

[Research Note]

**First record of *Cyathostoma*
(*Hovorkonema*) sp. (Nematoda:
Syngamidae) from a red-crowned
crane, *Grus japonensis*, in Kushiro,
Hokkaido, Japan**

Tomoo Yoshino^{1,2}, Hiroko Iima^{1,2}, Fumio Matsumoto¹
and Mitsuhiko Asakawa^{2*}

A red-crowned crane, *Grus japonensis*, chick died on August 6, 2013, in Kushiro, Hokkaido, Japan. In the postmortem examination, 20 individual nematodes were found in the nasal cavity and anterior thoracic air sacs. The chick died from severe inflammation of the air sacs, granulomatous pneumonia, and esophageal strangulation, caused by a heavy nematode infection. The nematodes were identified as *Cyathostoma* (*Hovorkonema*) sp., based on their measurements and morphological characteristics. This is the first host record of these nematodes from *G. japonensis* and the first geographical record from Japan. Considering the life cycle, the chick was infected with the nematodes by ingestion of earthworms, because the chick had been given many earthworms as food from the parents. Nematol. Res. 49(1), 7–11. (2019)

Key words: air sac inflammation, esophageal strangulation, gapeworm, granulomatous pneumonia, paratenic host

The red-crowned crane, *Grus japonensis* (Moller, 1766), is an endangered waterfowl and is distributed in China, Russia, Korea, and Japan (Hokkaido) (Masatomi, 2000). Several activities, such as artificial breeding, rescue, and postmortem, have been performed to conserve the crane at the Kushiro Zoo, since the zoo was established (Iima, 2016; Yoshino, 2017).

A female crane chick hatched at the Kushiro Zoo, Kushiro, Hokkaido, Japan, on July 4, 2013. Until about one week after hatching, the chick had grown steadily, but from a week old, it showed several symptoms, such as eyeball protrusion, an increased amount of eye mucus, a head swing, and neck tilt. Also from 12-days-old, the chick experienced symptoms, such as gloom and head shaking, and its growth seemed to be slow. About three weeks after hatching, it lost its voice, no longer took bait

on its own, and would not eat any food provided by the parents. As the chick appeared to be debilitated, the chick was captured for treatment. However, the chick showed open breathing, severe head shaking, lethargy, pulmonary noise was heard when breathing, and bait was expectorated with her mouth wide open. Then, gradually the lethargy increased, such that the chick was unable to stand and developed head drooping, and the chick died on August 6. In the postmortem examination performed at the Kushiro Zoo, 20 individual nematodes were obtained from the nasal cavity and the intraclavicular and anterior thoracic air sacs. The collected nematode specimens were fixed in 70% ethanol, and the localized gross regions of the organs were fixed in a 15% formalin solution. The specimens were taken to the Wild Animal Medical Center of Rakuno Gakuen University for taxonomical and microscopic examination. The nematode specimens were cleared in a lacto-phenol solution. The organs were sectioned (4 or 5 μ m thickness), stained with Hematoxylin and Eosin (HE), and mounted using Canada balsam solution. Morphological and biometric data were recorded using a camera lucida (OLYMPUS DP20). The specimens have been deposited at the Kushiro Zoo (Postmortem No. 130807R330).

The chick was remarkably thin and had no subcutaneous fat. Several nematodes pierced the wall of the air sac and formed a granuloma; the esophageal wall adhered with the thickened air sac, and the esophagus was strangulated (Fig. 1). Histologically, severe inflammation, due to eggs and nematodes on the cervical, intraclavicular, and anterior thoracic air sacs, subacute granulomatous pneumonia, and emphysema centered on the eggs were observed (Fig. 2).

The nematodes obtained from the respiratory systems of the chick had the following morphological characteristics: the inner wall of the buccal capsule was smooth, and six teeth were arranged at the base; a buccal collar was absent; the cuticle was smooth; the males and females were separated; the males had strongyloform bursa and externo-dorsal rays that were markedly shorter than the medio-dorsal ray, without medial projection that did not extend beyond the margin of the bursa copulatrix; and the spicules were thin and equal. According to the criteria from Baruš *et al.* (1978) and Lichtenfels (1980), the nematodes were identified as *Cyathostoma* (*Hovorkonema*) sp. (Strongyloidea: Syngamidae). Their morphological characteristics and measurements are given in Fig. 3 and Table 1.

The syngamid nematodes are called gapeworms and

¹ Kushiro Zoo, Kushiro, Hokkaido 085-0201, Japan

² School of Veterinary Medicine, Rakuno Gakuen University, Ebetsu, Hokkaido 069-8501, Japan

*Corresponding Author: e-mail: askam@rakuno.ac.jp

are well known as a pathogenic agent of wild and captive birds, and they can sometimes be a contributing cause of death. If hosts are infected with over 10 individuals of the nematodes, they produce a severe inflammatory response, especially, when worms embed and migrate within the walls of the digestive tract and ruffled feathers, leading to an inability to maintain body balance (Atkinson *et al.*, 2009; Friend and Franson, 1999). Gapeworm infections are well known to occur in cranes, and they are also known for some fatal cases in chicks of other crane species (Carpenter and Derrickson, 1987; Gibson, 2007; Olsen and Langenberg, 1996; Spalding *et al.*, 1996). In the present case, because the esophagus was strangled, it was thought that the chick could not swallow food, and it became severely thin and prostrated. Additionally, dyspnea and pulmonary noise occurred, due to parasitic nematode pneumonia and air sac inflammation. However, it showed several symptoms, such as eyeball protrusion, an increased amount of eye mucus, a head swing, and neck tilt, from 1 week of age,

suggesting that there was another pathogenic infection, in addition to the nematode infestation.

The nematodes belonged to the family Syngamidae Ryzhikov, 1967 (Nematoda: Chromadorea: Rhabditida: Strongylida: Strongyloidea); specifically, three genera, *Syngamus* Siebold, 1836, *Boydinema* Baruš & Tenora, 1972 and *Cyathostoma* Blanchard, 1849, have been recorded to cause bird infections (Lichtenfels, 1980). *Hovorkonema* Turemuratov, 1963 (nec. Jurásek, 1977) is classified as one of the subgenera of the genus *Cyathostoma* and has been known to infect several avian orders, mainly, Galliformes, Falconiformes, Strigiformes, and Anseriformes, that live in the Palearctic Region (Atkinson *et al.*, 2009; Baruš *et al.*, 1978; Krone *et al.*, 2007). One of the species, *Cyathostoma (Hovorkonema) variegatum* (Creplin, 1849) Chapin, 1925, has been reported previously in many avian species, belonging to families of the Falconiformes, Strigiformes, Anseriformes, and Gruiformes, that live in

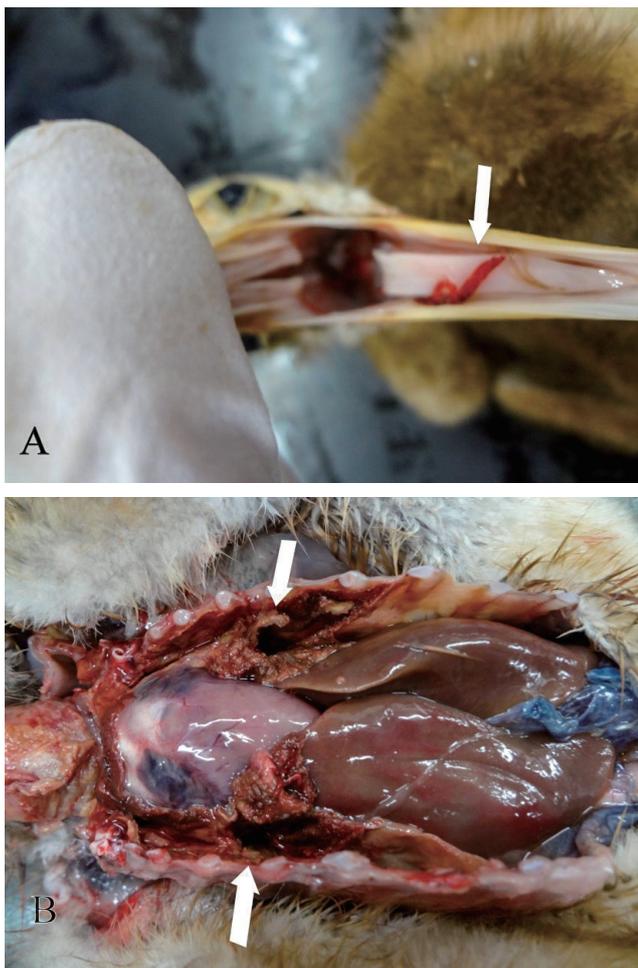


Fig. 1. Lesion identified at postmortem. (A) nematodes in the oral cavity (arrow), and (B) thickened air sacs (arrow).

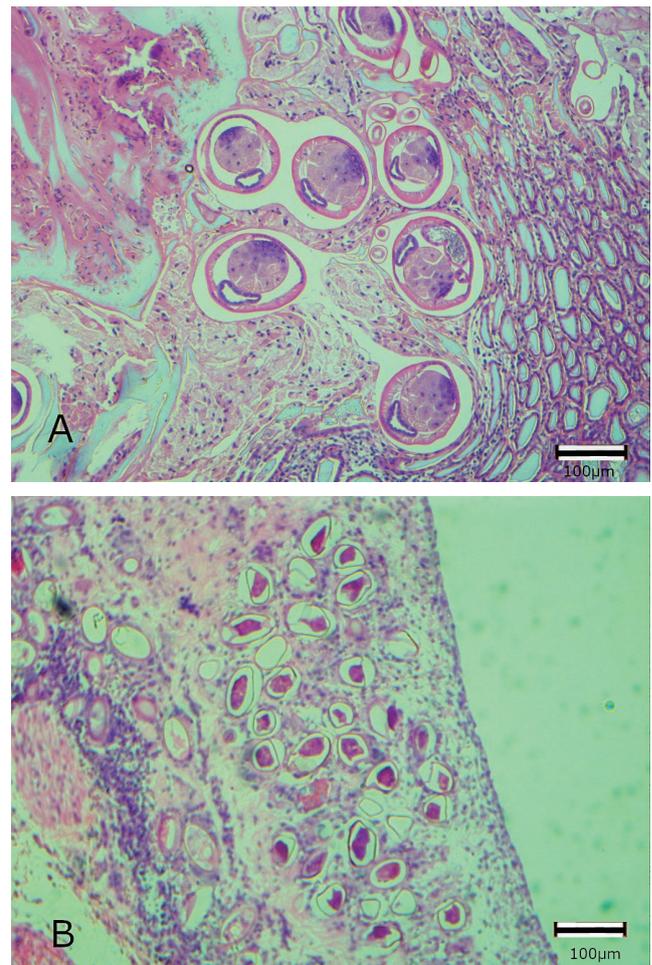


Fig. 2. Histopathological observation. (A) granulomatous pneumonia, with nematodes in the center, and (B) severe inflammation of the air sac, caused by nematode eggs.



Fig. 3. *Cyathostoma (Hovorkonema)* sp. obtained from the red-crowned crane. (A) Buccal capsule of male, (B) posterior extremity and spicules of the male.

the regions of Europe, Asia, America, and Australia (Atkinson *et al.*, 2009; Baruš *et al.*, 1978; Carpenter and Derrickson, 1987; Gibson, 2007; Krone *et al.*, 2007; Olsen and Langenberg, 1996; Spalding *et al.*, 1996). For the Gruidae, three crane species, including the common crane, *G. grus*, sandhill crane, *G. canadensis*, and whooping crane, *G. americana*, have been recorded as the hosts of *C. (H.) variegatum* (Carpenter and Derrickson, 1987; Gibson, 2007; Kanarek *et al.*, 2016; Krone *et al.*, 2007; Olsen and Langenberg, 1996; Spalding *et al.*, 1996); however, there are no records for infection by *Cyathostoma (Hovorkonema)* in *G. japonensis*. The present specimens were considered to be similar to *C. (H.) variegatum*, based on the morphological characteristics and measurements, but this could not be confirmed because the male specimens were immature.

Although four species of syngamid nematodes, namely, *Syngamus trachea* (Montagu, 1811) Siebold, 1836 (Host: Domestic chicken); *Syngamus* sp. (Host: Rhinoceros Auklet, *Cerorhinca monocerata*); *C. (Cyathostoma) lari* Blanchard, 1849 (Host: Grey Heron, *Ardea cinerea*, and Schrenk's Bittern, *Ixobrychus eurhythmus*); and *C. (C.) microspiculum* (Skrjabin, 1913) Turemuratov, 1963 (Host: Eurasian woodcock, *Scolopax rusticola*), have previously been reported from Japanese birds (Uchida *et al.*, 1991; Yokohata, 2002; Yoshino *et al.*, 2009), there has been no record of the subgenus *Hovorkonema* in wild or captive birds in Japan. For *G. japonensis*, a protozoan (*Eimeria* spp.), two trematodes (*Echinostoma gotoi* Ando and Ozaki, 1923 and *Apatemon*

Table 1. Body measurements for *C. (H.) variegatum* specimens collected from a red-crowned crane.

	Male		Female	
	Present specimens (n=2)	<i>C. (H.) variegatum</i> (Baruš <i>et al.</i> 1978)	Present specimens (n=10)	<i>C. (H.) variegatum</i> (Baruš <i>et al.</i> 1978)
Body length	3.75, 7.14	9.5 – 10.7	8.16 – 19.19	25 – 40
Maximum body width	0.18, 0.39	0.32 – 0.33	0.62 – 0.83	0.62 – 0.83
External diameter of buccal capsule	0.07, 0.13	0.145 – 0.150	0.24 – 0.36	0.41 – 0.56
Internal diameter of buccal capsule	0.05, 0.1	0.099*	0.18 – 0.27	0.352 – 0.384
Depth of buccal capsule	0.05, 0.11	0.109 – 0.132	0.19 – 0.27	0.304 – 0.368
Length of esophagus	0.43, 0.58	0.65 – 0.78	0.78 – 0.91	1.12 – 1.15
Maximum width of esophagus	0.08, 0.1	0.14 – 0.16	0.14 – 0.16	0.272 – 0.304
Maximum width of bursa	0.29, 0.32	0.512	–	–
Maximum depth of bursa	0.14, 0.17	0.208	–	–
Length of ventral rays	0.07, 0.09	0.140 – 0.165	–	–
Length of medio-lateral ray	0.10, 0.13	0.21 – 0.26	–	–
Length of externo-dorsal ray	0.13, 0.16	0.144 – 0.170	–	–
Length of medio-dorsal ray	0.20, 0.23	0.330 – 0.363	–	–
Spicules	0.46, 0.55	0.580 – 0.720	–	–
Egg size	–	–	0.071 – 0.079 x 0.037 – 0.044	0.073 – 0.083 x 0.052 – 0.056

Measurements are given as ranges (in mm).

* Variation is not given.

gracilis (Rudolphi, 1819) Szidat, 1928), and five nematodes (*Baruscapillaria* sp., *Contracecum* sp., *Paracuaria adunca* (Creplin, 1846) Anderson and Wong, 1981, *Syncuaria* sp., and *Viktorocara* sp.) species have previously been recorded in Japan (Oshima *et al.*, 2014; Sarashina *et al.*, 2006; Yoshino *et al.*, 2015). Therefore, the present case is the first record of *Cyathostoma* (*Hovorkonema*) sp. from *G. japonensis* worldwide and the first geographical record from Japan.

It is well known that the genus *Cyathostoma* has a direct life cycle but sometimes uses earthworms as a paratenic host. In *S. trachea*, a closely related species, the eggs hatch on the ground and grow to infective larvae about one or two weeks after hatching. Then, they infest their final hosts or earthworms by ingestion of the infective larvae (L3) and are transferred from the host's digestive tract to the respiratory system and mature after about three weeks of infection in their final hosts (Anderson, 2000). The chick showed remarkable symptoms, such as a lost voice and difficulty swallowing, since three weeks of age. Assuming that the growth of the present species is the same or close to that of *S. trachea*, it is thought that the chick was infected by nematode larvae soon after her birth. Additionally, red-crowned cranes live in wetlands, and they feed on several items, such as crayfishes, snails, fishes, earthworms, insects, and plants (Masatomi, 2000; Kobayashi *et al.*, 2002). In the present case, the parents brought many earthworms for their chick, so the chick's infection might have been the result of ingestion of a paratenic host (maybe earthworms) with infective larvae. Attention should be paid to the possibility that the nematode *Cyathostoma* (*Hovorkonema*) sp. causes epizootic infections among paratenic hosts (earthworms) and severe pathogenicity in final hosts, especially in zoos. Thus, a systematic epidemiological survey is required, not only for the healthcare of captive animals, but also for the conservation of wild cranes.

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タンチョウ *Grus japonensis* から得られた *Cyathostoma (Hovorkonema)* sp. (線虫綱: 開嘴虫科) の初記録

吉野智生・飯間裕子・松本文雄・浅川満彦 …………… 7

2013年8月6日に死亡したタンチョウのヒナを剖検したところ、鼻腔内と前胸および叉骨間気嚢から20個体の線虫が検出された。形態学的に検討した結果、円虫上科開嘴虫科の *Cyathostoma* 属 *Hovorkonema* 亜属の一種であった。種名決定は保留されたが、タンチョウを宿主とした

Cyathostoma 属線虫の報告は初であり、同時に、この属線虫の国内初となった。本属線虫はミミズを待機宿主として利用することが知られ、この個体は親からミミズを盛んに給餌されていたため、幼虫を含んだミミズを採食して感染したと考えられた。

キーワード：気嚢炎、食道絞扼、開嘴虫科線虫、肉芽腫性肺炎、待機宿主