山階鳥学誌 (J. Yamashina Inst. Ornithol.), **43**: 65–73, 2011

Short Note (短　報)

A Parasitological Survey on Introduced Birds in Japan

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Abstract. A parasitological survey was performed on 11 species of birds introduced into Japan: *Cygnus olor, Branta canadensis moffitti, Cairina moschata domestica, Phasianus colchicus karpowi, Bambusicola thoracica, Pavo cristatus, Psittacula krameri manillensis, Pycnonotus sinensis, Garrulax canorus, Leiothrix lutea, Lonchura malacca*. A total of 17 taxa of external and internal parasites, including one protozoan (*Eimeria* sp.), five nematodes (*Eucoleus perforans*, *Amidostomum anseris*, *Pseudaspidodera pavonis, Heterakis gallinarum* and *Synhimantus* (*Dispharynx*) *nasuta*), four trematodes (*Echinochasmus* sp., *Amphimerus anatis, Tanaisia* sp. and Dicrocoeliidae gen. sp.), one cestode (unidentified) and six arthropods (*Goniodes pavonis, Lipeurus maculosus, Ixodes turdus, Haemaphysalis flava, Leptotrombidium scutellare* and *Mouchetia* sp.) were obtained and identified. Among the obtained parasites, Dicrocoeliidae gen. sp., *I. turdus* and *L. scutellare* from *L. lutea,* and *Tanaisia* sp. and *L. scutellare* from *G. canorus,* are the first host records, while *P. pavonis* and *G. pavonis* are the first geographical records from Japan.

Key words: Introduced birds, Parasitic arthropods, Parasitic helminths.
キーワード: 外来鳥類, 寄生性節足動物, 寄生蠕虫類.

Recently, about 40 species of exotic and domestic birds have been introduced into Japan, and some of these have spread throughout the country (Japanese Society of Ecology 2002, Eguchi & Amano 2004). It has long been recognised that introduced birds can serve as reservoirs of pathogens which could then be transmitted to native birds (Long 1981, Asakawa *et al.* 2002, Japanese Society of Ecology 2002). For example, some viral, bacterial and protozoan diseases, including avian pox, parrot disease and avian malaria, have accompanied the invasion of introduced birds and reduced the number of native bird species in the Hawaiian Islands and Mascarenes Islands (Warner 1968, Pierce *et al.* 1977, van Riper *et al.* 1986, Dobson & May 1991, Simberloff 1992, Jones 1996), whereas in Europe, the roundworm *Heterakis gallinarum*, derived from introduced birds, has become prevalant (Tompkins *et al.* 1999, Prenter *et al.* 2004). A significant factor in the spread of parasites is the fact that these introduced birds are largely sympatric with native birds

Received 30 January 2011, Revised 28 May 2011, 2nd Revision 21 April 2011, Accepted 25 April 2011.

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and there is little compelling evidence for resource competition between introduced and native birds (Eguchi & Amano 2000, Japanese Society of Ecology 2002). Biological and ecological studies have been conducted on several introduced bird species in Japan, including studies of their habitats, feeding behavior, reproduction and resource competition (Eguchi & Masuda 1994, Amano & Eguchi 2002a, 2002b, Tanaka & Takahara 2003, Kawakami & Yamaguchi 2004, Nakamura 2004, Tojo & Nakamura 2004, Sato 2006, Kato 2009). However, there is little information regarding the diseases caused by helminth and arthropod parasites. A preliminary survey on parasitic helminths and arthropods in introduced birds carried out by Yoshino *et al.* (2003) examined only four specimens each of *Garrulax canorus* and *Leiothrix lutea*. To supplement these preliminary findings between 2003 and 2010 we carried out a detailed survey and obtained much new data.

We utilized three methods of parasitological survey; a helminthological survey on carcasses, a coprological survey, and a survey of external parasites and blood samples collected from captive birds.

A total of 46 carcasses of nine introduced bird species, namely *Cairina moschata domestica* (n=5), *Cygnus olor* (n=2), *Pavo cristatus* (n=6), *Phasianus colchicus karpowi* (n=8), *Bambusicola thoracica* (n=11), *Pycnonotus sinensis* (n=2), *G. canorus* (n=2), *L.*

Table 1. Introduced birds examined.

Order	Family	Species	Samples			Locality
			Carcasses	Feces	Blood Smears	
Anseriformes						
	Anatidae	*Cygnus olor*	2			Tomakomai & Toyako (Hokkaido)
		Branta canadensis moffitti		3		Yamakita (Kanagawa)
		Cairina moschata domestica	5		5	Iruma (Saitama)
Galliformes						
	Phasianidae	*Pavo cristatus*	6	15	4	Kohama & Ishigaki Islands (Okinawa)
		Bambusicola tholacica	11			Ichihara & Chiba (Chiba), Mizuho (Tokyo), Yokohama (Kanagawa), Iruma (Saitama)
		Phasianus colchicus karpowi	8			Ebetsu, Sapporo & Urakawa (Hokkaido)
Psittaciformes						
	Psittacidae	*Psittacula krameri manillensis*		1		Isehara (Kanagawa)
Passeriformes						
	Pycnonotidae	*Pycnonotus sinensis*	2			Tomigusuku (Okinawa)
	Timaliidae	*Garrulax canorus*	2	1	15	Hachioji (Tokyo), Fukuoka (Fukuoka)
		Leiothrix lutea	9		60	Hachioji (Tokyo)
	Estrildidae	*Lonchura malacca*	1			Tomigusuku (Okinawa)

lutea (n=9), and *Lonchura malacca* (n=1), were collected from several areas of Japan and examined (Table 1). Additionally, external parasites were examined for *G. canorus* (n=15) and *L. lutea* (n=60) that were captured by mist netting (mesh size 36 mm) conducted on five occassions between August 2002 and June 2004 at Tama Forest Science Garden, Tokyo. Parasitological examinations were performed at the Wild Animal Medical Center (WAMC) of Rakuno Gakuen University, Japan. All visceral organs from the carcasses were examined under a binocular microscope. The helminths and arthropods obtained were fixed and identified in 70% ethanol solution. To facilitate microscopic examination, nematodes were cleared in lacto-phenol solution, trematodes and cestodes were stained with heidenhain-iron hematoxylin solution, and arthropod parasites were mounted with Hoyer's medium. Morphological and biometric data were recorded using a camera lucida (OLYMPUS Model BH-2DA). All specimens are preserved in the WAMC or the Meguro Parasitological Museum, Tokyo, Japan.

Fecal samples of four introduced species (*P. cristatus* (n=15) collected in November 2003 at Kohama Island; *Psittacula krameri manillensis* (n=1) collected in June 2002 at Kanagawa Prefecture; *Branta canadensis moffitti* (n=3) collected in March 2010 at Lake Tanzawa, Kanagawa Prefecture; *G. canorus* (n=1) collected in June 2004 at Hachioji, Tokyo) were investigated by using a sugar flotation centrifugal technique (specific gravity =1.27). A fresh feces sample weighting was suspended in 60 ml water, and the suspension was filtered through a coarse strainer to remove any large particles. A 15 ml aliquot of the filtrate was transferred into two test tubes and centrifuged at 800 rpm for 10 min. The supernatant was discarded and the precipitate was re-suspended in a sugar solution and centrifuged at 1,500 rpm for 10 min. A cover slip was placed on the fluid surface of the solution in each test tube and allowed to stand for 20 min. Subsequently, the cover slip was removed, placed onto a glass slide and examined microscopically; based on the morphology of the numbers of eggs (EPG) and oocysts (OPG) per gram of feces were counted.

Blood smears were collected and investigated in captive *G. canorus* (n=15), *L. lutea* (n=60), *C. m. domestica* (n=5) and *P. cristatus* (n=4). Approximately 100 μl of blood was collected from each bird using an 18 gage needle. Thin blood smears were fixed in methanol solution, stained for 40 min with 3% Giemsa solution (pH 7.0), and scanned under an optical microscope.

A total of ten helminth and five arthropod parasite taxa were identified (Table 2). They include five nematode taxa (*Eucoleus perforans, Amidostomum anseris, Pseudaspidodera pavonis, H. gallinarum* and *Synhimantus* (*Dispharynx*) *nasuta*), four trematode taxa (*Amphimerus anatis*, *Tanaisia* sp., *Echinochasmus* sp. and Dicrocoeliidae gen. sp.), two biting louse taxa (*Goniodes pavonis*, *Lipeurus maculosus*), two ixodid ticks (*Ixodes turdus* and *Haemaphysalis flava*) and a feather mite (*Mouchetia* sp.). Although some segments of a cestode were recovered from *P. cristatus*, these specimens were heavily degenerated and we were unable to identify it definitively. No parasite species were recovered from *P. sinensis* and *L. malacca*, and no *Trichinella* larvae were observed in the muscle of any of the birds examined. Two arthropod parasites, including the ixodid tick (*H. flava*) from *G. canorus*, and the chigger mite *Leptotrombidium scutellare* from both *G.*

Table 2. Helminths and arthropods obtained from introduced birds.

Parasitic helminths	Site*	Host	Infected/ Examined	Prevalence	Intensity		
					Mean	SD	Range
Nematoda							
Enoplida							
Trichurioidea							
Capillariidae							
Eucoleus perforans	oe	*Pavo cristatus*	2/6	33.3	13.5	2.1	12–15
	oe	*Phasianus colchicus karpowi*	3/8	37.5	5.7	2.5	3–8
Strongylida							
Trichostrongyloidea							
Amidostomatidae							
Amidostomum anseris	gi	*Cygnus olor*	1/2	50.0	3	—	3
Ascaridida							
Heterakoidea							
Heterakidae							
Pseudaspidodera pavonis	ce	*P. cristatus*	5/6	83.3	174.8	236.3	1–443
Heterakis gallinarum	ce	*Bambusicola thoracica*	7/11	63.6	10.8	8.7	3–23
	si, ce	*P. c. karpowi*	1/8	12.5	194	—	194
Spirurida							
Acuarioidea							
Acuariidae							
Dispharynx nasuta	oe, pr	*P. cristatus*	2/6	33.3	63.5	16.3	52–75
Trematoda							
Echinostomatida							
Echinostomatidae							
Echinochasmus sp.	si	*P. cristatus*	1/6	16.7	3	—	3
Plagiorchiida							
Eucotylidae							
Tanaisia sp.	ki	*Garrulax canorus*	1/2	50.0	2	—	2
Ophitholchiida							
Opistholchidae							
Amphimerus anatis	li	*Cairina moschata domestica*	2/5	40.0	1	—	1
Dicrocoeliida							
Dicrocoeliidae							
Dicrocoeliidae gen. sp.	si	*Leiothrix lutea*	1/9	11.1	11	—	11
Cestoda							
Unidentified	si	*P. cristatus*	1/6	16.7	—	—	—
Insecta							
Phthiraptera							
Goniodidae							
Goniodes pavonis	bs	*P. cristatus*	3/6	50.0	7.7	7.4	2–16
Philopteridae							
Lipeurus maculosus	bs	*P. c. karpowi*	1/8	12.5	1	—	1
Archanida							
Acari							
Ixodidae							
Ixodes turdus	bs	*L. lutea*	1/9	11.1	1	—	1
Haemaphysalis flava	bs	*Garrulax canorus*	1/2	50.0	1	—	1
Avenzoariidae							
Mouchetia sp.	fe	*L. lutea*	8/9	88.9	5.6	4.4	1–14

* Abbreviation of site: oe, oesophagus; pr, proventriculus; gi, gizzard; si, small intestine; ce, caecum; li, liver; ki, kidney; bs, body surface; fe, feather

Table 3. Arthropods obtained from captive birds.

Arthropods	Site	Host	Infected/ Examined	Prevalence	Intensity		
					Mean	SD	Range
Archnida							
Acari							
Ixodidae							
Haemaphysalis flava	face	*Garrulax canorus*	2/15	13.3	1	0	1
Leptotrombididae							
Leptotrombidium scutellare	ear	*Leiothrix lutea*	1/60	1.7	5	—	5
	ear	*G. canorus*	1/15	6.7	6	—	6

Table 4. Results of coprological survey.

Species	Host	Infected/Examined	OPG or EPG		
			Mean	SD	Range
Eimeria sp.	*Pavo cristatus*	7/15	7.8	8.4	1.1–24.3
Nematode eggs	*P. cristatus*	5/15	2.9	1.7	0.7–3.5
	Branta canadensis moffitti	1/3	0.7	—	0.7
Cestode eggs	*Garrulax canorus*	1/1	1.7	—	1.7

canorus and *L. lutea*, were recovered from around the ear area of captive individuals (Table 3).

Coccidian oocysts of a single taxon, *Eimeria* sp. were recovered from *P. cristatus*, nematode eggs from *P. cristatus* and *B. c. moffitti,* and cestode eggs from *G. canorus*. Their prevalence and OPG/EPG are listed in Table 4. No coccidian oocysts and no parasite eggs or protozoan parasites were recovered from *P. k. manillensis*. Nematode eggs recovered from *B. c. moffitti* were considered as being a trichostrongylid species.

No blood parasites were evident in any of the samples examined.

For the introduced birds, *H. gallinarum* recovered from *P. c. karpowi* and *B. thoracica*, Schistosomatidae gen. sp., *Centrorhynchus turdi, Ornithoica bistativa, Ornithomya avicularia aobatonis* and *H. flava* recovered from *G. canorus*, and *Trypanosoma* sp., *Anonchotaenia* sp., *Ornithonyssus sylvialum* recovered from *L. lutea*, are parasite taxa previously reported in Japan (Uchida *et al.* 1991, Yoshino *et al.* 2003, Nagata 2006). In addition, one nematode species recovered in this study, *Pseudaspidodera* sp. from *P. cristatus,* had previously been reported from these birds kept in zoological gardens in Japan, (Sato *et al.* 2005). Among the parasites identified in the present survey, Dicrocoeliidae gen. sp., *I. turdus*, *L. scutellare* and *Mouchetia* sp. recovered from *L. lutea,* and *Tanaisia* sp. and *L. scutellare* recovered from *G. canorus*, are the first host records. *Pseudaspidodera pavonis* and *G. pavonis* are the first geographical records from Japan. Most biting lice and feather mite taxa live on the body of their host throughout their life cycles (Hopkins & Clay 1952, Gaud & Atyeo 1996). Some of them are known to move using other arthropods (e.g. louse flies), but this is not known for *Goniodes* spp. (Keirans 1975, Macchioni *et al.* 2005). It is also known that the heterakid nematode *P. pavonis can*

infect its definitive host directly (earthworms can be paratenic hosts) (Anderson 2000). In view of their respective life histories, these two species are likely to be alien parasites, introduced to Japan together with their hosts (Japanese Society of Ecology 2002, Asakawa 2005).

The nematode genera *Amidostomum* and *Heterakis* are well known as highly pathogenic agents for their hosts; the former causes heavily ventriculitis and ulcers, whereas the latter causes enteritis and the transmittable agents of *Histomonas meleagridis* that causes black-headed disease (Tuggle & Crites 1984, Friend & Franson 1999, Chalvet-Monfray *et al.* 2004, McDougald 2005). Moreover, by invading the intestinal wall of infected birds, larvae of *H. gallinarum* can cause typhlitis, with severe diarrhea, weight loss and depression, and lymphocytic infiltration and granuloma formation (Chalvet-Monfray *et al.* 2004, McDougald 2005); the epidemiological consequences of infection by *H. gallinarum* should be considered. It is known that nematodes belonging to the same family (or same genus) often have similar pathogencities. However, it is known that *Ascaridia dissimilis* (Heterakoidea: Ascaridiidae) could transmit *H. meleagridis* to their hosts, and that *H. bonasae* (Heterakoidea: Heterakidae) could not be the carrier of histomoniasis (Davidson *et al.* 1978, Norton *et al.* 1999). It remains unknown whether *Pseudaspidodera* spp. could be a carrier of *H. meleagridis*. Further investigation of the pathogencity of *P. pavonis* is required. Members of the genus *Eucoleus* are capillariid nematodes and cause severe esophagitis that has resulted in fatalities in wild and domestic galliform birds (Tateiwa 1960, Friend & Franson 1999).

The ixodid ticks, *I. turdus* and *H. flava*, are distributed throughout the Japanese Islands, China and Russia, and have been reported from many Japanese native birds (mainly passerine birds) (Yamauchi 2001). The chigger mite *L. scutellare* is known from many birds in Japan and is also distributed in China and Southeast Asia (Takada 1990, Nagahori 1998). Several arthropods, namely *H. flava*, *Ixodes* sp. and *Leptotrombidium* spp. were previously recorded from *G. canorus* and *L. lutea* (Tanskul & Gingrich 1986, Yoshino *et al.* 2003); these ticks and mites can act as be mediators of Lyme disease (Miyamoto *et al.* 2000) and human rickettsiosis (Takada 1990).

The coprological survey found few coccidian oocysts and parasite eggs in the feces of *B. c. moffiti*, but did recover *P. cristatus* and cestode eggs from the feces of *G. canorus*, and coccidian oocysts from the feces of *P. cristatus.* Eggs of trichostrongylid nematodes were recovered from *B. c. moffiti;* trichostrongylids are regarded as high pathogenic agents of their host birds, mainly anseriformes (e.g. *Amidostomum* spp.) (Friend & Franson 1999, Asakawa *et al.* 2002).

Although there exist several records of blood parasites, including the genera *Haemoproteus*, *Leucocytozoon*, *Plasmodium* and *Trypanosoma,* from *G. canorus* and *L. lutea* living in Southeast Asia (McClure *et al.* 1978), these parasites were not evident in the present survey.

Our study shows that birds introduced into Japan are parasitized by several external and internal parasites, including some highly pathogenic agents (e.g. *A. anseris*), and that some of the parasite fauna consists of parasites introduced together with them. However the impact of these introduced parasites on native birds and the ecosystem in Japan is still

unclear. A more detailed survey and monitoring of the parasites and infectious agents of introduced birds in Japan is warranted.

Acknowledgements

The present survey was supported in part by the Grant-in-Aid (Nos. 18510205, 20380163) and by the Support Project to Assist Private Universities in Developing Bases for Research from the Ministry of the Education, Science and Culture, Japan. We are grateful for the samples donated by Tama Forest Science Garden, Yamaha Resort Haimurubushi, Utonai Wildlife Rescue Center, Tokyo Agriculture University, Hamura Zoo, Tama Zoological Garden, Kanagawa Wildlife Support Network, Bird Clinic Kanesaka Animal Hospital, Prof. A. Uchida (Azabu University), Mr. K. Miyagi, and Mrs. Y. Miyagi. Thanks are also due to M. Fujita, Y. Iida, A. Ishikawa, N. Takahashi, J. Uemura, K. Aizawa, A. Miura, Y. Yabe and K. Ishii, for their assistance in sampling.

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日本における外来鳥類の寄生虫調査

日本には現在，愛玩・展示および家禽化を目的に毎年約200万羽の外国産鳥類が輸入され，外来種化したものも少なくない。外来鳥類が在来生態系に及ぼす影響の一つに感染症や寄生虫の持ち込みが知られるが，国内では調査が少なく，特に寄生虫の持ち込みやその影響は不明のままであった。そこで筆者らはコブハクチョウ *Cygnus olor*，オオカナダガン *Branta canadensis moffitti*，バリケン *Cairina moschata domestica*，コウライキジ *Phasianus colchicus karpowi*，コジュケイ *Bambusicola thoracica*，インドクジャク *Pavo cristatus*，ワカケホンセイインコ *Psittacula krameri manillensis*，シロガシラ *Pycnonotus sinensis*，ガビチョウ *Garrulax canorus*，ソウシチョウ *Leiothrix lutea* およびギンパラ *Lonchura malacca* の計11種について寄生蠕虫類，原虫類および節足動物の保有状況を調査した．その結果，*Eimeria* sp., *Eucoleus perforans*, *Amidostomum anseris*, *Pseudaspidodera pavonis, Heterakis gallinarum, Synhimantus* (*Dispharynx*) *nasuta*, *Echinochasmus* sp., *Amphimerus anatis, Tanaisia* sp., Dicrocoeliidae gen. sp.，属種不明条虫，*Goniodes pavonis, Lipeurus maculosus, Ixodes turdus, Haemaphysalis flava, Leptotrombidium scutellare* および *Mouchetia* sp. の計17種の寄生虫が確認され，その中には高病原性を示す線虫類の *P. pavonis, H. gallinarum* および *A. anseris* や，人獣共通感染症を媒介する *I. turdus* および *L. scutellare* が含まれていたため注目された．ソウシチョウの Dicrocoeliidae gen. sp., *I. turdus* および *L. scutellare* とガビチョウの *Tanaisia* sp. および *L. scutellare* は新宿主記録であり，*P. pavonis* および *G. pavonis* は国内初記録であった．血液原虫や消化管原虫類については殆ど検出されなかったが，今回検出された蠕虫類，節足動物も含め今後のモニタリングが必要であろう．

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