

A Case of *Eucoleus aerophilus* Infection in Jeju Weasel (*Mustela sibirica quelpartis*)

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Abstract: A dead male Jeju weasel with clinical signs of severe dehydration and moribund was submitted for the necropsy. Grossly, purple-red ecchymoses were scattered in the surface of the entire lungs. Milky-white mucoid materials occupied in the left bronchial lumens. Histopathologically, an adult worm with thick cuticle layer surrounded by chronic inflammatory cells was embedded in the left caudal lung lobe. Numerous oval or barrel-shaped parasitic eggs with muco-purulent exudates were observed in the dilated bronchus of another section of left caudal lobe. Operculate parasitic eggs showed typical morphologic characteristics such as barrel shape with asymmetry bipolar plugs and thick bi-layered wall. Based on the histopathologic findings and morphological characteristics of parasitic eggs in the pulmonary tissue section, this case was diagnosed as verminous pneumonia associated *Eucoleus aerophilus* infection in Jeju weasel.

Key words: Eucoleus aerophilus, Jeju weasel, lung, parasitic eggs, verminous pneumonia.

Introduction

Eucoleus (E.) aerophilus, previously known as Capillaria (C.) aerophila, is a little known trichuriod parasitic nematode affecting cats, dogs and wild carnivores (18). A few cases of human infection have also been reported (7). The adult lungworms live embedded in the epithelium of trachea, bronchi and bronchioles of the definitive host (10,14). The female worms lay eggs, which are subsequently coughed up, swallowed and release via feces into the environment (2). Basic knowledge for the life cycle of this nematode is scanty and, for instance, the route of transmission to vertebrate hosts remains to be understood (8). In general it is thought that animals become infected by ingesting environmental larvated eggs, but it has been also hypothesized, but never demonstrated, that larva might require the passage through earthworms to become infective for definitive hosts (8,15).

E. aerophilus has been recently described in pets from many countries of Europe and America (3,4). Canine and feline respiratory infection by *E. aerophilus* is considered sporadic and/or subclinical, but clinical cases characterized by bronchovesicular sounds, sneezing, wheezing, and chronic dry productive cough have been reported (1,2).

According to survey for fecal materials, *E. aerophilus* infestation was first reported in a fox reared in Korean Zoo in 1977 (9). However, there was no available data for the *E. aerophilus* infection in domestic and wild animals in Korea. In this study, we describe a case of *E. aerophilus* infection in a Jeju weasel (*Mustela sibirica quelpartis*) in Jeju. To the

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author's knowledge, this is the first case of *E. aerophilus* infection in wild animal in Korea.

Case

A moribund male Jeju weasel with severe dehydration was rescued at the near area of the Seogwipo Natural Recreation Forest (Fig 1). In spite of the fluid therapy, the weasel died three hours after rescue. The weasel was submitted to the Veterinary Pathology Laboratory in Jeju National University. At necropsy, purple-red ecchymoses were scattered in the surface of the entire lungs. Milky-white mucoid materials occupied in the left bronchial lumens.

Samples taken from the major parenchymal organs were fixed in 10% phosphate buffered formalin, processed in a routine manner, embedded in paraffin and stained with hematoxylin and eosin (H&E). Morphometric analysis for 10 barrel-shaped parasitic eggs in paraffin section of lung was performed using a Leica DM LB2 microscope and a Leica DFC 280 digital camera supported by the software Leica LAS V 3.7. Unfortunately, fecal materials were not collected at necropsy.

Histopathologically, the lungs showed diffuse congestion, hemorrhage and atelectasis. An adult worm with thick cuticle layer surrounded by chronic inflammatory cells such as macrophages and lymphocytes was embedded in the left caudal lung lobe (Fig 2). Three parasitic eggs were scattered in the surrounding inflammatory exudates, adjacent area of adult worm. Numerous oval or barrel-shaped parasitic eggs with muco-purulent exudates were observed in the dilated bronchus of another section of left caudal lobe (Fig 3). Multifocal hyperplasia, desquamation, degeneration, and necrosis



Fig 1. The Jeju weasel showed severe dehydration and mori-



Fig 2. A parasitic adult worm (W) was embedded in the pulmonary parenchyma. H&E. Bar = $500 \mu m$.

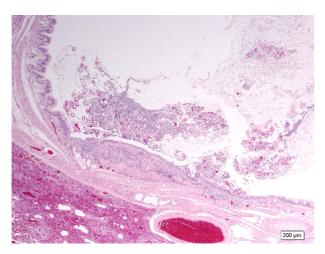


Fig 3. Cystic dilated bronchus contained many parasitic eggs with muco-purulent exudates. H&E. Bar = $200 \mu m$.

were presented on the lining epithelial cells of bronchus (Fig 4). Chronic inflammations also existed around blood vessels in lamina propria of bronchus. Operculate parasitic eggs showed typical morphologic characteristics such as barrel shape with asymmetry bipolar plugs and thick bi-layered (dense inner, striated outer) wall (Fig 5). The length of major

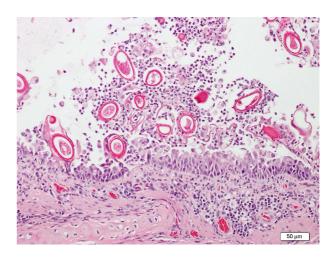


Fig 4. Parasitic eggs were oval or barrel-shape. Note hyperplasia, desquamation, degeneration, and necrosis of the lining epithelium in bronchus. H&E. Bar = $50 \mu m$.

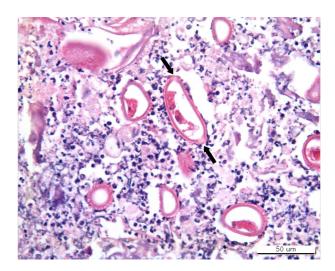


Fig 5. Barrel shaped parasitic eggs had asymmetry bipolar plugs (arrows) and thick bi-layered (dense inner, striated outer) wall. H&E. Bar = $50 \mu m$.

and minor axes of parasitic eggs were 43.1 \pm 5.8 μm (ranged 36.1 to 53.4) and 22.7 \pm 4.2 μm (ranged 16.2 to 28.6), respectively.

Based on the histopathologic findings and morphological characteristics of parasitic eggs in the pulmonary tissue section, this case was diagnosed as verminous pneumonia associated *E. aerophilus* infection in Jeju weasel.

Discussion

After ingestion of embryonated eggs of *E. aerophilus* in the definitive host, the ova hatch in the intestine, and the larvae penetrate the intestinal wall and migrate via bloodstream or the lymphatics to the lungs, where they invade the mucosa and reach adulthood (13). Their sexual maturity takes about 3-6 weeks post infection (2,17). The adult stages of *E. aerophilus* (length ~20-30 mm, width ~60-180 µm) are slender, whitish and filamentous nematode (8,14). The parasite can

induce the damage of pulmonary parenchyma and chronic bronchitis in dogs and cats. In cases of heavy parasitic burden or bacterial complications, the diseases can lead to mortality due to bronchopneumonia and respiratory failure (2,15,18).

Reported infestation rates of *E. aerophilus* in dogs and cats which are kept as pets in Europe and North America are generally less than 10% (15,18). However, a recent study from Italy showed the occurrence of clinical signs in 87.5% and 72.7% of dogs and cats, respectively, indicating that the prevalence of clinical disease might be underestimated (18).

E. aerophilus has been found on every continent, and occurs in a wide range of wild animals including martens, wolves, badgers, ferret, raccoons, raccoon dogs, coyotes, foxes, hedgehogs, and other carnivores (6,7,12). The red fox is considered the most important reservoir host in the wild environment (2,6,10). Several epidemiological studies showed that as many as 68.6% of red foxes in Canada and 84% of foxes in Serbia were parasitized with *E. aerophilus* (8,10).

Clinical disease was common in foxes farmed for fur where it was associated with poor husbandry practices (1). Infected animals suffered chronic respiratory disease with poor growth and decreased fur quality. Heavy infections could lead to significant mortality because of bronchopneumonia (10). The exact pathogenic role of *E. aerophilus* in wildlife is not well recognized, though this parasite has been considered as an agent of massive mortality in farmed foxes (8). In the previous report, authors demonstrated that a very low inflammatory response around the worms could be the result of a low pathogenicity or an immunosuppression in the mucosa caused by the parasite in the foxes (8). In this case, the Jeju weasel showed severe dehydration and in a moribund state without any respiratory signs such as sneezing, wheezing, and cough. Therefore, these clinical signs might be not closely related with E. aerophilus infection in this Jeju weasel.

According to the histopathologic survey in red foxes, microscopic lesions associated with *E. aerophilus* were typically centered around the bronchi and bronchioles (10). The bronchi frequently contained *E. aerophilus* alone or admixed with serocellular exudate. The bronchial mucosa showed variable degrees of edema and hyperplasia of the bronchial associated lymphoid tissue (BALT) and also contained numerous eosinophils infiltrating the submucosa. *E. aerophilus* was predominantly located in the caudal lung lobes of foxes. In this case, adult worm with chronic inflammation and parasitic eggs admixed with muco-purulent exudates were observed in the large bronchi of the left caudal lobe. However, the BALT hyperplasia and the infiltration of eosinophils were not obvious in Jeju weasel case.

Indeed, pulmonary capillariosis in humans is an occasional event, with only 12 cases described to date. Human infection may be transmitted directly by the consumption of vegetables contaminated parasitic eggs (7). Human cases of this disease have been documented in Russia and the Ukraine (eight cases), Morocco (one case), Iran (one case), France (one case), and Serbia (one case) (7). The clinical symptoms of human infection are bronchitis, coughing, mucoid or blood-tinged sputum, fever, dyspnea, and eosinophilia (7,15). The last described report was a cryptic case of infection in a woman from Serbia that resembled a bronchial carcinoma

(7). In that paper, more in depth analysis revealed the presence of *E. aerophilus* eggs in the bronchial biopsy, thus suggesting that the parasites likely died in the bronchial tree, causing abscesses which appeared as tumor-like lesions. Large chronic inflammatory focus composed of central adult worm and surrounding macrophages and lymphocytes was also demonstrated in the *E. aerophilus* case of Jeju weasel.

Canine and feline capillariosis is diagnosed by the detection through standard fecal flotation of the typical trichuroid eggs passed in the faeces by the infected hosts (15). This approach is the most common and the least expensive in routine practice, but the eggs of E. aerophilus need to be differentiated from the eggs of intestinal whipworm Trichuris (T.) spp. When the barrel-shaped eggs are found in samples following copromicroscopic analysis with flotation solutions (specific gravity = 1.2-1.35) their shell wall surface pattern, size and plug morphology must be carefully examined to achieve a reliable diagnostic result (15).

Under the microscopy, E. aerophilus eggs showed a typical morphology, as they were barrel-shaped and presented asymmetry of bipolar plugs (17). The egg walls had a network of anastomosing ridges and bridges. The lengths of major and minor axes of E. aerophilus eggs from dogs and a cat were $64.91 \pm 1.11 \, \mu m$ to $65.04 \pm 1.50 \, \mu m$ and from 34.89 \pm 3.34 µm to 36.96 \pm 3.15 µm, respectively. Conversely, eggs of T. vulpis were bigger (size ranging from 72-94 µm long and 31-42 µm wide) than those of E. aerophilus and exhibited a brownish, thick and smooth shell with symmetrical mucoid plugs (17). In that paper, authors stressed that the net-like wall of E. aerophilus eggs being the most important key features to identify the eggs and comparison for T. vulpis eggs. In the present histopathologic study for Jeju weasel, barrel-shaped parasitic eggs had typical asymmetry bipolar plugs and dense inner and striated outer bi-layered wall. The mean length of major and minor axes of parasitic eggs were $43.1 \pm 5.8 \ \mu m$ and $22.7 \pm 4.2 \ \mu m$. The overall size of parasitic eggs in the present was smaller than those of E. aerophilus in previous literatures (15,17). These results may be closely associated with tissue fixation and processing for histopathologic examination. Generally, formaldehyde fixation of tissue and tissue processing protocol, alcohol dehydration, clearing in xylene, and infiltrating with paraffin produced as much as a 20% decrease in linear dimension of the tissues (5).

Recently, the efficiency of a seminested PCR for the specific molecular identification of *C. aerophila* in naturally infected dogs and cats also has been demonstrated (3).

Little information has been published on anthelmintic treatment of capillariosis (15). Oral fenbendazole (50 mg/kg, daily for 14 days) was reported to be effective in the treatment of one dog infected with *E. aerophilus* (2). Specific treatment protocols have not been evaluated but fenbendazole, levamisole and ivermectin have been used successfully in cats (1). A recent experimental prospective study demonstrated that imidacloprid 10% / moxidectin 1% (Advocate; Bayer Animal Health) spot-on formulation is safe and effective on the treatment of feline lung capillariosis caused by *E. aerophilus* (16).

Wild carnivores highly infected with *E. aerophilus* are infection reservoirs for dogs and cats in urban areas (6). In a

previous study, distinct genetic population of *E. aerophilus* was discovered in Europe and out of total 15 haplotypes characterized (haplotypes I-XV), five were shared between pets such as dogs and cats and wildlife including foxes and beech martens (4). This finding indicated the existence of common patterns of transmission between wild and domestic carnivores in the same areas (4). As the disease has a zoonotic character, the risk of human infection, through cats and dogs is possible.

The Jeju weasel is one of the principle predators in Jeju and is valuable in maintaining ecological balance in wildlife populations. The Jeju weasel is distributed equally from low-lying ground of Hallan Mountain up to 1,600 meters (11). These animals are usually observed in the surroundings of private house, and occasionally found in farmland, fruit farm, and road. Therefore, Jeju weasel may be important reservoir for certain diseases and parasites such as *E. aerophilus* transmissible to domestic animals. More in-depth study for *E. aerophilus* infection in wildlife should be warranted to clarify the ecologic importance and to develop the preventive measure of zoonotic disease.

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