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Concurrent Transitional Meningioma and Ceruminous Gland Adenocarcinoma in a Scottish wildcat hybrid (*Felis silvestris*)

S. J. Drew†, D. Perpiñán‡ and J. Baily†

†Easter Bush Pathology and ‡Exotic Animal and Wildlife Service, Royal (Dick) School of Veterinary Studies, The University of Edinburgh, Roslin, Midlothian, UK.

Corresponding author: Stephen J Drew, Easter Bush Pathology, Royal (Dick) School of Veterinary Studies, The University of Edinburgh, Roslin, Midlothian, UK. EH25 9RG; Tel: 0131 651 7455; Stephen.Drew@ed.ac.uk

Summary

The Scottish wildcat (*Felis silvestris*) is an iconic and endangered, sub-population of the European wildcat (*Felis silvestris silvestris*). As such, there is much research devoted to its’ ecology, genetics and conservation but little published information on pathology and disease. The investigation and reporting of such information is vital to furthering our understanding of the effects of hybridisation, a factor which is crucial if we are to secure a future for the Scottish wildcat. This report describes the clinical presentation, gross post-mortem and histological findings, in an elderly Scottish wildcat hybrid with concurrent transitional meningioma and ceruminous gland adenocarcinoma. To the authors’ knowledge there have been no previous reports of meningioma or ceruminous gland adenocarcinoma in the European wildcat (*Felis silvestris silvestris*) and there are only isolated reports of primary central nervous system neoplasia in other non-domestic felid species.
The Scottish wildcat is an isolated sub-population of the European wildcat (*Felis silvestris silvestris*) and is also considered by some scientists to be a distinct sub-species (*Felis silvestris grampia*) (Miller, 1912; Beaumont et al., 2001; Kilshaw et al., 2010). The future of the European wildcat, and particularly the Scottish wildcat, is uncertain due to habitat loss, hunting and persecution throughout the 18th and 19th centuries (McOrist and Kitchener, 1994), and more recently the effects of hybridisation, as a result of inter-breeding with domestic cats (*Felis silvestris catus*), (Daniels et al., 2001; Davis and Gray, 2010; Kilshaw et al., 2010), along with the potential transmission of common infectious diseases from domestic cats (McOrist et al., 1991). Now classified as endangered, they are legally protected under both UK and European legislation (Kilshaw et al., 2010). As such, the majority of published literature is focused on its ecology, genetics and conservation.

There have been a limited number of publications regarding pathology and disease of the European wildcat. Most publications are serological studies focusing on the prevalence of common feline viruses (McOrist et al., 1991; Watt et al., 1993; Daniels et al., 1999; Leutenegger et al., 1999; Millán and Rodriguez, 2009; Wasieri et al., 2009), although there are isolated reports dealing with other pathogens such as *Chlamydophila* sp. (Millán and Rodriguez, 2009), *Toxoplasma gondii* (McOrist et al., 1991; Herrmann et al., 2013), lungworm (Falsone et al., 2014), hemoplasmas (Willi et al., 2007), and other endoparasites (Burt et al., 1980; Krone et al., 2008). Neoplasia has been rarely reported in the European wildcat; from 79 necropsy examinations reported in several articles (Jefferies, 1991; McOrist et al., 1991; Watt et al., 1993; Krone et al., 2008; Wasieri et al., 2009; Hermann et al., 2013;
Falsone et al., 2014), only one tumour (a pulmonary lymphoma) was found (Hermann et al., 2013).

This report describes the presentation, gross post-mortem, and histological findings in a Scottish wildcat hybrid with concurrent transitional meningioma and ceruminous gland adenocarcinoma. To the authors’ knowledge, there have been no previous reports of meningioma or ceruminous gland adenocarcinoma in the European wildcat (*Felis silvestris silvestris*).

A 16.5 year old, neutered male, Scottish wildcat hybrid from a zoological collection reportedly suffered from sporadic episodes of incoordination and mild lethargy over a four week period. Although this individual was deemed to be a hybrid (*Felis silvestris silvestris x Felis silvestris catus*), rather than a true wildcat, according to the records of the zoological institution, a sample of skeletal muscle was tested using a 35 Single Nucleotide Polymorphism (SNP) marker test developed by the Royal Zoological Society of Scotland from a panel of markers published by Nussberger et al., (2013) in an attempt to confirm this and investigate the degree of hybridisation. Two extracts of DNA were conducted and three replicates of the assay were run but the assay failed to prove or disprove the hybrid status of this individual due to insufficient DNA quality, presumably due to sample degradation. The cat was transferred from a wildlife centre when it was 9 years old, and had since lived with a female Scottish wildcat in an outdoor enclosure. Prior medical history was unremarkable.

Husbandry and nutrition were considered appropriate for the species. Physical examination under general anaesthesia revealed loose skin, thought to be consistent with poor hydration or recent weight loss, a round bony proliferation on the right stifle joint and a hard mass behind the right ear. Body condition score was considered acceptable (4/9) with a weight of 4.5kg, teeth were in an excellent condition for an old cat and vital parameters (heart and respiratory rate, rectal temperature) were unremarkable. The cat initially appeared to recover well from
anaesthesia, but remained recumbent and died 2 hours later. The animal was subsequently submitted for post-mortem examination at the Royal (Dick) School of Veterinary Studies (The University of Edinburgh, Roslin, Midlothian, UK).

Post-mortem examination revealed the entrance to both external ear canals to be obscured by dark grey to blue black, well demarcated, multi-lobular, firm, occasionally cystic, exophytic, polypoid masses ranging from 1 to 5mm in diameter and a cream, well demarcated, 60x30x20mm, multi-lobular, firm, subcutaneous mass at the base of the right ear which contained a dark red, central, well circumscribed, depressed area 3mm in diameter on cut section. The right pre-scapular lymph node was moderately enlarged and the brain contained a cream to yellow, well demarcated, multi-lobular, expansile and compressive mass, approximately 15mm in diameter, between the cerebral hemispheres in the region of the falx cerebri (Fig.1). Samples of all gross lesions along with representative samples from all tissues and internal organs were collected, fixed in 10% buffered formalin and routinely processed according to current histological methods. Sections 5µm thick were stained with haematoxylin and eosin.

Histopathological examination of the external ear canal masses (Fig.2) revealed a moderately acanthotic, stratified squamous keratinising epithelium elevated by numerous, multi-focal, dilated, cystic, glandular structures lined by a flattened to low cuboidal epithelium. Many cysts contained amorphous, tan to pale brown, granular material (cerumen) and low numbers of large foamy macrophages also contained this granular brown material. Within a mass from the right external ear canal, the superficial dermis was focally expanded by more dense proliferations of polygonal to cuboidal cells forming tubules, acini and fronds. Cells had variably well-defined cell borders, a moderate amount of eosinophilic, finely granular cytoplasm, open-faced, oval to round nuclei with 1-2 prominent nucleoli. Anisocytosis and anisokaryosis were mild with an average of 1 mitotic figure per high power field (x400). Foci
of necrosis expanded the centre of these cellular proliferations. Examination of the right ear base mass (Fig.3) was consistent with the pre-existing architecture of a lymph node, 80% of which was effaced by tubules and acini of cells, similar to those described in the mass from the right external ear canal, with evidence of lymphatic invasion. Similar cell proliferation was present in the right pre-scapular lymph node.

The histopathological appearance of the auricular lesions was consistent with a diagnosis of bilateral ceruminous gland dilatation and hyperplasia (feline ceruminous cystomatosis) with unilateral (right side) ceruminous gland adenocarcinoma and metastasis to cervical and pre-scapular lymph nodes.

Sections of cerebral cortex, lateral ventricle and meninges showed the leptomeninges of the cingulate sulcus to be expanded by a large, densely cellular, well demarcated, finely encapsulated, compressive, nodular mass (Fig.4). The mass consisted predominantly of lobules of loosely to densely packed cells forming concentric whorls, occasionally surrounding blood vessels or containing central areas of necrosis. Adjacent cells formed long, interlacing fascicles streaming around the whorls and were supported by a loosely arranged, eosinophilic, fibrillar stroma. Cells were fusiform to polygonal with indistinct cell borders and variable amounts of eosinophilic, wispy, fibrillar cytoplasm. Within the centre of concentric whorls, cells adopted a more epithelioid morphology. Nuclei were round to elongated with loosely packed, finely stippled chromatin and 1 to 2 nucleoli. Anisocytosis and anisokaryosis were moderate with occasional multi-nucleate forms. Mitoses were rare (1 figure per 10 high power fields (x400)). Moderate numbers of neutrophils, occasional lymphocytes, plasma cells, pyknotic cells, small clusters of foamy macrophages and occasional acicular clear spaces (cholesterol clefts) were present within the mass. Both the grey matter and white matter tracts of the adjacent cerebral cortex contained mild diffuse vacuolation with mild perivascular clearing and spacing (oedema).
The histopathological findings of the cerebral mass were consistent with a diagnosis of transitional (mixed) meningioma containing features of both meningothelial (characterised by moderately cellular lobules of polygonal cells) and fibrous tumours (long interlacing fascicles of fusiform cells).

Also identified following post-mortem and histological examination were bilateral stifle joint osteoarthritis, nodular hyperplasia of both thyroid glands, mild hepatic lipidosis, mild, multifocal, chronic cholangiohepatitis and mild, multifocal, chronic interstitial nephritis with a focal, chronic, renal infarct.

Ceruminous gland adenocarcinomas are the most commonly diagnosed tumour of the external acoustic meatus in cats, accounting for up to 2% of all feline neoplasms (Njaa and Wilcock, 2012) and are more frequently diagnosed than adenomas in domestic cats (Moisan and Watson, 1996; London et al., 1996). They exhibit locally invasive behaviour (London et al., 1996) and metastasis to regional lymph nodes, lungs and viscera can occur in up to 50% of cases (Njaa and Wilcock, 2012). No evidence of pulmonary or visceral metastasis was found in this case.

Differentiation between ceruminous adenoma and adenocarcinoma can be challenging unless there is evidence of local invasion or metastatic disease, as in this case (Wilcock et al., 2002).

Meningiomas are the most common primary central nervous system (CNS) neoplasm of domestic cats (Koestner & Higgins, 2002; Troxel et al., 2003; Tomek et al., 2006; Motta et al., 2012) typically occurring in cats older than 9 years (Troxel et al., 2003; Tomek et al., 2006) with an increasing incidence with age. Domestic shorthaired cats seem to be predisposed but no significant sex predilection has been found (Troxel et al., 2003; Tomek et al., 2006). Transitional meningiomas, as reported here, and fibrous subtypes are most frequently encountered in domestic cats. Meningiomas are typically slow growing, with the exception of the uncommon anaplastic (malignant form), rarely metastatic and approximately
50% of cases do not exhibit any clinical signs. To the authors’ knowledge this is the first report of meningioma in a European wildcat. There are only isolated additional reports in the literature of central nervous system neoplasia in non-domestic felids, such as meningioma in a Bengal tiger (*Panthera tigris tigris*) (Akin et al., 2013) and intracranial oligodendroglioma in a lion (*Panthera leo*) (Tucker et al., 2008).

The findings reported here pose the question for future studies to determine the incidence of common neoplasms of domestic felines in pure-bred wildcats and their hybrids. This may help to elucidate whether their occurrence may be an associated effect of hybridisation, or purely a reflection of the increasing age of individuals living in zoological collections in comparison to their free living relatives.

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Conflict of Interest Statement: The authors declare no conflict of interest.

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Legend for Figures:

Fig 1. Brain, showing the presence of a nodular mass between the cerebral hemispheres.
Fig 2. Section of tissue from the right external ear canal showing acinar arrangement of neoplastic epithelial cells with central necrosis. Epithelial cells are present within the lumen of the thin-walled vessel at the top of the image. HE.
Fig 3. Section of the right cervical lymph node showing replacement of normal architecture by metastatic neoplastic epithelia forming acinar structures. HE.
Fig 4. Section from the brain mass showing whorls and bundles of neoplastic spindle cells consistent with the transitional form of meningioma. HE.