CASE REPORT

Solitary (primary) uveal T-cell lymphoma in a horse

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Abstract
A 22-year-old Australian stockhorse gelding was presented with anterior uveitis in the right eye which was nonresponsive to anti-inflammatory therapy. Clinical examination revealed corneal edema and vascularization, marked hypopyon, and thickening of the dorsal iris, which was confirmed by ultrasonography. Hematologic and biochemical analyses, abdominal and thoracic ultrasonography, and abdominocentesis with cytologic and biochemical analysis revealed no significant abnormalities. Cytological examination of an aqueous humor sample revealed a population of predominantly large lymphoblasts with high nuclear-to-cytoplasmic ratio, round or irregular nuclei, clumped nuclear chromatin, multiple large prominent nucleoli, and a small volume of basophilic cytoplasm. The cytologic diagnosis was intraocular lymphoma. Biopsy of the right submandibular lymph node revealed no evidence of neoplastic invasion. Euthanasia and a complete necropsy were performed and revealed no evidence of neoplasia in any tissue other than the right eye, which had an extensive, well-defined infiltrate of neoplastic lymphocytes expanding the ciliary body and iris, infiltrating the ciliary epithelium, and extending into the pars plana and peripheral choroid. Immunohistochemistry confirmed that neoplastic cells expressed the T-cell marker CD3. To the authors’ knowledge, this is the first description of primary, solitary uveal T-cell lymphoma in a horse. Although apparently rare, lymphoma should be considered in horses with uveitis, even when inflammation is unilateral and in the absence of extraocular signs of neoplasia. Aqueocentesis and cytological examination provided an antemortem diagnosis in this case and should be considered as a diagnostic tool for investigation of uveal thickening and hypopyon.

Key Words: aqueocentesis, cytology, equine, histology, immunohistochemistry, lymphoma, T-cell lymphoma

INTRODUCTION
Lymphoma is the most frequently diagnosed neoplasm of the equine hematopoietic system and can be classified as multicentric, alimentary, mediastinal, cutaneous, or solitary.1-3 Of these, multicentric lymphoma is reportedly the most common form in horses and is characterized by widespread involvement of lymph nodes and a variety of organs. The liver, spleen, intestine, kidney, and bone marrow are most commonly affected,1.4,5 but involvement of the upper airway, central nervous system, heart, adrenal glands, reproductive organs, and eye have all been reported.6-17 In a report of 37 horses with lymphoma,4 34 had tumors located in multiple lymphoid tissues plus abdominal or thoracic organs, and 3 were restricted to the skin and subcutis. Reports regarding the most common subtype of lymphoma in the horse are contradictory with some authors reporting T-cell-rich, B-cell lymphoma most commonly,18,19 and others report T-cell lymphoma as the most common subtype.2,4 To the authors’ knowledge, ocular manifestations of lymphoma have been reported only in horses with the multicentric form of disease. Even a retrospective case series emphasizing ocular lesions in 21 horses with lymphoma, 4 of which had uveitis as their primary complaint and histologically confirmed uveal infiltration with neoplastic cells, revealed that all 21 horses had multicentric lymphoma.20 A case report described a horse with
bilateral uveal lymphoma; however, this horse also had splenomegaly and hepatomegaly. Aqueocentesis of the left eye in this horse performed postmortem confirmed intraocular T-cell lymphoma as part of multicentric disease. Clinical signs of ocular involvement in equine multicentric lymphoma include intermittent eyelid swelling, chronic ocular discharge, third eyelid edema, unilateral exophthalmos, corneoscleral masses, and chronic uveitis that is unresponsive to treatment.

To the authors’ knowledge, the present report is the first to describe antemortem diagnosis of unilateral uveal lymphoma in a horse by aqueocentesis, and the first to document primary solitary ocular lymphoma in a horse confirmed by complete necropsy.

CASE REPORT

A 22-year-old Australian stockhorse gelding was referred to the University of Melbourne Equine Centre for investigation of severe uveitis of the right eye (OD) of 10 days duration that was poorly responsive to treatment with atropine 1% ophthalmic solution (0.1 mL OD q8 h) and phenylbutazone (2.2 mg/kg orally q12 h). Clinical examination revealed the gelding was in poor body condition (body condition score 3/9), but all other physical parameters were within normal limits. The horse behaved as if sighted. There was mild to moderate blepharospasm OD. There was no ocular discharge from either eye and no facial staining suggestive of prior discharge. The periocular skin and eyelids were mildly swollen OD; otherwise no facial asymmetry was noted. There was marked anisocoria due to wide mydriasis in the left eye (OS) and marked miosis OD. Direct and consensual pupillary light reflexes were absent in both eyes (OU). Menace response, and dazzle and palpebral reflexes were complete OU, and globe position and movements were normal OU. Following sedation and an auriculopalpebral nerve block OU, both eyes were examined by use of diffuse and focal light, slit-lamp biomicroscopy, and monocular indirect and direct ophthalmoscopy. There was moderate conjunctival hyperemia without chemosis OD. There was mild diffuse corneal edema along with mid-stromal corneal blood vessels extending approximately 2–3 mm axially from all 360 degrees of the corneoscleral limbus OD. Dense hypopyon occupied the ventromedial 1/4 to 1/3 of the anterior chamber OD. Dorsal to this, there was marked (3+’21) aqueous flare. The iris was diffusely thickened OD and had 2 ridge-like regions of more notable thickening extending vertically from the iris root to the pupillary margin dorsally (Fig. 1). These regions of the iris were affected by rubeosis iridis. The lens appeared within normal limits. Examination of the lens and intraocular structures caudal to it was limited due to opacification of the cornea and anterior chamber and intense miosis OD. No abnormality other than fixed mydriasis was detected OS. Fluorescein staining of both corneal surfaces failed to identify any corneal defects. Intraocular pressure was not measured because a tonometer was not available at the time of examination. Ophthalmic clinical diagnoses at this stage were chronic active uveitis with regional iridal thickening or masses OD. Due to the absence of other lesions OS, the mydriasis of that eye was suspected to be due to inadvertent atropine application or due to systemic effects secondary to frequent atropine application OD prior to referral. Etiologic diagnoses considered for the uveitis OD were ocular lymphoma or other intraocular neoplasia, lepto- tospirosis or other infectious causes, immune-mediated disease, and penetrating trauma (with an entry wound caudal to the those regions observed clinically).

Ocular ultrasonography was performed OD, due to inability to examine the posterior segment OD and to better characterize the iridal lesions. The gelding was sedated with intravenously administered detomidine (0.01 mg/kg) and butorphanol (0.01 mg/kg), and transcorneal ultrasonography was performed OD using a variable frequency (5–13 MHz) linear transducer (Acuson x300, Siemens Medical Solutions, Mountain View, California, USA). The dorsolateral aspect of the iris was hypoechoic (Fig. 2) and thickened (3.03 mm) relative to the dorsomedial aspect (1.58 mm) and relative to published reference values [2.5 ± 0.66 mm].’22 The lens, vitreous body, posterior wall of the globe, and retrobulbar structures OD were ultrasonographically normal.

A subpalpebral lavage system was placed through the ventromedial aspect of the lower eyelid to facilitate topical administration of medications,’23 and the gelding was started on flunixin meglumine (1.1 mg/kg IV q12 h) and atropine 1% ophthalmic solution (0.2 mL OD q12 h) via the subpalpebral lavage system. Re-examination the following day revealed mild improvement in the degree of blepharospasm, but marked miosis and hypopyon were still present OD. Repeat fluorescein staining of the right cornea failed to identify any corneal epithelial defects, and topical administration of dexamethasone (0.2 mL OD
with acepromazine (0.03 mg/kg IV) and romifidine toxoid were administered. The gelding was premedicated nylbutazone (4.4 mg/kg IV), and tetanus toxoid and anti-penicillin (22 mg/kg IM), gentamicin (6.6 mg/kg IV), phenylbutazone OD and biopsy of the right submandibular lymph node were recommended under general anesthesia. Prior to referral, aqueocentesis OD and biopsy of the right submandibular lymph node revealed regional expansion of the cortex with small, densely packed, dark lymphoid cells, large numbers of lymphocytes within a predominant population of large lymphoblasts with high nuclear-to-cytoplasmic ratio, round or irregular nuclei, clumped nuclear chromatin, multiple large prominent nucleoli, and a small volume of basophilic cytoplasm (Fig. 3). The cytologic diagnosis was intraocular lymphoma. Histopathologic examination of the right submandibular lymph node revealed regional expansion of the cortex with small, densely packed, dark lymphocytes with a low mitotic index (Fig. 4). These were immunophenotypically identified as T cells. Infiltration of the lymph node capsule was not seen, and no evidence of neoplasia was noted. The histologic diagnosis was reactive lymphadenopathy.

The owner elected to euthanize the horse 7 days after referral and permitted a complete postmortem q8 h) was initiated. Re-examination 24 hours later (2 days after admission) revealed no improvement OD, but the degree of mydriasis OS had reduced, supporting the conclusion that this was very probably due to inadvertent atropine application to that eye or due to systemic effects secondary to frequent topical administration OD prior to referral. A venous blood sample was drawn for hematologic and biochemical analyses. Abdominal and thoracic ultrasonography and abdominocentesis with cytologic and biochemical analysis were performed. No significant abnormalities were identified on any of these examinations.

Due to failure to detect a cause of uveitis with extraocular testing, notable thickening of the iris evident clinically and ultrasonographically, and a poor response to anti-inflammatory therapy by Day 5 after referral, aqueocentesis OD and biopsy of the right submandibular lymph node were recommended under general anesthesia. Prior to anesthesia, a 14-gauge 5.25” intravenous catheter (Angiocath, Becton Dickinson Infusion Therapy Systems Inc., Utah USA) was placed in the right jugular vein. Procaine penicillin (22 mg/kg IM), gentamicin (6.6 mg/kg IV), phenylbutazone (4.4 mg/kg IV), and tetanus toxoid and antitoxin were administered. The gelding was premedicated with acepromazine (0.03 mg/kg IV) and romifidine (80 µg/kg IV), and anesthesia induced with ketamine (2.2 mg/kg IV) and diazepam (0.05 mg/kg IV) and maintained with isoflurane in 100% oxygen following orotracheal intubation with a 24-mm diameter endotracheal tube. Physiologic monitoring throughout the procedure consisted of pulse oximetry, direct arterial blood pressure measurement, capnography, end-tidal gas monitoring (Mindray PM 9000, Shenzhen Mindray Bio-Medical Electronics, Shenzhen China), and blood gas analysis (ABL 625, Radiometer, Denmark). Blood pressure was maintained above a mean arterial pressure of 70 mmHg throughout the procedure. The ocular surface OD was disinfected with 1:50 povidone iodine solution, and an eyelid speculum was placed. A 23-gauge 1” needle was inserted through clear cornea just anterior to the corneoscleral limbus, and directed into the hypopyon in the ventral aspect of the anterior chamber. Once the needle was appropriately positioned gentle aspiration was performed. The needle appeared obstructed by the highly cellular contents of the anterior chamber and neither aqueous humor nor cellular material was visible in the syringe. However, the needle was removed and a grossly visible sample was expressed onto a glass slide and air-dried for cytologic examination. Because of the apparent viscosity of the aqueous humor and to ensure a diagnostic sample, a 20-gauge 1” needle then was inserted through an adjacent site in clear cornea just anterior to the corneoscleral limbus, and approximately 0.5 mL of aqueous humor was slowly aspirated. The sample was placed into a vacuum tube containing EDTA and submitted for cytologic assessment. A small amount of aqueous humor leaked from the aspiration site following removal of the 20-gauge needle. This port was closed with a single, simple interrupted suture using 8–0 polyglactin 910. Atropine, chloramphenicol, and dexamethasone solutions were applied to the cornea while the horse was under general anesthesia. Wedge biopsy of the right submandibular lymph node was performed, the sample placed into formalin and submitted for histopathologic examination. Skin overlying the biopsy site was closed using 2–0 polypropylene in a simple interrupted pattern. The gelding recovered uneventfully from general anesthesia and was maintained on flunixin meglumine (1.1 mg/kg IV q12 h), atropine (0.1 mL OD q12 h), dexamethasone (0.2 mL OD q8 h), and chloramphenicol (0.2 mL OD q8 h).

Cytologic examination of air-dried direct smears and a cytospin preparation of the aqueous humor revealed a large population of lymphoid cells, large numbers of disintegrated cells, and abundant cellular debris. The lymphoid cell population included small- and medium-sized lymphocytes within a predominant population of large lymphoblasts with high nuclear-to-cyttoplasmic ratio, round or irregular nuclei, clumped nuclear chromatin, multiple large prominent nucleoli, and a small volume of basophilic cytoplasm (Fig. 3). The cytologic diagnosis was intraocular lymphoma. Histopathologic examination of the right submandibular lymph node revealed regional expansion of the cortex with small, densely packed, dark lymphocytes with a low mitotic index (Fig. 4). These were immunophenotypically identified as T cells. Infiltration of the lymph node capsule was not seen, and no evidence of neoplasia was noted. The histologic diagnosis was reactive lymphadenopathy.

The owner elected to euthanatize the horse 7 days after referral and permitted a complete postmortem

Figure 2. Transverse plain transcorneal ultrasonographic image of the right eye of the horse shown in Fig. 1. The lateral aspect of the iris is hypoechoic and thickened when compared to the medial aspect. Note also the iridal folds as seen clinically.
examination. A wide selection of tissues were collected at autopsy, including both globes, brain, spinal cord, and lymph nodes of the head and body cavities. Both eyes were examined histologically. In the right globe, an unencapsulated but moderately well-defined infiltrate of discrete round cells consistent with lymphocytes expanded the ciliary body and iris root, infiltrated the ciliary epithelium, and extended into the pars plana and peripheral choroid dorsally. These cells also invaded adjacent scleral collagen and were present in the anterior chamber and iridocorneal angle. The neoplastic cells formed sheets that separated and effaced stromal collagen and whorled around blood vessels of the iris (Fig. 5). Neoplastic cells had sparse cytoplasm and round to irregular nuclei with coarse or stippled chromatin. There was moderate anisocytosis and anisokaryosis. The mitotic rate was less than 1 per 10 high power fields. Immunohistochemistry using antibodies directed against CD3 (Dako 1:25) and Pax 5 (BD, 1:25) identified the neoplastic cells as T lymphocytes, with small numbers of normal B lymphocytes also present (Fig. 6). Small capillaries extended from the iris stroma to form a vascular network on the anterior iridal surface (pre-iridal fibrovascular membrane), and there was moderate to severe anterior chamber hemorrhage. There was mild, full thickness corneal stromal vascularization, and epithelial edema. Lens, retina and optic nerve OD were unremarkable. No lesions were seen in the left globe. No abnormal lymphocytic infiltrates were seen in any other tissue examined. The histologic diagnosis based on these findings was primary uveal T-cell lymphoma.

DISCUSSION

To the authors’ knowledge, this case report provides the first description of solitary intraocular lymphoma despite relatively frequent reports of ocular involvement in horses.
Ocular manifestations of lymphoma are common in other species. Dogs with ocular lymphoma commonly have uveal infiltrates or tumors, intraocular hemorrhages, and retinal hemorrhages, and approximately 25% of dogs with uveitis in one study were diagnosed with neoplasia. Uveal involvement was noted in 100% of cats with ocular and multicentric lymphoma in one study. By contrast, solitary ocular lymphoma appears to occur very uncommonly. To the authors’ knowledge, there is a single case report of primary oculocerebral B-cell lymphoma in a cat and one report of solitary intraocular lymphoma in dogs. Likewise, primary intraocular lymphoma in humans is rare, representing < 1% of patients diagnosed with lymphoma. In humans, the most common forms of primary intraocular lymphoma are vitreoretinal or choroidal, rarely predominantly iridal as described in the current horse. Importantly, ocular lymphoma in humans precedes the development of central nervous signs in 50–80% of cases, with patients developing central nervous system disease within 2 years of diagnosis. Development of neurological disease in dogs (Personal communication, KT Wiggans) and cats with solitary ocular lymphoma may occur. By contrast, the present case exhibited no neurological signs, and neither gross nor histologic examination postmortem identified neurological involvement.

To the authors’ knowledge, this case report also provides the first description of antemortem diagnosis of intraocular lymphoma using cytological examination of a sample obtained by aqueocentesis in a horse. Aqueocentesis was performed in the present case because of the clinically and ultrasonographically detectable thickening of the iris, poor response to treatment, and the lack of significant findings on extracocular diagnostic tests. Aqueocentesis was performed under general anesthesia because of the risk of intraocular damage (especially to the iris and lens). This procedure may be possible in standing sedated horses, if auriculopalpebral akinesia, periocular anesthesia, and topical anesthesia of the cornea are employed. Cytologic examination of the sample revealed an abnormal lymphoid cell population which is consistent with reports in humans, where exfoliation is believed to result in neoplastic lymphoid cell population which is consistent with reports in humans, where exfoliation is believed to result in neoplastic lymphoid cell population which is consistent with reports in humans, where exfoliation is believed to result in neoplastic lymphoid cell population which is consistent with reports in humans, where exfoliation is believed to result in neoplastic lymphoid cell population which is consistent with reports in humans, where exfoliation is believed to result in neoplastic lymphoid cell population which is consistent with reports in humans, where exfoliation is believed to result in neoplastic lymphoid cell population which is consistent with reports in humans, where exfoliation is believed to result in neoplastic lymphoid cell population which is consistent with reports in humans, where exfoliation is believed to result in neoplastic lymphoid cell population which is consistent with reports in humans. Fine needle aspiration of intraocular masses may also be performed, and is claimed to have numerous advantages over centesis alone including simultaneous collection of adjacent intraocular fluids (vitreous and/or aqueous); ability to obtain sufficient cells to perform cytological and immunocytochemical analyses; and that they are less invasive and cause fewer complications than do iridal or chorioretinal biopsy. Fine needle aspiration of the iris was not performed in the horse of this present report because of concerns regarding hemorrhage from the iris or its pre-iridal fibrovascular membrane, and because the aqueous humor was so cellular that a diagnosis was considered likely based on centesis alone. Indeed the fluid was so highly cellular and proteinaceous that there was concern that an adequate sample was not collected using a 23-gauge needle. Therefore, a 20-gauge needle was used in a subsequent centesis, and a highly cellular fluid sample was collected. Use of cytopin technology was useful in the present case to ensure adequate cells were available for cytologic assessment.

Immunophenotyping of human lymphomas is performed to gain a more refined prognosis, institute strategic therapy, and allow therapeutic monitoring. Similarly, histologic categorization of equine lymphomas has been recommended, but has proven difficult because of marked tumor heterogeneity. Thus, immunophenotyping is considered necessary for further differentiation of equine lymphoma. However, retrospective studies
attempting to classify equine lymphoma morphologically and immunohistochemically have produced conflicting results with regards to the most common subtype. Meyer et al. classified 26 of 37 equine lymphomas as of T-cell origin. But subsequently, Durham et al. identified T-cell-rich, B-cell lymphoma as the most common subtype in 87 of 203 cases reported. The tumor described here was identified as being of T-cell origin, which is consistent with a previous report of a malignant equine T-cell lymphoma with uveal involvement. In human oncology, lymphomas of T-cell origin are considered to be more aggressive and to carry a poorer prognosis when compared with B-cell lymphoma. Further investigation of this is warranted in horses.

Previous reports suggest that the prognosis following diagnosis of equine lymphoma in which there is intraocular involvement is extremely poor with 20 of 21 horses euthanized or dead within 6 months of diagnosis. The prognosis for humans diagnosed with primary intraocular lymphoma is better with approximately 60% of humans surviving for 5 years following diagnosis. Treatment options described for horses with lymphoma include enucleation, enucleation combined with local chemotherapeutic agents, systemic chemotherapy, and brachytherapy or teletherapy. Reports regarding response to therapy for equine lymphoma are scarce due to the advanced nature of the disease upon diagnosis, costs involved in treatment, concerns regarding radiation safety and appropriate disposal of chemotherapeutic agents in the field, the ability to isolate horses following radiation therapy, and the reported poor prognosis. None of these options were exercised in the case described here due to financial considerations.

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REFERENCES


