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Distinctive tumour of the tongue in 3 horses

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Summary

Tumours arising from the dorsal surface of the tongue occurred in 3 horses from 14-23 years of age. Tumours were surgically excised at a referral hospital (1 case) and on the farm (2 cases) and submitted for histopathology. All tumours were multilobular and composed of vaguely nested, bland, oval to slightly elongate cells with an infiltrative growth pattern. Mitotic activity was not detected. Immunohistochemical studies found that tumour cells were often positive for S-100 and cytokeratin and were occasionally positive for vimentin. Tumour cells were negative for glial fibrillary acidic protein, neuron specific enolase, synaptophysin, muscle actin, and chromogranin A. Follow up obtained from 7 months to 2 years following tumour removal indicated no evidence of regrowth or metastasis. The origin of these distinctive tumours is not clear, but the immunohistochemical profile suggests the possibility of origin from lingual taste buds. These cases and review of the literature indicate that successful surgical excision of tongue tumours can be performed by practitioners in private practice as well as by surgeons at referral hospitals.

Introduction

Tumours reported to involve the tongue in horses are rhabdomyosarcoma (Castleman *et al*. 2011; Hanson *et al*. 1993), squamous cell carcinoma (Schuh 1986), benign peripheral nerve sheath tumour (Schneider *et al*. 2010), atypical perineurial cell proliferative lesions (Vashisht *et al*. 2007), multiple myeloma (Markel and Dorr 1986), lymphoma (Rhind and Dixon 1999), mast cell tumour (Seelinger *et al*. 2007), and chondrosarcoma (Wilson and Anthony 2007). This report describes a previously unreported distinctive tumour arising in the tongue of horses. Despite locally invasive behavior all tumours were able to be completely excised, and excision appeared to be curative.

Case 1

A 23-year-old Quarter horse gelding was presented with a history of mild weight loss and difficulty masticating food. Oral examination revealed an ulcerated 3 cm diameter mass arising from the center of the dorsal surface of the caudal tongue (**Fig 1**). The mass was not obviously painful to the horse. The horse was referred to Oregon State University for surgery. The horse was premedicated with acepromazine by intravenous injection, followed 1 hour later by intravenous injection of xylazine. General anaesthesia was induced with 0.24 % (1.2 gram) ketamine hydrochloride in 10% guaifenesin, an endotracheal tube was inserted, and anaesthesia was maintained using inhaled isoflurane. With the horse in lateral recumbency a mouth speculum was placed and the mass was excised using a scalpel and electrocautery. Simple interrupted sutures using polyglactin 910 absorbable suture material were placed to appose the cranial edges. The caudal edge was judged to be too difficult to suture and was left open for second intention healing.

Following surgery the horse was fed soaked pelleted feed mixed with 1 gram of phenylbutazone twice daily, as well as hay and pasture grazing, for 1 week. The wound healed with no complications and the horse returned to use as a pleasure riding horse with a bitted bridle.

Case 2

A 14-year-old Standardbred gelding presented with a history of bleeding from the mouth. A 4 cm diameter mass was found on the dorsal surface of the tongue approximately 15 cm from the tip. Palpation did not elicit pain. A punch biopsy was obtained following local infusion of 2% lidocaine. Following biopsy diagnosis excisional biopsy was performed at the farm. The horse was sedated with a mix of intravenous xylazine and butorphanol and general anaesthesia was induced with intravenous valium followed immediately by intravenous ketamine hydrochloride. Surgery was performed with the horse in lateral recumbency. The mass was excised with a scalpel. Due to deep invasion general anaesthesia was prolonged with a second dose of intravenous valium and ketamine. The wound was closed with 2 layers of polydioxanone absorbable suture material in a simple interrupted pattern. The owners were instructed to feed a soaked mash for 2 weeks. Dehiscence of the superficial suture line was detected on examination 1 week later and the wound was left to heal by second intention healing. The horse returned to work in harness 1 month following surgery. At examination 22 months after surgery the site was healed with an irregular surface, although the tongue was approximately 6-8 cm shorter than before surgery (**Fig 2**).

Case 3

Case 3 was a 26-year-old Quarter horse mare, no longer being actively ridden, with no apparent clinical signs until the owner saw a 1.5 cm diameter mass on the left side of the dorsal surface of the tongue, approximately 4 cm from the tip. The mass did not appear to be painful to the horse. The mass was surgically excised at the farm following sedation with intravenous detomidine hydrochloride, placement of a mouth speculum, and injection of local anaesthetic (2% lidocaine) at the surgery site. Excision was achieved with a scalpel and the wound was closed with polydioxanone absorbable suture material in a simple interrupted pattern. The mare was on a diet of soaked pellets as well as pasture grazing due to age-related dental issues, and the same diet was continued after surgery. The site healed with minimal scarring.

Pathology

Grossly all 3 masses were raised, relatively round, ulcerated, slightly firm, and resembled granulation tissue. Sections from each were initially stained with haematoxylin and eosin. Based on findings sections were further stained with periodic acid-Schiff for glycogen, and argentaffin and argyrophil reactions, and immunohistochemistry for S-100[1], glial fibrillary acidic protein[1], neuron specific enolase[1], synaptophysin[1], muscle actin[1], vimentin[1], and chromogranin A[1] was performed. As findings did not lead to a definitive diagnosis, immunohistochemistry for cytokeratins[1] and for Melan A[1] was performed on representative sections from the tumours in Case 1 and 2; tissue from Case 3 was not available for these additional tests. Immunohistochemistry was performed using an automated immunostainer[1] and included appropriate positive and negative controls. All

antibodies reacted appropriately with positive control tissue except for Melan A, which did not recognize melanocytes in normal equine skin.

Histologic features of all cases were very similar. Epithelium overlying the tumours was often ulcerated, with associated granulation tissue, mixed inflammation, and superficially embedded plant material. Tumours were poorly defined and formed multiple lobules extending from close to the ulcerated surface into the underlying musculature (**Fig 3**). Lobules were composed of sheets of vaguely nested, relatively homogeneous and bland oval to elongate cells with round hyperchromatic nuclei and a moderate amount of clear cytoplasm (**Fig 4**). Tumour nests were supported by fine reticular stroma. Case 1 also had multiple foci of tumour necrosis. Mitotic activity was rare in all tumours, with no mitoses detected in 10 high power fields in any case. Cytoplasmic granules were not detected with PAS stain, and argentaffin and argyrophil stains did not detect neurosecretory granules. A small number of tumour cells in each case were positive for cytoplasmic vimentin. Tumour cells in all cases exhibited frequent strong nuclear reaction and occasional strong cytoplasmic reaction following incubation with S100 antibodies and strong cytoplasmic immunoreactivity to cytokeratins. All other antibody preparations were negative.

Normal tissue was present at surgical margins in cases 1 and 3. Only a small portion of the tumour was submitted from case 2, and additional tissue removed after obtaining the biopsy diagnosis was not submitted for histopathology.

Follow up

Follow up information was obtained for all horses. Case 1 was euthanized 7 months after biopsy diagnosis due to strangulation of the small intestine. The tongue was examined grossly and microscopically. Scarring was minimal and there was no gross or histologic evidence of tumour regrowth. Case 2 was examined at 2 years following tumour removal and case 3 was examined 1 year following tumour removal and there was no evidence of tumour regrowth in either case. Case 2 was reported to be dropping some grain while eating but was maintaining good weight and was otherwise clinically normal. No evidence of metastatic disease was detected in any horse.

Discussion

The histopathologic features of these 3 tumours were almost identical, but the tumour cell of origin remains unclear. No PAS positive granules to suggest granular cell tumour were found, immunohistochemistry findings do not suggest a tumour of endocrine or neuroendocrine cells (negative for chromogranin A, synaptophysin, and neuron specific enolase), muscle origin (negative for muscle actin), or peripheral nerve sheath origin (negative for GFAP). Although tumour morphology and behavior does not suggest melanoma, an atypical melanoma was considered but we were unable to replicate the findings of LeRoy et al. (2005), who reported positive reaction of equine melanocytes with Melan A antibody. However, an atypical melanoma would not be entirely ruled out even if tumour cells were negative, as a cytologically confirmed equine melanoma was reported to be negative for Melan A (LeRoy *et al.* 2005). The positive immunoreactivity for both S-100 (a neural marker) and cytokeratins (an epithelial marker) is unusual and

suggests the possibility of origin from lingual taste buds (Shin *et al.* 1995). Evaluation of tumour cell ultrastructure might be useful in determining cell of origin, but electron microscopy was not available for these cases.

Findings indicate that, despite a locally invasive growth pattern, successful surgical excision of these tongue tumours in horses can be achieved by veterinary practitioners in the field as well as by veterinary surgeons at referral hospitals. Prior reports indicate that excision of other equine tongue tumours can also be curative, as surgery was apparently curative in 2 horses with rhabdomyosarcoma of the tongue (Castleman *et al*. 2011; Hanson *et al*. 1993), in 1 horse with a tongue chondrosarcoma (Wilson and Anthon 2007), and in 1 horse with a mast cell tumour of the tongue (Seeliger *et al*. 2007).

In conclusion, surgery is a viable option for tumours of the tongue of horses, and surgical excision of these distinctive tongue tumours was apparently curative in the 3 horses in this report.

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Figure Legends

Fig 1. Horse tongue tumour, case 1. Image of the tumor on the dorsal surface of the tongue, taken during surgery.

Fig 2. Horse tongue tumour, case 2. Postoperative image of the tongue, taken 22 months after surgery.

Fig 3. Horse tongue tumour, case 3. Lobules of tumour cells (T) extend into skeletal muscle (M). The epithelium (E) in this area is intact. Haematoxylin and eosin. Bar = 100 μm

Fig 4. Horse tongue tumour, case 2. Tumour cells are relatively bland oval cells forming poorly defined nests. Haematoxylin and eosin. Bar = 50 μm.