of other cell types (e.g., the eosinophil) may vary from antigen to antigen, but the fundamental cellular process appears more or less the same, whatever the nature of the inciting antigen.

**SUMMARY**

An electron microscope study of the morphologic responses of the rabbit lymph node after skin homografting is described. The results demonstrate: (a) the appearance of large, round, ribosome-rich cells—hemocytoblasts—during the first set reaction; (b) a striking proliferation of plasma cells during the second set reaction; (c) the absence of hemocytoblasts in the “6-MP tolerant” animals, and (d) large numbers of two types of unusual cells in
the "6-MP resistant" animal. The presence of increased numbers of plasma cells in the second set homograft reaction is in agreement with the findings of circulating antibodies in transplantation immunity and indicates the fundamental unity of the dynamics of the cellular response in various immunological phenomena.

**SUMMARIO IN INTERLINGUA**

Es describite un studio de microscopia electronic del responsas morphologic del nodo lymphatic del conilio post homograffage cutanee. Le resultatos demonstra (a) le apparition de grande, ronde cellulas ric in ribosomas (hemocytoblastos) durante le reaction a un prime graffo, (b) un frappante proliferation de plasmocytos durante le reaction a un seconde graffo, (c) le absentia de hemocytoblastos in animales tolerante pro 6-mercaptopurina, e (d) grande numeros de duo typos inusual de cellulas in animales resistente contra 6-mercaptopurina. Le presentia de augmentate numeros de plasmocytos durante le reaction a un secunde graffo es de accordo con le constatation de circulante anticorpore in immunitate de transplantation e indica le unitate fundamental del dynamica del responsa cellular in varie phenomenos immunologic.

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**REFERENCES**

14. Watson, M. L.: Observations on a granule associated with chromatin in the
MORPHOLOGIC RESPONSES TO HOMOGRAFTS


37. Feldman, M., Elson, D., and Glober-
sen, A.: Antibodies in ribonucleo-
1960.
Electron microscopy studies of ribo-
somal clusters synthesizing hemoglo-
39. Wissler, R. W., Fitch, F. W., La Via,
M. F., and Gunderson, C. H.: The
cellular basis of antibody formation.
J. Cell. & Comp. Physiol. 50(suppl.
40. Holub, M.: Potentialities of the small
lymphocyte as revealed by homotrans-
plantation and autotransplantation ex-
41. Roberts, J. C., Dixon, F. J., and Weigle,
W. O.: Antibody producing lymph
node cells and peritoneal exudate
cells. Morphological studies of transfers
to immunologically inert rabbits. 
A.M.A. Arch. Path. 64:324, 1957.
42. Porter, K. A., and Cooper, E. H.: Trans-
formation of adult allogenic small
lymphocytes after transfusion into
newborn rats. J. Exper. Med. 115:
43. Mims, C. A.: Experiments on the origin
and fate of lymphocytes. Brit. J.
M., and Egahl, R. H.: Transplanta-
tion antigenicity of lymphoid cells
1963.
45. Marshall, W. H., and Roberts, K. B.: 
The growth and mitosis of human
small lymphocytes after incubation
with a phytoagglutinin. Quart. J.
46. Berman, L., and Stulberg, C. S.: Pri-
mary cultures of macrophages from
normal human peripheral blood. Lab.
47. Schooley, J. G., and Berman, I.: 
Morphologic and autoradiographic ob-
servations of H3 thymidine labeled
thoracic duct lymphocytes cultured 
48. McGregor, D. D., and Gowans, J. L.: 
The antibody response of rats de-
pleted of lymphocytes by chronic
drainage from the thoracic duct. J.
49. Dunn, T. B.: Normal and pathological
anatomy of the reticular tissue in labo-
ratory mice. J. Nat. Cancer Inst. 14:
1281, 1954.
50. Ehrlich, W. E., and Harris, T. N.: The
formation of antibodies in the popli-
teal lymph node in rabbits. J. Exper.
Med. 76:335, 1942.
51. Trowell, O. A.: Some properties of
lymphocytes in vitro and in vitro. 
52. Dixon, F. J., Weigle, W. O., and Rob-
erts, S. C.: Comparison of antibody
responses associated with the transfer
of rabbit lymph nodes, peritoneal
exudate and thymus cells. J. Im-
munol. 79:56, 1957.
53. Speirs, R. S.: Antigenic material; per-
sistence in hypersensitive cells. 
 Theory of Acquired Immunity. Cam-
bridge, Cambridge University Press,
1959.
55. Stetson, C. A., and Demopoulos, B.: 
Reaction between skin homografts 
and specific immune sera. Ann. New 
56. Tranzer, J. P., Porte, A., Kempf, J., 
and Fruhling, L.: Différenciation pla-
smocytaire d’éléments réticulaires après 
stimulation antigénique par de la fer-
ritine hétérologue. Compt. rend. Acad.
57. Fishman, M., and Adler, F. L.: Anti-
body formation initiated in vitro. II.
Antibody synthesis in x-irradiated re-
cipients of diffusion chambers contain-
ing nucleic acid derived from macro-
phages incubated with antigen. J.
58. Schulze, W.: Untersuchungen über die
capillaren und post capillaren venen
Rec. 135:421, 1925.
59. Smith, C., and Henon, B. K.: Histol-
ogical and histochemical studies of the
high endothelium of post capil-
lar veins of the lymph node. Anat. 
60. Gowans, J. L.: The fate of parental
strain small lymphocytes in F1 
hardt, W. O.: Cellular migration streams in the haemopoietic system. 
In The Kinetics of Cellular Proliferation. F. Stohlman, Jr., ed. New York, 
Grune & Stratton, 1959, p. 69.
62. Amos, D. B., Gorer, P. A., Billingham, 
aspects of the immune response to 
64. Kretschmer, R. R., and Pérez-Tamayo, 
R.: The role of humoral antibodies 
in rejection of skin homografts in 
rabbits. I. Passive transfer of isoi-
immune serum to conditioned hosts. J. 
65. Leduc, E. H., Coons, A. H., and Conol-
ly, J. M.: Study on antibody produc-
tion. II. The primary and secondary 
responses in the popliteal lymph node 
of the rabbit. J. Exper. Med. 102: 
61, 1955.
66. Rebuck, J. W., and Logrippo, G. A.: 
Characteristics and interrelationships 
of the various cells in the reticulum 
cell, macrophage, lymphocyte and 
plasma cell series in man. Lab. In-
67. Makinodan, T., and Albright, J. F.: 
Cellular variation during the immune 
response: one possible model of cel-
lular differentiation. J. Cell & Comp. 
Physiol. 60 (suppl. 1):129, 1962.
68. Nossal, J. G. V., and Makela, O.: Auto-
radiographic studies in immune re-
sponses. I. The kinetics of plasma cell 
proliferation. J. Exper. Med. 115:209, 
1962.
69. Speirs, R. S.: Advances in the knowl-
edge of the eosinophil in relation to 
70. Litt, M.: Studies on experimental eosin-
ophila. V. Eosinophils in lymph nodes 
of guinea pigs following primary anti-
genic stimulation. Am. J. Path. 42: 
529, 1963.
71. Schwartz, R. S., and André, J. A.: The 
chemical suppression of immunity. In 
Mechanism of Cell and Tissue Dam-
age Produced by Immune Reactions. 
P. Miescher, and P. Grabar, eds. 
Basel, Bruno Schwabe & Co., 1962, 
p. 385.

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