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LYMPHOSARCOMA IN AN INFANT NORTHERN FUR SEAL (*Callorhinus ursinus*)¹

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Abstract: An infant northern fur seal (*Callorhinus ursinus*) died in a rookery on St. Paul Island, Pribilof Islands, Alaska. Grossly, slight enlargement of the mesenteric lymph nodes was seen. Microscopically, a lymphosarcoma composed of sheets of monomorphic lymphoid cells in sections of lymph node and tonsil was seen. Electron microscopy of formalin-fixed tissues revealed several structures that were possibly of viral origin in the cytoplasm of the neoplastic cells.

INTRODUCTION

There are few reports of neoplasms in pinnipeds. In recent years three neoplasms, including a squamous cell carcinoma, a transitional cell carcinoma, and a nephroblastoma, were documented,¹ as well as a bile duct carcinoma,² all in California sea lions, *Zalophus californianus*. Also noted was a malignant granulosa cell tumor in a southern elephant seal, *Mirounga leonina*.³ In a chapter that included mention of eight neoplasms in various pinnipeds,⁴ reference was made to an earlier paper that reported lymphosarcoma in two harbor seals, *Phoca vitulina geronimensis*.⁵

This report documents a lymphosarcoma in an infant northern fur seal, *Callorhinus ursinus*, and describes structures that morphologically resemble viruses.

CASE REPORT

History

A female northern fur seal pup was found dead in a rookery on St. Paul

Island, Pribilof Islands, Alaska. It was necropsied as part of a continuing survey of infant mortality in the herd.

Necropsy Findings

The pup weighed 4.4 kg and was 55.1 cm long. From its size, weight, and dentition it was judged to be less than 1 month old, probably between 1 and 3 weeks of age.

Grossly, slight enlargement of the mesenteric lymph nodes, sanguinocatharrhal enteritis, and some liquefaction of subcutaneous fat were noted.

PREPARATION FOR MICROSCOPY

Specimens from all organ systems except the skeletal and nervous systems were fixed in neutral buffered 10% formalin, embedded in paraffin, sectioned at 6 μm, and stained with hematoxylin and eosin. Naphthol AS-D choracetate esterase (Leder) and acid phosphatase reactions were performed on selected sections.

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¹ The opinions or assertions contained herein are the private views of the authors and are not to be construed as official or as reflecting the views of the Department of the Army, of the Air Force, or of Defense.

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Selected tissues were cut from paraffin blocks, deparaffinized, and postfixed in osmium tetroxide. Thick sections were cut at 1 μ m from Epon-embedded specimens and stained with toluidine blue. Thin sections were cut, stained with uranyl and lead citrate, and examined with a Hitachi HS 8-F electron microscope.

RESULTS

Light-Microscopic Findings

In the tonsil and in two sections of lymph nodes, one of which was identifiable as the mesenteric, sheets of monomorphic lymphoid cells with no follicular architecture and very scant stroma were seen (Fig. 1). This pattern was especially prominent in the unidentified lymph node, which also had areas of hemorrhage, necrosis, and early fibrosis. Small accumulations of lymphoid cells also were present in the capsule, and in some areas lymphoid cells obliterated the peripheral sinus. The salivary gland tissue adjacent to the tonsil had several localized infiltrations of lymphoid cells. Mitotic activity was noted in one of these foci. The mesenteric lymph node had several perivascular accumulations of rather homogenous lymphoid cells in its capsule and serosal fat. This lesion was continuous with a similar serosal lesion in the adjacent colon.

The colon had excessive aggregates of homogenous lymphoid cells suprajacent and subjacent to the muscularis mucosa. More superficially in the lamina propria and deeper in the submucosa, some areas contained many plasma cells. The mucosa was intact except for its luminal surface, which showed postmortem changes.

In most of the areas mentioned, but especially in the lymph nodes, the lymphoid cells were rather homogenous and had medium to medium-large nuclei with slightly irregular, rounded to ovoid shapes and distinct nuclear envelopes. Nucleoli were prominent in most cells. A modest amount of lightly eosinophilic to amphophilic cytoplasm with rounded to polygonal outline was seen. Many

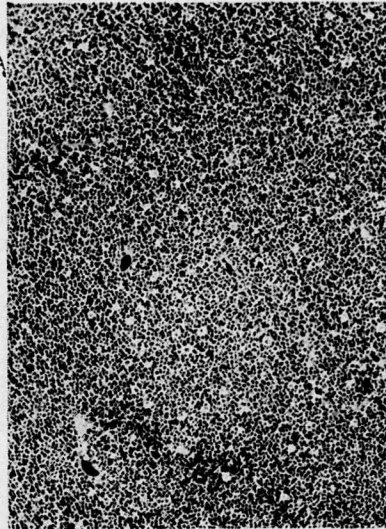


FIGURE 1. Lymph node. Broad sheets of rather uniform lymphoid cells are interrupted only by delicate vessels (dark foci) and histiocytic cells, which impart a "starry sky" appearance. X115. AFIP Neg. 76-2152.

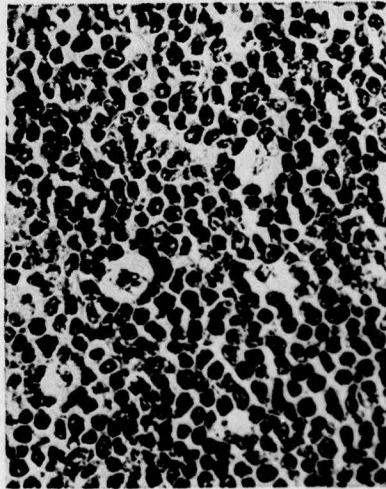


FIGURE 2. Greater magnification of Figure 1. The lymphoid cells are fairly well differentiated. X530, AFIP Neg. 76-2155.

mitotic figures were present (up to five mitotic figures per high-power field). Necrotic lymphoid cells or histiocytic cells were scattered through the tissue, imparting a "starry sky" appearance (Figs. 1 and 2).

The Leder stains and acid phosphatase stains showed no activity in the neoplastic cells.

Electron-Microscopic Findings

As expected, tissue preservation was considerably less than optimal, owing to formalin fixation of routine necropsy tissue. Nevertheless, the relative homogeneity of the lymphoid cells was demonstrated (Fig. 3). Most cells had modest numbers of poorly preserved mitochondria and indistinct plasma membranes. Heterochromatin was clumped at the periphery of the nuclei. The nuclear envelopes were well defined, and the outer layers were studded with ribosomes.

The presence of unidentified round structures was detected in the cytoplasm of some of the cells (Figs. 4 and 5). These structures were circular and had dense, sometimes granular outer margins and variably less dense cores. They were 60 to 90 nm in diameter, with most falling in the 80- to 90-nm range.

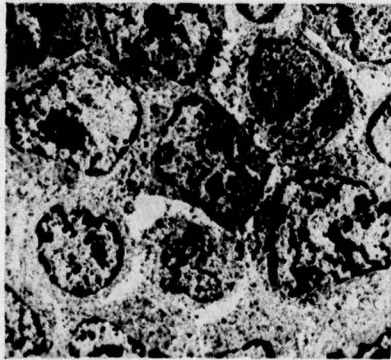


FIGURE 3. Electron micrograph. Lymph node. Heterochromatin is clumped primarily at the periphery of the nuclei, which have distinct envelopes. Prominent nucleoli are seen in some nuclei. X3,700. AFIP Neg. 76-5399.

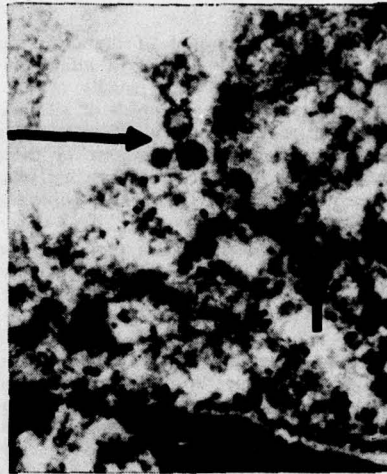


FIGURE 4. Electron micrograph. Neoplastic lymphoid cell. Heterochromatin is at the periphery of the nucleus (bottom). The outer layer of the nuclear envelope is studded with many ribosomes. Size of the circular structures (arrow) ranges mostly from 80 to 90 nm. An autolytic mitochondrion is seen to the right of the circular structures (short arrow). X87,500. AFIP Neg. 76-7702.



FIGURE 5. Similar section to Figure 4. Additional circular structures were seen in other neoplastic lymphoid cells. X91,500. AFIP Neg. 76-7703.

DISCUSSION

A minimal amount of literature has been addressed to lesions in infant pinnipeds, and there is a paucity of neoplasms reported in pinnipeds of all ages. This scarcity may be a reflection of a low prevalence of neoplasms in these animals or simply of the fact that relatively few pinnipeds have been subjected to a thorough necropsy.

To our knowledge this represents the first reported case of a neoplasm of the lymphoid tissue in this species. In a recent report Brown *et al.*¹ described severe enlargement of mesenteric lymph nodes in northern fur seal pups infected with hookworms, a common and often fatal disease of this species. The neoplastic lesions in the case reported herein were easily distinguished microscopically from the hyperplastic nodes described by Brown. In the authors' experience the hyperplastic lymph nodes in pups with hookworm infection contained a mixture of cell types, in contrast to the monomorphic sheets of neoplastic lymphoid cells present in the lymph nodes and tonsils of our case. The colonic lesions were more difficult to interpret as to

involvement with the neoplastic process. Perivascular areas adjacent to the muscularis mucosa had collars of monomorphic lymphoid cells of similar morphologic pattern to those present in the neoplastic nodes and tonsils. In other areas the colon showed a definite inflammatory reaction, as mature lymphocytes and plasma cells predominated.

Definitive conclusions cannot be made concerning the nature of the circular structure of particles observed in the cytoplasm of the neoplastic cells. Their size was roughly in the range (70 to 75 nm) of the intracellular Type A oncornavirus particles of the mammary tumor agent of the mouse, but the two concentric spherical shells that characterized the Type A particle were either absent or had been obscured by the poor fixation. That the structures represent artifacts formed from normal cytoplasmic structures is a distinct possibility because of the less than optimum processing of the specimen for ultrastructural evaluation. For example, they closely resembled microsomes prepared for ultrastructural examination by cellular-fractionation techniques.

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LITERATURE CITED

1. BROWN, R. J., A. W. SMITH, M. C. KEYES, W. P. TREVATHAN and J. L. KUPPER. 1974. Lesions associated with fatal hookworm infections in the northern fur seal. *J. Am. vet. med. Ass.* 165: 804-805.
2. GRINER, L. A. 1971. Malignant leukemic lymphoma in two harbor seals (*Phoca vitulina geronimensis*). *Am. J. vet. Res.* 32: 827-830.
3. MAWDESLEY-THOMAS, L. E. 1971. An ovarian tumor in a southern elephant seal (*Mirounga leonina*). *Vet. Path.* 8: 9-15.
4. RIDGWAY, S. H. 1972. Homeostasis in the aquatic environment. In: *Mammals of the Sea. Biology and Medicine*. Ed. by S. H. Ridgway. Charles C. Thomas, Springfield, Ill. pp. 590-747.
5. SCHROEDER, R. V., C. A. D. QUADRI, R. W. McINTYRE and W. A. WALKER. 1973. Marine mammal disease surveillance program in Los Angeles County. *J. Am. vet. med. Ass.* 163: 580-581.
6. SWEENEY, J. C. 1974. Common diseases of pinnipeds. *J. Am. vet. med. Ass.* 165: 805-810.

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