THE OCCURRENCE OF PLASMA CELLS AFTER IONIZING IRRADIATION IN DOGS *

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Histopathologic examination in dogs subjected to whole body x-irradiation† regularly has shown replacement of lymphocytes in lymphoid tissue by cells of a type that meets some or all of the morphologic criteria of the plasma cell. The appearance of plasma cells in hemopoietic organs of humans has been mentioned by Ross¹ (after the presence for 3 years of a radium needle in the interventricular septum of the heart) and by Liebow, Warren, and De Coursey² in atomic energy casualties. It has also been described in animals by Thomas and Bruner³ (chronic radium poisoning in rats), by De Bruyn⁴ (lymph nodes), and by Murray⁵ (spleen). To the best of our knowledge a systematic investigation has not been made previously.

METHOD

Healthy dewormed dogs of the beagle type were divided into three groups, as follows:

I. “High dose” animals given doses varying from 250 to 400 r. (24 dogs)
II. “Low dose” dogs given exposures from 100 to 150 r. (10 dogs)
III. Unirradiated controls (17 dogs)

A 2-million-volt Van de Graaff x-ray generator at the Massachusetts Institute of Technology, with 12.5 mm. Cu h.v.l. was used as a source of radiation. The animals were irradiated bilaterally (whole body irradiation to 2 dogs at a time) at a distance of 3 meters from the target to the midline of the animal. The average dose rate at the animal midline was about 12 r. per minute.

Seventeen of the 24 “high dose” dogs (group I) died. All of the “low dose” dogs (group II) survived. The surviving dogs in both groups were sacrificed between the 30th and 40th post-radiation days except one group II dog that was sacrificed 6 months after exposure.

Necropsies on dogs which died were usually performed within a 2 to 3 hour post-mortem period or less. Tissues were fixed in Zenker’s formol solution and 10 per cent neutralized formalin. The routine

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stain used was hematoxylin and eosin, plus such other stains as seemed to be indicated including phloxine and methylene blue, methyl-green-pyronine, and stains for reticulum and hemosiderin.

Results

Incidence of plasma cells in each of the three groups was recorded by a scale of $-$, $\pm$, $+$, and $++$ in the following structures: lymph nodes, spleen, tonsils, intestine, and bone marrow, as shown in Tables I and II. Any method of recording this type of observation obviously is a compromise. Thus, the bone marrow in fatal cases may be devoid of all cellular elements except the type cell, which, in absolute numbers, might be relatively small. Another point to be mentioned is the very dense lymphocytic peripheral layer of lymph follicles in normal dogs as compared to the loose pattern of depleted lymphoid tissue in the x-irradiated animal. In the normal dog a few plasma cells may escape observation so that possibly some cases which were graded as minus might otherwise be placed in a plus-minus category. We believe that these potential inadequacies do not modify our results substantially.

As mentioned before, the cell we refer to does not always show all of the characteristics of the typical plasma cell; Liebow, Warren, and De Coursey\(^2\) have termed it “plasmacytoid.” The nucleus frequently is not cartwheel-like, but rather shows dense, evenly distributed chromatin particles or even a completely homogeneous pyknotic aspect. However, basophilia of the cytoplasm and the perinuclear halo is always outstanding. Similar cells are seen in peripheral blood of dogs during the terminal stage of x-irradiation illness. With white blood cell counts below 400 per cmm., only a few neutrophils are present. The remainder are not readily identifiable. Many resemble the plasma cell seen in tissues.

**Lymph Nodes (Table I)**

According to Maximow and Bloom,\(^6\) plasma cells commonly are found in human lymph nodes, especially in medullary cords. Their number is subject to marked variations under physiologic, and, to even a greater degree, under pathologic, conditions. In some animals, such as rats and mice, they may be particularly numerous; however, the dog was not mentioned. Our experience is that plasma cells do **not** belong to the constant cellular constituents of lymph nodes in dogs. In 32 lymph nodes of 13 nonirradiated dogs we found no plasma cells in the cortex. In medullary cords these elements are not obligatory,
since 10 of 32 nodes were negative in this respect. In 12, a rare plasma cell was found and in only 6 was the incidence higher. But among the 6 lymph nodes 5 were not normal. One showed in the medullary cords and in the sinusoids abundant polymorphonuclear leukocytes besides the plasma cells. Thus the latter cells in this case were probably a manifestation of an inflammatory reaction the cause of which has not been determined. Another lymph node contained some tuberculoid granulomas; such lesions in lymph nodes, lungs, and other organs of

<table>
<thead>
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<th>Group</th>
<th>Dosage</th>
<th>No. of dogs</th>
<th>No. of lymph nodes studied</th>
<th>Area of study</th>
<th>Graded incidence of plasma cells</th>
<th>Total</th>
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<tr>
<td>I</td>
<td>250–400 r.</td>
<td>24</td>
<td>50</td>
<td>Cortex</td>
<td>25 10 6 9</td>
<td>50</td>
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<td></td>
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<td></td>
<td>Medulla</td>
<td>5 6 11 24</td>
<td>46</td>
</tr>
<tr>
<td>II</td>
<td>100–150 r.</td>
<td>10</td>
<td>25</td>
<td>Cortex</td>
<td>18 7 0 0</td>
<td>25</td>
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<td></td>
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<td></td>
<td>Medulla</td>
<td>8 7 4 5</td>
<td>24</td>
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<tr>
<td>III</td>
<td>0</td>
<td>13</td>
<td>32</td>
<td>Cortex</td>
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<td></td>
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<td>Medulla</td>
<td>10 12 1 6</td>
<td>29</td>
</tr>
</tbody>
</table>

our dogs are not uncommon; they frequently are due to worm-larvae infestation. The remaining plasma-cell-positive lymph nodes were "hemolymph nodes."

It is not appropriate to discuss here the existence of hemolymph nodes in the dog. It may suffice to say that in these structures, sinusoids were filled with erythrocytes, erythrophages, and blood-pigment-carrying macrophages. Lymph nodes of this type have been found by Fahr in dogs in what he calls the portal ring. Even by macroscopic observation they are to be differentiated from other nodes by their rusty brown color. Fahr ascribed to them an important part in iron metabolism. In contradistinction, mesenteric lymph nodes of the dogs are light gray and do not exhibit, in sinusoids, any erythrocytes or erythrophages, except for small areas directed towards the liver hilus in some lymph nodes attached to the root of the mesentery. The co-existence of many plasma cells in these lymph nodes in our material probably is not a casual occurrence. Significantly, in animals in which only one of several nodes was a "hemolymph node," this exhibited numerous plasma cells which were not found in the others. This observation was made also on dogs receiving small amounts of x-irradiation in whom there were no clinical manifestations of x-ray injury. We obviously do not assign a causal relationship to this phenomena but
rather prefer to consider them to be two coordinated manifestations of a special metabolic function of these lymph nodes. In any case, this fact has to be kept in mind in the analysis of the findings in the x-irradiated dog.

In cases of fatal severe radiation injury the major part of the lymph node was occupied by extensive hemorrhages. The cortex was reduced to scantly and poor reproduction of its usual architecture. No germinal centers, not even follicles, were to be found. The diffuse lymphoid tissue, attached to the capsule, not only was markedly depleted, with a rather loose structure, but also was very polymorphous in its cellular composition. In addition to degenerating lymphocytes, reticulum cells were seen, many in all stages of degeneration. There were also large atypical cells, difficult to classify; often these showed pathologic mitotic figures. Generally, however, the plasma cell element was predominant. These cells showed degenerative changes too, especially karyorrhexis. The plasma cells might be phagocytized by reticulum cells. In these nodes the medullary cords were reduced in size, although less so than the cortex. The cytologic picture monotonously showed plasma cells.

In some fatal cases the hemorrhagic process in the lymph nodes was less outstanding. There sometimes were more circumscribed extravasations of blood in otherwise fairly well preserved cortical lymph follicles. Even then the marginal and the medullary sinusoids were filled with erythrocytes. In such cases there were abundant plasma cells in the medullary cords.

In evaluating results, the occurrence of plasma cells in the medullary cords of lymph nodes of normal dogs must be considered. The data presented indicate, however, that the grade of positive findings increases with the x-ray dose. We believe the difference would have been even more marked had we sacrificed all dogs within 30 days after exposure.

Our findings show that x-irradiation effects are more frequently observed in the medulla than in the cortex of lymph nodes. Therefore, we assume a more pronounced tendency to production of plasma cells in this location.

*Spleen (Table II)*

The occurrence of plasma cells in the pulp of the human spleen is a matter of controversy. In the experience of one of us (F.J.W.), they are by no means usually present, although it is accepted that considerable numbers may be encountered without any obvious cause. In contrast we saw in 2 only of 12 untreated dogs a few plasma cells
in the pulp, especially in the perifollicular zones. The picture was completely different after heavy x-irradiation injury. The malpighian bodies were few and small, with a loose arrangement of their cells, and among them plasma cells were predominant. Plasma cells were found in the pulp also. In 2 cases there was outstanding ectopic hematopoiesis. In such foci, also, plasma cells were found.

Plasma cells after ionizing irradiation

Tonsils (Table II)

In normal dogs the tonsil rarely contains plasma cells. With severe x-irradiation injury, necrotizing tonsillitis was constantly found. In such cases lymph follicles were subject to intensive destruction with formation of abundant loose chromatin particles. Among the better preserved cellular elements plasma cells were predominant, although some of them showed varying stages of disintegration. We do not know whether this is a primary x-irradiation effect or a process secondary to a nonspecific necrotizing process. Probably both factors are concerned.

Gastrointestinal Tract

Intestine (Table II)

The mucosa of the normal dog's intestine contains varying amounts of small round-cells, among which are some elements resembling plasma cells. These are chiefly accumulated in a thin basal layer, adjacent to the muscularis mucosae. However, neither the solitary nor the aggregated lymph follicles in the intestinal wall of our normal cases showed any plasma cells. According to De Bruyn, the irradiation effect on
lymphoid tissue in the gastrointestinal tract should be identical to that on isolated lymph nodes. In our series the pertinent changes were both less constant and less pronounced, although similar in kind. Although plasma cells were found, in few instances only did they represent the predominant cellular constituent.

**Bone Marrow (Table II)**

In most cases marrow was taken from both the sternum and the femoral shaft. Many sections from the sternum were technically unsatisfactory. Curettings from the femoral shaft provided better visualization of cellular elements. Normally in this area there are compact islands and some scattered elements of active bone marrow. Plasma cells are absent or very rare. With severe x-irradiation injury, both sternum and femur were composed largely of fat tissue with or without extensive hemorrhage. If there existed other cells, these seemed to be reticulum cells plus macrophages containing iron pigment, but most of the cellular elements present were plasma cells. In dogs sacrificed more than 1 month after radiation with small doses, the marrow might be hyperplastic; in such cases plasma cells were sparse, although perhaps a little more numerous than in non-irradiated animals.

**Thymus**

No plasma cells were found in the thymus glands of 3 untreated or of 6 x-irradiated dogs.

**Comment**

Plasma cells occasionally are present in hematopoietic organs of dogs, chiefly in the medullary cords of lymph nodes. X-irradiation results in the appearance of these cells where they did not occur before or where they were very scanty. In view of the known immediate and prolonged effect of ionizing irradiation upon lymphoid tissue, it is difficult to postulate regeneration of plasma cells from hematopoietic tissue *per se*. Generally, it is thought that plasma cells are derived from lymphocytes. The acute and prolonged effect of ionizing radiation on lymphocytic tissue makes it difficult to reconcile this impression with what we have noted. The alternative is that the plasma cell does not arise from lymphoblasts and that it is in fact, a derivative of the reticulo-endothelial system. We have observed numerous instances in which we think such has been the case.

It is known that in cases of severe radiation injury reticulum cells may be the only surviving elements. Our hypothesis is that reticulum
is the precursor of the cells we describe although we are fully aware of the difficulty in proving this hypothesis. One might even assume that with blockage of hematopoietic cells, reticulo-endothelial cells have potentially more area for development.

We cannot offer even a hypothesis as to the pathogenesis and significance of this plasma cell proliferation. In some instances it seems to be a manifestation of trivial inflammatory reaction in the face of deficiency of the granulocytic system. For example, with tonsillitis of human agranulocytosis or aplastic anemia there is a marked predominance of plasma cells in the inflammatory infiltrations. But in the intestines of our dogs we did not find any conspicuous relationship between mucosal necrosis and the appearance of plasma cells in the lymph follicles.

We might assign a rôle to products of disintegration from destroyed single cells (lymphocytes), but we have no proof for that assumption.

**Summary**

Small numbers of plasma (plasmacytoid) cells may be found in the hematopoietic organs of normal dogs. These are more numerous in the medullary cords of the lymph nodes and, particularly, in those nodes which exhibit the picture of the so-called hemolymph nodes. Without exception, the cortex of lymph nodes of the normal dog was found free of such cells. After x-irradiation, plasma cells are to be found in considerable numbers in the spleen, intestinal lymph follicles, tonsils, and bone marrow. In lymph nodes both the cortex and medulla may be the seat of deposition of these cells, although the medullary cords seem to exhibit the proclivity to produce them. No plasma cells were found in the thymus in either untreated or radiated dogs.

The source of the plasma cells in x-irradiated dogs seems to be reticulum cells. We cannot comment upon the function of these plasmacytoid elements.

**REFERENCES**


LEGENDS FOR FIGURES

Fig. 1. Mesenteric lymph node from a dog given 300 r. of total body x-irradiation, dying on the 18th post-irradiation day. Practically all of the cells in the medullary cord shown here are of the plasmacytoid type. Hematoxylin and eosin stain. × 800.

Fig. 2. Plasmacytoid cell in the peripheral blood of a dog given a mid-lethal dose of ionizing irradiation. This appears similar to those seen in the tissues. There is a perinuclear clear zone; however, the nucleus does not show a cartwheel arrangement of the chromatin. This cell is rarely if ever seen in the peripheral blood of the normal dog. After x-irradiation, it usually appears before clinical manifestations develop. In the late stages, it is very common. Wright's stain. × 3,200.

Fig. 3. A hemolymph node in an unirradiated control dog. There are erythrophages in the sinusoids. Of note is the mixture of lymphocytes and plasmacytoid cells. In the normal dog, it is extremely rare to find the plasmacytoid cell except in the hemolymph node. Hematoxylin and eosin stain. × 600.