Rates of Proliferation and Interrelationships of Cells in the Mesenteric Lymph Node of the Rat

By William O. Rieke, Ruth W. Caffrey and N. B. Everett

THE STRUCTURE and cellular composition of lymph nodes in various species have been thoroughly studied with both the light1-5 and the electron microscope.6-8 These studies have adequately served to describe and classify the cells in the lymph node and have led to general agreement on the appearance and location of the various cell lines. From these studies have also come descriptions of structural transitions which underlie most of what is currently believed relative to the origin, developmental sequence and interrelationships of the various cell types. Thus, since Downey and Weidenreich1 first reported finding cells which morphologically appeared to be intermediates between fixed reticular cells and lymphocytes, many have believed that lymphocytes arise from reticular cells.2,7,8,9,10 Similar findings and reasoning have led to the conclusion that plasma cells also develop from reticular cells7,10,11. Experiments designed to test the hypothesis that lymphocytes arise from reticular cells have often depended either on the physical association of reticular cells and lymphocytes or on the finding of apparent transition forms between these cell types.14-16

Radioautography with tritiated thymidine labeling provides a direct approach to the problems of cellular proliferation and interrelationships. It is the purpose of the present paper to report developmental rates and some possible interrelationships of the various cell types in the mesenteric lymph node of the rat as shown by single and multiple injections of tritiated thymidine.

MATERIALS AND METHODS

Male rats of either the Lewis or Sprague-Dawley strain were injected with tritiated thymidine (TTH, Schwarz Bio Research, Orangeburg, N. Y.), specific activity either 1.9 or 6.05 c./mM) according to one of the schedules which follow.

Injection Schedules and Their Purposes

Group I. Single injection. Nine rats weighing 141 to 143 Gm. were injected intravenously with 1 µc./Gm. body weight of TTH and were sacrificed at 15 minutes; 4 and 8 hours; 1, 2, 3, 5, 10 and 14 days after injection. The animal sacrificed 15 minutes after TTH allowed the identification of cells in DNA synthesis and capable of division. The per cent of each cell type labeled 15 minutes after TTH indicates the fraction of the cells' generation time spent in DNA synthesis. The DNA synthetic times of the various populations were assumed to be approximately comparable, and, therefore, differences in per cent label were indicative of differences in generation times of the populations. Animals sacrificed at later intervals provided for following the labeling patterns resulting from cell division and maturation.
Group II. Intensive injections. Six rats weighing 150 to 250 Gm. were injected intraperitoneally with 1 μc./Gm. body weight of TTH every 4 hours for 36 hours. All were sacrificed 4 hours after the last injection. This schedule assured the labeling of all cells formed during the 36-hour period of injections, and, therefore, the increment in the percent of any cell type labeled was a direct measure of its rate of proliferation.

Group III. Cumulative injections. Eleven rats each with an initial weight of approximately 65 Gm. were injected intraperitoneally with 0.75 μc./Gm. body weight of TTH every 6 hours for 1 to 11 days. One animal was sacrificed 6 hours after the fourth injection and the remainder were sacrificed daily with the autopsy in each case timed for 6 hours after the last injection. This series provided a period of several days during which the increment of labeling, and hence the rates of proliferation of the various cell types, could be serially studied.

Group IV. Multiple injection interval. Six rats weighing 60 Gm. each were given a total of 12 intraperitoneal injections of 0.5 μc./Gm. body weight of TTH during a 16-day period. One animal was sacrificed 4 hours after the last injection and the remainder were sacrificed at 2, 4, 6 and 8 weeks after TTH. This schedule provided for labeling a large percent of slowly proliferating cells and allowed investigation of the relationship of this group to rapidly proliferating cells.

A portion of the mesenteric lymph node chain from each rat was teased and smeared for radioautography as previously described.17 Another portion was fixed in Zenker's fluid, embedded in methacrylate and sectioned at 1 μ. Mounted sections were carried in the Dominici staining technic through the Orange C step prior to radioautographic processing. Staining the sections in this manner was found effective in complexing and inactivating the heavy metals in Zenker's fixative which otherwise may cause chemical reduction and artifacts in the emulsion during radioautography. After radioautographic processing, the sections were further stained with nuclear-fast red and finally counterstained with MacNeal's tetrachrome. Radioautographic exposures of 5 and 8 weeks were made.

RESULTS

The labeling patterns of the various cell types are reported separately below and summarized for all types in table 1. To facilitate identification, a brief description of the morphologic characteristics of the several varieties of cells is presented together with the labeling patterns. Only those cells which may be identified clearly from the characteristics noted are included in the following data.

Reticular Cells (Including Macrophages)

These cells are well represented in both cortex and medulla of the lymph node and although they vary widely in size and shape, are easily recognized. In smear preparations they exhibit round, oval or irregular nuclei, which are 8 to 15 μ across and contain a delicate chromatin network with one or more pale blue nucleoli (fig. 2). Although the nucleus is bounded by a definite nuclear membrane, the homogeneous, grey-lilac staining cytoplasm is not as sharply limited. The smearing procedure commonly disrupts the cytoplasm leaving the cells to appear as bare nuclei. In tissue sections where cell size is approximately one-half of what it is in smears, reticular cells are found with widely varying nuclear shapes but always possess a pale blue chromatin network and usually one distinct centrally placed nucleolus (fig. 3). The cytoplasm is usually abundant, though pale, and is not sharply limited.
Table 1.—Per Cent of Cells Labeled in Mesenteric Lymph Node

<table>
<thead>
<tr>
<th>Animal Groups</th>
<th>Single Injection</th>
<th>Intensive Injection</th>
<th>Cumulative Injection</th>
<th>Multiple-Injection Interval</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>15 min.</td>
<td>72 hr.</td>
<td>36 hr.</td>
<td>5 days</td>
</tr>
<tr>
<td>Number of animals*</td>
<td>4</td>
<td>2</td>
<td>6</td>
<td>2</td>
</tr>
<tr>
<td>Reticular cells</td>
<td>6</td>
<td>0</td>
<td>12</td>
<td>50</td>
</tr>
<tr>
<td>Hemohistioblasts and histioblasts</td>
<td>50</td>
<td>0</td>
<td>100</td>
<td>100</td>
</tr>
<tr>
<td>Large lymphocytes</td>
<td>33</td>
<td>65</td>
<td>60</td>
<td>60</td>
</tr>
<tr>
<td>Medium lymphocytes</td>
<td>0</td>
<td>1-2</td>
<td>3</td>
<td>23</td>
</tr>
<tr>
<td>Small lymphocytes</td>
<td>75</td>
<td>weak</td>
<td>100</td>
<td>100</td>
</tr>
<tr>
<td>Plasmablasts</td>
<td>0</td>
<td>50</td>
<td>50</td>
<td>100</td>
</tr>
<tr>
<td>Proplasmacytes</td>
<td>0</td>
<td>50</td>
<td>50</td>
<td>100</td>
</tr>
</tbody>
</table>

*In addition to the 31 animals shown, another 30 animals of different post-TTH intervals have been analyzed and found to fit the patterns presented in the table.

from neighboring cells. Free or fixed macrophages have morphologic characteristics similar to reticular cells and are included in this group.

The labeling patterns of reticular cells are strikingly different from any other cell type in lymph node. Immediately following a single injection of TTH, only a small per cent (6 per cent) are labeled. These are intensely labeled, and several weeks later there are still numerous intensely labeled cells present. At longer intervals after a single injection, a marked and unusual variation in labeling intensity appears with some reticular cells being almost black with silver grains while others evidence only three to five grains. Most of the labeled reticular cells at longer post-TTH intervals are of the weakly labeled variety. Because phagocytosis of labeled small lymphocytes (figs. 4 and 5) is commonly observed in reticular cells and has been reported from this laboratory before, it may be that reutilization of this material gives origin to the weak labeling noted.

Only when TTH is administered over a period of many days are high percentages of reticular cells labeled. For example, cumulative TTH injections given throughout a 10-day period label 85 per cent of these cells, but even then, only one-third are intensely labeled. It is to be recognized that during such a period injected animals are adding approximately one-third to their body weight and thus the intensely labeled reticular cells may represent nothing more than the increment in the population expected with growth.

When TTH is administered for a period of days and then discontinued, high percentages of labeled reticular cells persist for weeks (figs. 6 and 7). In such multiple injection animals, 61 per cent of reticular cells are labeled 8 weeks after TTH, and although labeling intensity gradually decreases with time, many cells are still intensely labeled. The reticular cells labeled at long post-TTH intervals are morphologically indistinguishable from those labeled at short post-TTH intervals.

Together, these observations indicate that reticular cells are a slowly
Fig. 1.—The disappearance of radioactivity in hemohistioblasts (including histioblasts) is illustrated, and from this the generation time is calculated to be approximately 12 hours.

proliferating population which are perhaps formed in relation to body growth. A thorough search of tissue sections noting reticular cells of all descriptions and locations (round, oval or stellate in the cortex or medulla, isolated or surrounding capillaries) and all functional activities (phagocytic or not) has failed to produce evidence that any group of these cells is rapidly proliferating.

**Hemohistioblasts (Histioblast)**

Although fewer in number than reticular cells, these cells are also found both in the cortex and medulla of lymph nodes. They are always associated with developing lymphocytes. Together with the plasmablast, they constitute the largest cells present and in smears often have oval nuclei with diameters of approximately 10 μ x 15 μ (fig. 8). The nucleus is not bounded by a distinct nuclear membrane and the chromatin strands it contains are intermixed with irregular areas of basophilic-staining material usually without a sharply defined nucleolus. The cytoplasm appears stringy, is moderately basophilic and has irregular borders. These cells are easily damaged and
in smears are most often found at the end or tail where they are surrounded by other cell types.

In tissue sections, hemohistioblasts (fig. 9) may be identified by their large oval nucleus with a dark nucleolus and by their abundant basophilic cytoplasm which has definite boundaries and contains numerous white granules (probably mitochondria).
As will be noted in the Discussion, the hemohistioblast as described here is believed to be functionally similar to the cell which Ferrata called by this name. Because of this, Ferrata's term is retained even though the morphology of the present cell is not the same as the hemohistioblast as Ferrata described it. The present hemohistioblast resembles what others have called a hematopoietic reticulum cell, while Ferrata's hemohistioblast cannot be distinguished from the general grouping of reticular cells.

The histioblast (fig. 10) is sometimes distinguished from the hemohistioblast on the basis of its cytoplasm being less basophilic. Because the labeling patterns of these two cells are identical, they are here considered together.

Hemohistioblasts incorporate tritiated thymidine readily and show very consistent labeling patterns. Fifteen minutes after TTH, 50 per cent of them are labeled and all of them are labeled by 36 hours of intensive TTH injections. Seventy-two hours after a single TTH injection, no cell of this type is labeled with more than three grains, and 2 weeks after multiple injections there is no labeling at all. During cumulative injections, their division rate is such that their labeling intensity remains relatively constant while that of reticular cells progressively increases. Together, these observations indicate that hemohistioblasts are rapidly proliferating cells, and this conclusion is supported by the findings from grain counts (fig. 1) which indicate that their generation time is approximately 12 hours.

Lymphocytes

Found in all parts of the lymph node, lymphocytes have a high nuclear-cytoplasmic ratio and are usually round but occasionally irregular. They are commonly divided into large, medium and small. A previous report from this laboratory suggests that in smears, large cells include those with nuclear diameters of 10 µ x 11 µ or greater; that medium cells have nuclear diameters less than large but greater than 7 µ x 8 µ; and that small cells include all

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All photomicrographs are of smear preparations or of one micron sections of the mesenteric lymph node of the rat.

Fig. 2.—The typical morphology of reticular cells (center) in smears is shown. Note the indistinct cytoplasmic boundaries.

Fig. 3.—Reticular cells (r), plasmablasts (p) and more mature members of the plasma cell series are seen in a plasma cell islet of the medulla of a lymph node.

Fig. 4.—A labeled phagocytized small lymphocyte and another partially digested cell are seen in this radioautograph of a reticular cell taken from an animal which had received H3-thymidine injections every 4 hours for 36 hours.

Fig. 5.—A labeled phagocytized small lymphocyte and other unlabeled inclusions are seen in this radioautograph of a reticular cell (r) in a tissue section taken from the same animal as in fig. 4.

Fig. 6.—A labeled reticular cell (left) and small lymphocyte (right) are shown in this radioautograph from an animal which had received multiple injections of H3-thymidine 24 days before autopsy. Occasional medium lymphocytes and these cells constitute the major types labeled at long post-thymidine intervals.

Fig. 7.—A radioautograph from the same animal described in fig. 6 illustrates the persistence of radioactivity in reticular cells long after dividing elements [plasmacytes (p)] have diluted out their label.
lymphocytes with nuclear measurements of $7 \mu \times 8 \mu$ or less. It is to be noted that within any of the three groups of lymphocytes, several different appearing types may be found. In general, these subtypes form a continuous series (figs. 11–13) grading from a round cell with a centrally placed nucleus surrounded by a narrow rim of very basophilic cytoplasm to an irregular cell with an eccentric nucleus containing nucleoli and surrounded by abundant...
pale-staining cytoplasm. In the case of large lymphocytes, the cells at either end of this series have been called lymphoblasts (or hemocytoblasts) and reticular lymphocytes respectively. In the medium and small classes, the same series of types appear as described for the large class, but small lymphocytes have a further subtype which shows essentially no cytoplasm and has clumped chromatin. In tissue sections it is difficult to recognize the various subtypes, but the major classes of lymphocytes are easily identified by their size, distinct cell outline, and the presence of a single acidophilic core in the nucleus. Because no differences were found in the labeling patterns of the subtypes in a given class, lymphocytes are here considered in the groups of large, medium and small.

The labeling pattern of large lymphocytes is identical to that of hemohistioblasts (table 1) and thus large lymphocytes form another group of rapidly proliferating cells. The difference in morphology between lymphocytes and hemohistioblasts, plus the presence of large lymphocytes in thoracic duct lymph where hemohistioblasts do not occur, make it very probable that these are truly separate cells. At the same time, similarities in both the percent label and intensity of labeling render it likely that they belong to the same cell line with large lymphocytes probably deriving from hemohistioblasts.

It has previously been noted that in addition to arising by division of large lymphocytes, medium lymphocytes also may derive from long-lived small lymphocytes which have enlarged as a consequence of antigenic stimulation. It is, therefore, not surprising that the percent of labeled medium lymphocytes is influenced by the growth and the state of health of an animal and varies considerably. One-third of these cells may be labeled 15 minutes after a single injection of TTH (table 1) and, therefore, medium lymphocytes do synthesize DNA in preparation for mitosis. However, some medium lymphocytes remain unlabeled after 11 days of cumulative TTH injections and differ markedly in this respect from large lymphocytes, all of which are labeled following 36 hours of TTH administration. Medium lymphocytes differ further from large lymphocytes in that while most divide and dilute out their radioactivity, a few remain labeled for long post-thymidine intervals (2 percent at 8 weeks). No large cell retains label for more than a few days. It is felt that these observations give support to the concept of a dual origin

Fig. 8.—A hemohistioblast (at the bottom) is shown along with a large lymphocyte (lymphoblast or hemocytoblast) at the top.

Fig. 9.—The appearance of interphase and dividing hemohistioblasts in tissue sections (h) are illustrated.

Fig. 10.—Although the histioblast shown differs from the hemohistioblast (fig. 8) by having stranded chromatin and less basophilic cytoplasm, its H3-thymidine labeling pattern is identical to that of the hemohistioblast.

Figs. 11–13.—Three medium lymphocytes (1) are shown to illustrate the variety of appearances within the class of medium. A similar series of forms is found in large and small lymphocytes. H3-thymidine labeling patterns are the same for all members of the series within any given class.
of this class and indicate further that within the class the majority of cells are relatively rapidly proliferating.

No small lymphocyte is labeled 15 minutes after TTH and it has been shown that small lymphocytes become labeled only as a consequence of their being formed by the division of labeled larger precursors. Only a small per cent (1.5 per cent) of these cells are labeled in lymph node 72 hours after a single injection of TTH and high percentages can be labeled only by administering TTH over a period of days. It has previously been reported that there are two types of small lymphocytes in the rat with respect to circulating life span: one (short-lived cells) which persists approximately 5 to 6 days, and another (long-lived cells) which has some members that circulate for at least as long as 6 months. That both of these types are seen in lymph node is evidenced from the multiple injection interval series (table 1). Here a rapid decrease in labeled small lymphocytes during the first 2 weeks after TTH is followed by a very gradual decrease during the ensuing several weeks. Because, as noted above, the appearance rate of labeled small lymphocytes in lymph node is slow, it is believed that the majority of these small cells belong to the long-lived population. This contrasts to the appearance rate of labeled small cells in thymus where the evidence indicates that the majority of small thymocytes belong to the short-lived population.

**Plasmablasts and Proplasmacytes**

These cells are found in the medullary cords often surrounded by groups of cells which contain all stages of the plasma cell series. In smears, the plasmablast is a large oval cell with an oval nucleus measuring 12 μ x 15 μ or more and containing basichromatin. The cell borders are distinct and the cytoplasm is highly basophilic except for a distinct paranuclear white area (fig. 14). In tissue sections, a plasmablast (fig. 3) is easily distinguished by its large size, oval outline, basophilic cytoplasm with its pale paranuclear area, and by the presence of significant quantities of acidophilic material in the nucleus.

Proplasmacytes are smaller than plasmablasts, have an irregular or polygonal outline and exhibit a small to moderate amount of intensely basophilic cytoplasm (fig. 15). They contain basichromatin and in tissue sections have acidophilic material in their nuclei.

Plasmablasts and proplasmacytes show similar labeling patterns and appear to be the most rapidly proliferating cell line in lymph node. Seventy-five per cent of these cells are labeled 15 minutes after a single TTH injection and 100 per cent are labeled by 36 hours of intensive TTH injections. The labeling intensity of these cells is greater than that of hemohistioblasts and lymphocytes (fig. 16); yet their division rate is so rapid that 72 hours following a single TTH injection only an occasional weakly labeled cell is found. Comparisons with the per cent label and generation time of the hemohistioblast lead to the conclusion that the generation time of plasmablasts is approximately 9 hours. This agrees well with a report by Nossal and Mäkelä who calculated the generation time to be 12 hours or less.
Plasma Cells

The morphology of mature plasma cells is sufficiently appreciated to make further description unnecessary. Like small lymphocytes, mature plasma cells are not labeled 15 minutes after TTH and become radioactive only as they arise by division of labeled precursors. Unlike small lymphocytes, however, 50 per cent of plasma cells are labeled 72 hours following a single TTH injection and 100 per cent are labeled by 5-day cumulative injections. In the same lymph node, only 23 per cent of small lymphocytes are labeled by 5 days of injections. Mature plasma cells differ further from small lymphocytes in that their life in lymph node is less than 2 weeks. These data indicate that plasma cells comprise a non-dividing, short-lived population which can be entirely renewed in not more than five days.

Miscellaneous Cells

An occasional fat cell or capillary endothelial cell is labeled in the first day following a single injection of TTH, but significant numbers do not label even with intensive TTH injections given for a few days. Only in multiple-injection-interval animals which received TTH over a period of 16 days are as many as 10 per cent of these cells labeled. A similar labeling pattern is seen in mast cells. These are best studied in tissue sections because they do not tolerate smearing procedures well. Fat cells, endothelial cells and mast cells may all be considered as elements which normally proliferate slowly.

Discussion

The present study allows some observations relative to interrelationships of the various cell lines in lymph node. Particular interest centers on the reticular cell, for as noted in the introduction there exists a substantial body of morphologic evidence which implicates the reticular cell as a stem cell. Because there are different concepts of the role and meaning of a stem cell, it is here defined as an undifferentiated cell which by mitosis is capable of producing both its own kind and cells of at least one different type. The present results based on the generally accepted hypothesis that DNA synthesis evidences impending cell division clearly indicate that many, and possibly all, reticular cells are not rapidly proliferating and do not demonstrate the labeling patterns expected of the stem cells of the rapidly renewing populations. At a time (short post-TTH intervals) when many or all of the hemohistioblasts, plasmablasts, proplasmacytes and large lymphocytes are labeled, very few (6 to 12 per cent) reticular cells are labeled (fig. 17). During the first 3 days after TTH when the rapidly proliferating lymphocytic and plasmacytic elements progressively dilute their radioactivity by division, the labeling intensity of the few reticular cells which are radioactive does not change significantly. Moreover, at a time when many reticular cells are labeled (long post-TTH intervals), there is no label at all in the rapidly proliferating cells just mentioned.

It is recognized that reticular cells as grouped here may not be functionally homogeneous and that they have commonly been divided into phagocytic
and non-phagocytic types. It is possible that a small fraction of one of these types may behave as stem cells. If this be true, however, it has not been possible to identify this subgroup by differences in labeling pattern, morphology or location. For example, the reticular cells in germinal centers, in plasma cell islets, or surrounding capillaries all are morphologically similar and are
most frequently not labeled when all surrounding elements are radioactive (fig. 17).

Finally, there is a possibility that a few reticular cells may modulate or transform into some sort of stem cell before beginning to proliferate. Bessis\(^f\) follows the work of Ferrata\(^g\) in suggesting such a transformation and indicates that some reticuloendothelial cells differentiate into stem cells called hemohistioblasts. Although the concept of hemohistioblasts as stem cells is believed to be valid, it must be noted that the hemohistioblasts of Ferrata do not resemble the cells described as hemohistioblasts in the present study, and, in fact, cannot be distinguished from reticular cells. The question, then, is whether hemohistioblasts (meaning stem cells) of any description arise by transformation from reticular cells. Relative to the hemohistioblasts of Ferrata, it may be said that since they are indistinguishable from reticular cells and since no subgroup of reticular cells appears to proliferate rapidly, there probably are not significant numbers of stem cells arising by transformations within the reticular cell group. Relative to the cells identified as hemohistioblasts in the present work, it should be noted that both the differences in labeling patterns and in labeling intensities between these and reticular cells make it improbable that many hemohistioblasts arise by direct transformation from reticular cells.

It is recognized that the methods employed do not absolutely exclude transformation of a few cells from one type to a stem cell type. Just as a few reticular cells may modulate into blast forms, so it may also be that a few mature lymphocytes transform to more primitive cells. Recently, MacKinney and co-workers\(^h\) have shown enlargement and DNA synthesis in lymphocytes cultured from blood. In addition, Goodman and Hodgson\(^i\) have provided convincing evidence that, under appropriate conditions, blood leukocytes behave as stem cells for both myeloid and erythroid elements. Thus, there may be some stem cells derived from such sources under normal conditions. However, it should be emphasized that when total populations are considered, the labeling patterns of hemohistioblasts and plasmablasts strongly indicates that these cells are adequate to function as stem cells by themselves and neither need nor receive significant supplementation of their number by trans-

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**Fig. 14.**—A plasmablast (top center) is shown along with an immature plasmacyte (bottom).

**Fig. 15.**—The angular or irregular outline of the proplasmacyte (bottom) may be seen together with its characteristic basichromatin.

**Fig. 16.**—Various members of the plasma cell series are labeled in this radioautograph of a section through the medullary cords of a lymph node. Note that labeled reticular cells are not present. Tritiated thymidine was injected every 4 hours for 36 hours before autopsy. The intensity of labeling in the plasma cell series exceeds that of the lymphoid series from the same animal illustrated in fig. 17.

**Fig. 17.**—Radioautograph made from a germinal center in the same tissue section of the animal described in fig. 16. Note that while active lymphopoiesis is evidenced by the extensive labeling seen, reticular cells (r), in the main, are unlabeled and give no indication of the mitotic activity expected of stem cells.
formations from other cell types. The fact that hemohistioblasts are morphologically intermediate between reticular cells and large lymphocytes probably explains why they have frequently been classified with one group or the other and have not usually been given independent status as stem cells.

The same sort of morphologic evidence, viz., the finding of "transition forms," which constitutes much of the justification for reticular cells being considered stem cells, also underlies many reports that plasma cells may derive from lymphocytes. The present results do not support this view although neither do they absolutely exclude the origin of plasma cells from large lymphocytes or hemohistioblasts. The finding of the plasmablast, which actually proliferates more rapidly than the members of the lymphocyte series, as well as the physical separation of these two cell lines in lymph node, make such conversions unlikely in the normal or unstimulated animal. There may be exceptions to this in the antigenically stimulated animal. Nossal and Mäkelä reported that rats stimulated to a secondary response to Salmonella antigens shortly after receiving TTH show many labeled plasma cells, and concluded that these probably derive from large lymphocytes. It is difficult, however, to exclude the plasmablast and proplasmacyte as the source of these cells for the present study shows that these precursors are normally present and are synthesizing DNA. It appears even more improbable that mature small lymphocytes in lymph node convert directly to plasma cells, for at long intervals after multiple injections of TTH, labeled small lymphocytes are frequently encountered but label is never observed in plasma cells which are continually developing. The transformation of small lymphocytes to plasma cells via some intermediary cell, as suggested by McGregor and Gowans, remains a possibility in the antigenically stimulated animal. However, since even "unstimulated" lymph nodes normally encounter some antigens and constantly produce plasma cells, some transformations of this type might be expected in the present animals. From the data it is clear that these do not occur unless they are accompanied by sufficiently frequent divisions of the intermediary cells (at least four) to dilute out the radioactive label before the formation of the earliest recognizable member of the plasma cell series.

**Summary**

Single and multiple injections of tritiated thymidine were combined with radioautography to study the rates of proliferation and interrelationships of the various cell lines in the mesenteric lymph node of the rat. The appearance and labeling patterns of the different cells are described from studies of both smears and tissue sections. Reticular cells exhibit wide variations in labeling intensity, phagocytize labeled lymphocytes, and become labeled in high percentages only when TTH is administered over a period of many days. Other slowly proliferating cell types include small lymphocytes, fat cells, endothelial cells and mast cells.

Rapidly proliferating cell lines include plasmablasts, hemohistioblasts, proplasmacytes and large lymphocytes. The generation time of plasmablasts and
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hemohistioblasts was determined to be approximately 9 and 12 hours respectively. Mature plasma cells constitute a non-dividing population which is renewed in lymph node in not more than 5 days.

Evidence is presented that the most primitive cells in the lymphocyte and plasma cell lines are the hemohistioblasts and plasmablasts respectively. Reticular cells most probably are not stem cells. No evidence is found to support previous reports that plasma cells derive from lymphocytes.

SUMMARIO IN INTERLINGUA

Simple e multiple injectiones de thymidina a tritium esseva combinate con radioautographia pro studiar le proratas de proliferation e le interrelationes del varie lineas de cellulas in le mesenteric nodo lymphatic del ratto. Le apparentia e le distribution del radiomarcage del diverse cellulas es describite a base de studios tanto de froths como etiam de histosectiones. Cellulas reticulari exhibi extense variationes in le intensitate del marcation. Illos deveni radiomarcate in alte procentages solmente quando le thymidina a tritium es administrate duringo periodo de plure dies. Altere typos cellulari de lente proliferation include micre lymphocytos, cellulas adipose, cellulas endothelial, e mastocytos.

Lineas de cellulas con rapide proliferation es plasmoblastos, hemohistioblastos, proplasmocytos, e grande lymphocytos. Le tempore de generation de plasmoblastos e hemohistioblastos eseva determinate a approximativemente 9 e 12 horas, respectivemente. Matur cellulas de plasma constitue un population non-divisive que es renovate in le nodos lymphatic intra non plus que 5 dies.

Es presentate evidentias que le cellulas le plus primitive in le lineas del lymphocytos e del plasmocytos es le hemohistioblastos e le plasmoblastos, respectivemente. Cellulas reticular, probabilissimemente, non es cellulas primordial. Nulle evidentias es trovate in supporto de previe reportos que plasmocytos se deriva ab lymphocytos.

REFERENCES

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