LETTERS

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B Cell Lymphoma in an Adult Female Mule Deer (*Odocoileus hemionus*) from Wyoming, USA

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ABSTRACT: A free-ranging adult female mule deer (*Odocoileus hemionus*) from Wyoming, US, was euthanized due to an open wound on its head. Postmortem examination yielded a diagnosis of multicentric B cell lymphoma associated with severe skin ulceration. Sequencing of frozen neoplastic tissue found no evidence of an exogenous viral etiology.

A free-ranging adult female mule deer (*Odocoileus hemionus*), from Sheridan County, Wyoming, US (44°44′0.9″, 106°54′31″), was reported to the Wyoming Game and Fish Department (WGFD; Laramie, Wyoming, US) on 22 July 2024, due to the presence of a "severe lesion on its head" (Fig. 1A, B). A WGFD employee euthanized the deer by gunshot to the neck; noted its poor body condition; performed a field necropsy; and submitted the frozen head and cranial neck, along with samples of liver, tongue, lung, esophagus, thyroid gland, and spleen, to the Wyoming State Veterinary Laboratory (WSVL; Laramie, Wyoming, US).

At necropsy, cranial to the right eye was an open skin wound (deep ulcer) covered by a dark crust of dried blood and necrotic material invaded by fly larvae (Fig. 1B). Multiple ovoid subcutaneous masses were palpated on the right side of head. Once incised, the masses were tentatively identified, based on their location and shape, as regional lymph nodes with the parenchyma completely effaced and replaced by a homogeneous, solid, pale tan infiltrate (Fig. 1C). The right retropharyngeal lymph node was similarly enlarged (6 cm greatest diameter, versus 3 cm for the left, normal-appearing node) and replaced by solid pale tan infiltrate. The left parotid, submandibular and retropharyngeal lymph nodes were smaller, with a distinct cortex and medulla on cut surface. The skin, subcutis, and muscle around the ulcer were thickened by a similar infiltrate, occasionally mixed with green suppurative exudate, that partially effaced regional dermal structures (Fig. 1A). The infiltrate extended into the nasal cavity, through the nasal bones, and into the right eye orbit. Samples of affected lymph nodes and skin, plus the left retropharyngeal lymph node (unaffected control) were fixed in 10% formalin and processed for routine histopathologic examination. Routine tests for specific pathogens including chronic wasting disease (ELISA on the left retropharyngeal lymph node), aerobic bacterial culture (lung, liver, and spleen), and cervid herpesvirus 4 detection (PCR on unaffected lymph node) were performed. No agents were detected.

Histopathologically, the masses were confirmed to be lymph nodes given the presence of an outer capsule of fibrous connective tissue extending into their parenchyma forming trabeculae, and the presence of small lymphocyte clusters. In the right parotid, submandibular, and retropharyngeal lymph nodes, neoplastic sheets of round cells supported by the regional collagenous stroma effaced and replaced the normal architecture, extended to adjacent soft tissues, and expanded the dermis over the nasal planum. Neoplastic cells had moderate amounts of eosinophilic cytoplasm, occasionally with a hint of a perinuclear pallor suggesting a well-developed Golgi apparatus (Fig. 1D). The nuclei were round, ovoid, indented or irregular, with moderate to marked variation in size, and clumped chromatin; a few larger nuclei had a large magenta nucleolus. Mitoses (counted in areas with least autolysis and freeze-thaw artifact) were uncommon, averaging (mean) 0–1 per 10

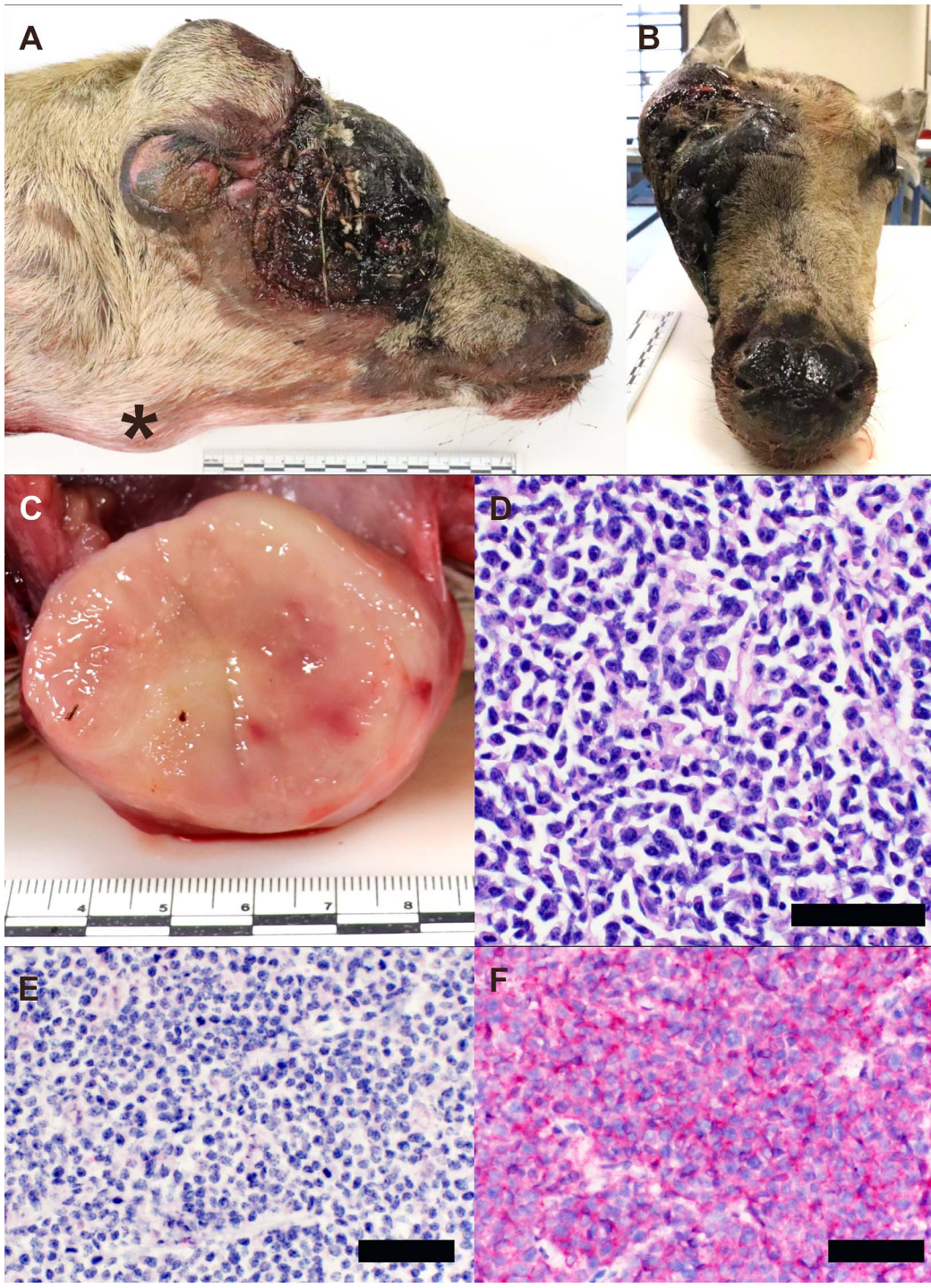

FIGURE 1. Mule deer (*Odocoileus hemionus*) from Wyoming, US, with ulcerated lesion cranial to the right eye (A, B) and enlarged submandibular lymph node (A, asterisk). The enlarged submandibular lymph node was firm, had a pale tan discoloration, and had no cortico-medullary distinction on cut surface (C). Histopathologic examination of the lymph node revealed a homogeneous population of neoplastic round cells (D, H&E) that failed to stain with CD3 antibodies (E), but had strong membranous staining with CD20 antibodies (F), yielding a diagnosis of B cell lymphoma. Scale bars, 50 μm.

random high-power fields (40×), but some may have been missed due to poor tissue preservation. Where the neoplastic population abutted the ulcerated skin, there was secondary bacterial infection and thick crusting. Neoplastic cells had invaded and destroyed the cranial bones, and reached the meningeal space of the frontal lobe, forming a nonencapsulated but well-demarcated mass invading the leptomeninges and extending into the perivascular (Virchow–Robin) spaces in the neuropil. Neoplastic cells had also infiltrated the subconjunctival region of the right eye. No significant lesions were present in the sections of left retropharyngeal and parotid lymph nodes, liver, tongue, lung, esophagus, thyroid gland, and spleen examined. Moderate tissue autolysis and marked freeze-thaw artifact were present.

Immunohistochemic staining with CD3, CD20, and CD79a antibodies (catalog no. A0452, Dako, California, US; catalog no. RB9013, Epredia, Michigan, US; catalog no. CM067C, Biocare Medical, California, US, respectively) was performed on an affected lymph node and on the left (unaffected) parotid lymph node and spleen (as species controls). Neoplastic cells did not stain with CD3 antibody (Fig. 1E) and exhibited strong immunolabeling with CD20 antibody (Fig. 1F). Immunolabeling with CD79a mostly mimicked that of CD20, albeit with a much weaker signal. Immunolabeling of lymphocytes in control tissues confirmed that the antibodies used worked appropriately for this species.

A portion of one of the affected lymph nodes was refrozen and submitted to the Iowa State University Veterinary Diagnostic Laboratory (Ames, Iowa, US) to investigate the possible presence of a viral agent through unbiased next generation sequencing of both DNA and RNA libraries. No exogenous viral reads were detected; however, a low number of reads were detected, with 98–99% nucleotide identity to cervid endogenous gammaretrovirus (GenBank no. JN592050.1).

A diagnosis of B cell lymphoma in the right cranial lymph nodes with invasion of the adjacent skin, skull, and frontal cerebrum was reached. Although we cannot rule out the presence of tumor in internal organs, the sections of liver, lung and spleen examined contained no evidence of neoplastic cells.

Neoplasia in cervids is uncommon but often of lymphocytic origin (Pewsner et al. 2017) and, for those reports that include immunophenotyping, of T cell lineage (Madson and Opriessnig 2009; Kleinschmidt et al. 2012). We could not find reports of B cell lymphomas in North American deer, or of any neoplasia in mule deer specifically, in peer-reviewed literature. A mean of 62 mule deer examinations (necropsy and histopathology) are performed at the WSVL each year, thus approximately 1,550 mule deer carcasses have been examined by a pathologist between 1999 and 2024. Of those, neoplasia was diagnosed in 10 individuals (0.6%), and 5 (0.3%) were identified as lymphomas. Four of those lymphomas were typed by immunohistochemistry: all were of T cell origin.

The gross presentation in this mule deer resembled that of the ethmoidal tumor in moose (*Alces alces*): a squamous or sarcomatous neoplasia that arises from the mucous membranes of the eponymous bone, extending to soft tissues and producing the "hole-in-the-head disease" (Borg and Nilsson 1985; Laaksonen and Paulsen 2015). Ethmoidal tumors also occur in roe deer (*Capreolus capreolus*) and are suspected to have a viral etiology (Borg 1987). Although the lymphocytic nature of the tumor in this mule deer differed from that of ethmoidal tumors in moose and roe deer, the possibility of a viral etiology was considered. Exogenous retroviruses, known to cause tumors in various wild species (Pesavento et al. 2018), were not detected in our mule deer. We did find an endogenous retrovirus with 98–99% nucleotide identity to that of a cervid endogenous gammaretrovirus first reported in wild mule deer in the neighboring state of Montana, US (Elleder et al. 2012). Cervid endogenous retroviruses, which are stably integrated in the host genome and transmitted vertically, have also been reported from hunter-harvested mule deer in Utah, US, and Wyoming (Kamath et al. 2014). Whether endogenous retroviruses are potentially oncogenic in wildlife is unknown (Pesavento et al. 2018).

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