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Feline lungworm infection. An emerging concern?

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Abstract

Infectious feline upper respiratory tract disease is very common and *Aelurostrongylus abstrusus* is the most often diagnosed parasitic cause, although clinical signs may go unnoticed and diagnosis is challenging as it has several limitations.

Noting that more cases have been recorded in recent years, the authors, in the present work, describe in detail the features of five cases of *Aelurostrongylus abstrusus* registered in their work and influence area, to alert animal health professionals to this emerging concern.

Keywords: feline, lungworm infection, *Aelurostrongylus abstrusus*, *A. abstrusus*

Introduction

Infectious feline upper respiratory tract disease (URDT) is very common and has several causes, including viral and bacterial agents in kittens (≤ 12 months of age – paediatric age), increased by a still immature immune system and absence of complete vaccination plans, as well as lungworms also in kittens (*Troglostrongylus brevior*; < 12 m) and adults (*Aelurostrongylus abstrusus*, > 12 m) (Table 1) (Cavalera *et al.*, 2018).

Both lungworms *Troglostrongylus brevior* and *Aelurostrongylus abstrusus* are Nematodes from Sercenentea subclasse, Strongylyda order and Metastrongyloidea superfamily, being the last one better-known, since it is more often diagnosed (Table 2). Both share the same definitive (domestic and wild cats), intermediate (gastropod like snails and slugs) and paratenic hosts (rodents, frogs, reptiles and birds that feed on infested gastropods), in these bypassing even the seasonality (Giannelli *et al.*, 2017; Cavalera *et al.*, 2018; Gueldener *et al.*, 2019).

Other lungworms found in the respiratory tract, despite being less frequent in European studies, also include *Eucoleus aerophilus*¸ *Angiostrongylus chabaudi* and *Oslerus rostratus*, (Table 1) (Cavalera *et al.*, 2019), the former and the latter also detected in Portugal (Waap *et al.*, 2014, Giannelli *et al.*, 2017).

Table 1. EURTD aetiological agents

Agent		**Taxonomy**
Virus	Feline Herpesvirus 1	
	Feline Calicivirus	
Bacteria	*Bordetella bronchiseptica*	
	Chlamydofila felis	
	Mycoplasma felis	
Parasites	*Troglostrongylus brevior*	Metastrongyloidea; Crenosomatidae
	Aelurostrongylus abstrusus	Metastrongyloidea; Angiostrongylidae
	Eucoleus aerophilus	Trichuroidea; Capillariidae
	Oslerus rostratus	Metastrongyloidea; Filaroididae
	Angiostrongylus chabaudi	Metastrongyloidea; Angiostrongylidae

(Adapted from: Waap *et al.* (2014), Giannelli *et al.* (2017), Cavalera *et al.* (2018), Cavalera *et al.* (2019)).

Regarding the life cycle of those metastrongyloid nematodes, infected cats shed the first larval stage (L1) in faeces, which is ingested by the intermediate host, developing up to the third stage (L3). When cats ingest intermediate and paratenic hosts, during their predatory activity, L3 penetrate the intestinal mucosa, moult into L4 and reach lungs via lymphatic system where develop adult worms, which by breeding, giving eggs that hatch in L1 (Gueldener *et al.*, 2019).

Alternative transmission pathways include the release of L3 in mucus and/or from dead snails submerged in water and snail-to-snail transmission, named *intermediesis*. Also, vertical transmission from the queen to the litter has been hypothesised. Thus these transmission pathways has been contributing to the increase of lungworm exposure in both, wild and domestic, felines (Cavalera *et al.*, 2019).

The most common signs are chronic cough, sneezing, nasal discharge and dyspnoea (Giannelli *et al.*, 2017) but unspecific clinical signs are also common and include anorexia, lethargia, weight loss, enlarged lymph nodes and diarrhoea (Cavalera *et al.*, 2019). Also, cats infected with *A. abstrusus* could not have manifested respiratory signs though they had radiographic changes in their lungs (Genchi *et al.*, 2014).

The diagnosis of feline lungworms are coproscopic procedures relying detection of L1 in faecal samples, being the Baermann technique the gold standard for diagnosis, but it has several limitations leading to the underestimation of the number of effectively diagnosed cases. Also, prevalence rates of infections in European countries are highly variable and very difficult to compare due to different study designs, although thus represent a topic of increasing interest in the field of veterinary sciences (Giannelli *et al.*, 2017; Gueldener *et al.*, 2019). These limitations are linked to several reasons listed below:

- the fact that regular deworming erroneously excludes this hypothesis of diagnosis;

- the absence of diagnostic cases, erroneously leads to thinking that the region is endemic;

- Baermann technique, the most employed test, has long time of execution (24 h), requires fresh and individual faecal samples, operator skills in the identification of L1 species and could revealed lack of sensitivity with low parasite burdens;

- the diagnosis is based on larvae identification but during the pre-patency, larvae can be absent in faeces, and, even in the patency, the larval shedding can be intermittent (e.g. in the case of immature or single-sex infections), even in presence of clinical signs, but especially in chronic infections;

- obtaining fresh faecal samples in animals with outdoor access, those with higher risk of infection, where they defecate;

- overlapping morphological features between lungworm from parasitic different species larvae, namely between *T. brevior* and *A. abstrusus* (Table 2);

- the clinical signs can be unspecific or ranging from asymptomatic to very severe respiratory stress, even lead to death but are often mild and subtle, so that the disease often stays unobserved (Waap *et al*., 2014; Soares *et al*., 2017; Cavalera *et al*., 2019; Gueldener *et al*., 2019);

- when clinical signs are present, they are often, at least initially, misinterpreted (e.g. as feline asthma) (Gueldener *et al*., 2019).

- diagnoses based on the collection of a single faecal simple per animal which may not contain larvae (Giannelli *et al*., 2017).

The limitations listed can be overcome by combining an highly specific and sensitive serological test (ELISA) for lungworms, since seroconversion occurs at the earliest 15 days post-infection and may persist 4-8 weeks after anthelmintic treatment which represents a valid method allowing reliable results to epidemiological surveys (Gueldner *et al*., 2019; Cavalera *et al*., 2019).

Table 2. Lungworms characteristics

Agent	Feline age	Respiratory system habitat	Main morphological differences	
			Anterior extremity	**Posterior extremity**
T. brevior	< 12 m	Bronchi Bronchioles	Clear and pointed With a subterminal oral opening	Tail featured a deep dorsal incision, which divided the extremity into two appendices (a shallow ventral one and a slender dorsal one) Tail without obvious S-shape Ending straight and gradually tapered
A. abstrusus	> 12 m	Bronchioles Alveolar ducts Alveoli	Slender With a short and terminal oral opening leading into a narrow vestibule	Tail bent into an S-shape With a visible dorsal kink, distinct deep dorsal and ventral incisures Terminal knob-like extremity

Objectives

The aim of the present work was to describe in detail the features of *A. abstrusus* as one of the main causes of URDT, since it is a frequent worldwide feline lungworm, with cases registered in our work and influence area, to alert animal health professionals to this emerging concern.

Material and Methods

Five clinical cases, summarized in Table 3, from the centre of Portugal (Viseu district) were presented to the Laboratory of Pathological Anatomy Veterinary (LPAV) for complementary diagnostic purposes.

Exclusion criteria were cases with lesions strongly compatible with lungworms but which evidence had not been confirmed by microscopic observation.

In cases nº 1, 3 and 4 a complete necropsy was performed and samples of several organs were collected, immediately fixed in 10 % buffered formalin solution, embedded in paraffin wax and 3 µm thick sections were stained for routine histopathological diagnosis with Haematoxylin and Eosin (HE).

In case nº. 2 a standard DiffQuik Staining Procedure (DF), according manufacturer's instructions was performed.

Results

All the clinical cases were from male cats and only two presented age-related information, which was very different: 2 years and 13 years; respectively (Table 3).

Only one clinical case presented description of specific respiratory symptoms, dyspnoea, and it was the case that had lungworm infection as the main diagnosis. All others revealed nonspecific symptoms (Table 3).

Of these four clinical cases, three died and *A. abstrusus* lesions were the direct and immediate cause of death, although they were not considered the primary diagnosis (Tables 3 and 4).

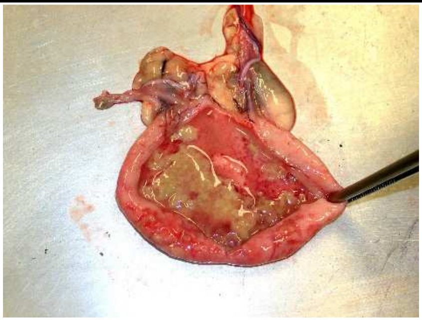
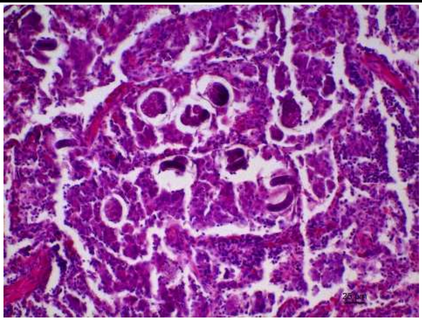
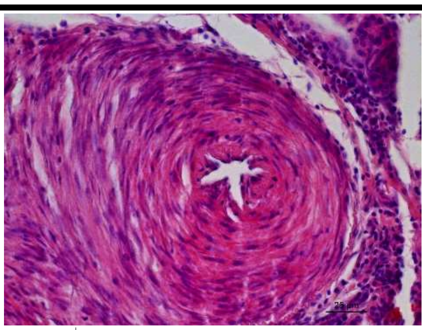
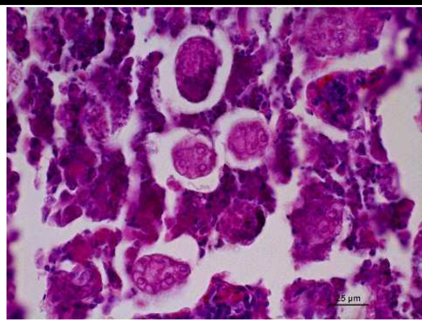
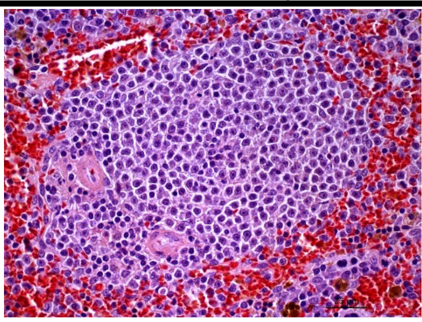
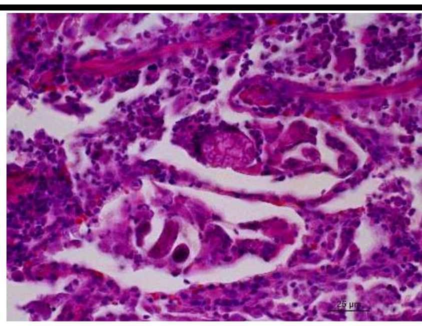
The main lesions and parasite characteristics that led to the diagnosis are summarized in Table 4 and included mixed inflammatory cell infiltrate, rich polymorphonuclear neutrophils, rarely eosinophils, macrophages and lymphocytes were present in lung parenchyma of the tree cases submitted to anatomopathological study, with areas of alveolar compensatory emphysema. Hyperplasia of peribronchial glands was also seen, as well as desquamation of the bronchiolar epithelium, smooth muscle hypertrophy in the walls of bronchioles and alveolar septa. However the most evident lesion was thickening of the smooth muscle in the tunica media of lung arterioles.

Table 3. Clinical cases description

Case nº	Gender	Age	Clinical signs	Main diagnosis
1	M	w/d	Hind limb locomotion loss	CRF and cystitis
2	M	2y	Dyspnoea	SCC
3	M	w/d	BW loss Dehydration	*A. abstrusus*
4	M	13y	BW loss Seizures Obesity	Lymphoma

w/d, without data; HL, Hind Limb; BW, Body Weight; CRF, Chronic Renal Failure; SCC, Squamous Cell Carcinoma.

Table 4. Clinical cases graphical description

Case nº	Main lesion	*A. abstrusus*
1	 Bladder wall thickening with necrosis and fibrin deposition.	 Morulated eggs; cross and longitudinal sections of adult worms in alveolar lumina, encircled by an abundant inflammatory infiltration (HE).
2	 Spherical aggregates of neoplastic epithelial cells revealing pleomorphism and anisocariosis (DF).	 Larvae with clear and pointed anterior extremity, with a visible dorsal kink and curved posterior extremity (DF).
3	 Marked hyperplasia of the middle tuna of the pulmonary arterioles tunica media, some of which with obliterated or irregular lumen (HE).	 Morulated eggs in alveolar lumina, encircled by an abundant inflammatory infiltration (HE).
4	 Undifferentiated lymphoid population with eucromatic nuclei, distinct nucleoli and angular edges, occupying splenic germinal centers (HE).	 Cross and longitudinal sections of larvae adult worms in alveolar lumina, encircled by an abundant inflammatory infiltration (HE).

Discussion

Although all clinical cases were in male cats, we subscribed that no statistical association was verified until now, regarding prevalence of lungworm infection and gender or neutering status, even though intact male cats were considered at higher risk in the past (Cavalera *et al.*, 2019; Gueldner *et al.*, 2019), even due to the limited number of cases of this work.

From the most common respiratory specific signs described we have recorded only dyspnoea in the clinical case nº. 2, which is consistent with *A. abstrusus* asymptomatic infections or with unspecific signs, underline the importance of taking these infections seriously because they represent a highly relevant cause of death, particularly during anaesthesia procedures (Gueldener *et al.*, 2019).

Death occurred in the other three clinical cases, in accordance with descriptions that death could also have occurred due to high infection burdens (Giannelli *et al.*, 2017) and although in most cases lungworm infection emerged as a secondary diagnosis.

Cats with regular deworming should also be considered at risk, as chemoprevention requires specific agents (spot-on preparations containing emodepside, eprinomectin, moxidectin and selamectin) which could eliminate larval shedding by at least 90% but only if part of a wider parasite control strategy and only theoretical. It should be noted that stray or feral cats are consistent reservoirs, which are also the animals with the highest infection rates, contributing to the maintenance of the *A. abstrusus* life cycle. Eprinomectin and moxidectin are promising drugs against pre-patent developmental larvae stages but still under study (Elsheikhaa *et al.*, 2016).

The recommended effective anthelmintic treatment is fenbendazole (50 mg/kg sid) performed for 5 days (Soares *et al.*, 2017).

The presence of these parasitic forms at the lungs triggers an inflammatory reaction and parenchymal tissue damages, lesions noted in all the cases submitted to histopathological evaluation (Giannelli *et al.*, 2017; Gueldener *et al.*, 2019). Hyperplasia of peribronchial, desquamation of the bronchiolar epithelium, smooth muscle hypertrophy in the walls of bronchioles and alveolar septa were also lesions observed, in line with other authors (Traversa *et al.*, 2014). However the most evident lesion observed, the thickening of the smooth muscle in the tunica media of lung arterioles was not evident in the study of Traversa *et al.* (2014) having been, in our opinion, largely responsible for the poor outcome of three of the reported cases.

Regarding *A. abstrusus*, recent studies have shown an apparent increase in clinical cases in several countries and regions (Soares *et al.*, 2017), including our work and influence area, which is probably indicative of the growing importance of these agents (Soares *et al.*, 2017), requiring the attention of animal health professionals to the introduction of new sanitary control measures, even in areas registered as non-endemic, although most adopted cats have an increasingly indoor lifestyle.

These findings are probably related to climate and ecological changes in recent years, where rising winter temperatures (above -2 ° C) allowed the dissemination of populations of intermediate hosts. Notwithstanding *A. abstrusus* L1 can outlive freezing and thawing for several times, on the other hand experimental infections of *Helix aspersa* with *A. abstrusus* L1 showed that warmer temperature conditions (18.8–29.5 °C) and constant humidity allow a higher rate of larval development in the snails (Cavalera *et al.*, 2019; Gueldner *et al.*, 2019).

In the same direction, Gueldner *et al.* (2019) obtained higher prevalence of *A. abstrusus* in humid and low altitude areas, which may be correlated with the presence intermediate or paratenic hosts. Snails and slugs are poorly mobile organisms which cannot evade unfavourable environments over long distances and ideal habitats are represented by wetland biotopes (natural streams, rivers, marshland, alluvial zones), which offer cover (in form of vegetation such as undergrowth and reed beds) from potential predators, solar radiation and constitute sustainable food resources (Gueldner *et al.*, 2019).

Concerning the expected paratenic hosts, the correlations between *A. abstrusus* prevalence and the occurrence of paratenic hosts appears speculative, as information on amphibians, reptiles, rodents or birds infected with larval stages is very scant (Gueldner *et al.*, 2019).

Finally, some studies have found statistical association with lungworm infection and Feline Immunodeficiency Virus (FIV) infection, as expected, but this association reinforces the likelihood of being animals with outdoor access, since the main mode of transmission of FIV is through cat-to-cat contact, namely by bites (Cavalera *et al.*, 2019).

In the same way, Gueldner *et al.* (2019) did not find seroprevalence significantly higher, as expected, in outdoor cats, attributing this result to the fact that the indoor cats had access to a balcony or a terrace, where they could also ingested intermediate hosts.

Conclusions

The authors consider that the registration of 4 clinical cases in only three years in the central region of Portugal, Viseu district, constitute an alert that requires passing to take into serious consideration this hypothesis in the differential diagnosis.

These cases indicate that the biological cycle has the potential to develop in this region, fact that it is most likely due to the rise in winter temperatures and the absence of snow in last year's enabling intermediate and paratenic hosts' survival.

Therefore, it will be crucial, in the very short term, to initiate strategies that include prophylactic measures to control the biological cycles of common lungworms, especially given the role of stray cats, still in large numbers in our country and region, and those with free outdoor access.

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