Anatoecus species (Phthiraptera: Philopteridae) from Anseriformes in North America and taxonomic status of *Anatoecus dentatus and Anatoecus icterodes*

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Abstract—*Anatoecus* Cummings (Phthiraptera: Philopteridae) is a genus of chewing lice with four species infesting Anseriformes in North America: *Anatoecus cygni* (Denny), *Anatoecus dentatus* (Scopoli), *Anatoecus icterodes* (Nitzsch), and *Anatoecus penicillatus* Kéler. Males of *A. dentatus* and *A. icterodes* are distinguishable by their genitalia; however, there are no known anatomical characteristics to distinguish females. *Anatoecus dentatus* and *A. icterodes* are recorded from at least 55 of the same host species worldwide. The mitochondrial gene cytochrome c oxidase subunit I was examined from the four *Anatoecus* species, specifically to examine the taxonomic status of *A. dentatus* and *A. icterodes*. When sequences from these species were analysed using neighbour joining analysis, *A. dentatus* and *A. icterodes* were recovered in a well-supported cluster. However, *A. dentatus* and *A. icterodes* were mixed within the same cluster. The average interspecific genetic distance of *A. dentatus* and *A. icterodes* (0.04%) was almost the same as the average intraspecific genetic distances of *A. dentatus* and *A. icterodes* 0.02% and 0.05%, respectively. Therefore, we formally synonymise *A. dentatus* and *A. icterodes* as *Anatoecus dentatus* (new synonymy). In addition two new hosts for *A. penicillatus* were recorded: *Branta canadensis* (Linnaeus) (Anseriformes: Anatidae) and *Chen caerulescens* (Linnaeus) (Anseriformes: Anatidae).

Résumé—Quatre espèces de mallophages du genre *Anatoecus* Cummings (Phthiraptera: Philopteridae) infestent les ansériformes d'Amérique du Nord: *Anatoecus cygni* (Denny), *Anatoecus dentatus* (Scopoli), *Anatoecus icterodes* (Nitzsch), et *Anatoecus penicillatus* Kéler. Les mâles A. *dentatus* et *A. icterodes* se distinguent par leurs organes génitaux, mais aucune caractéristique anatomique ne distingue les femelles. De plus, on rapporte dans le monde au moins 55 espèces d'ansériformes infestées à la fois par *A. dentatus* et *A. icterodes*. Nous avons examiné le gène mitochondrial cytochrome c oxydase des quatre espèces d'*Anatoecus* spp. dans le but d'étudier le statut taxonomique de *A. dentatus* et *A. icterodes*. Suite à l'analyse des séquences de ces espèces par la méthode NJ (*neighbour-joining*), *A. dentatus* et *A. icterodes* se sont retrouvés dans un clade monophylétique très fortement supporté. Toutefois, *A. dentatus* et *A. icterodes* étaient paraphylétiques l'un par rapport à l'autre. La distance interspécifique moyenne entre *A. dentatus* et *A. icterodes* (0.04%) était presque la même que les distances intraspécifiques moyennes entre *A. dentatus* et *A. icterodes*, soit respectivement 0.02% et 0.05%. Par conséquent, nous considérons formellement *A. dentatus* et *A. icterodes* comme étant *Anatoecus dentatus* (nouvelle synonymie). De plus, deux nouveaux hôtes ont été recensés pour *A. penicillatus*: *Branta canadensis* (Linnaeus) (Anseriformes: Anatidae) et *Chen caerulescens* (Linnaeus) (Anseriformes: Anatidae).

Introduction

Chewing lice (Insecta: Phthiraptera) are permanent ectoparasites found on birds and mammals (Marshall 1981). Since parasites can have detrimental effects on their hosts, it is important to understand their natural history. In order to accomplish this accurately, confident identifications of all members of the community under study must be achieved.

Species of *Anatoecus* Cummings (Phthiraptera: Philopteridae) infest ducks, geese, swans (Anseriformes), and flamingoes (Phoenicopteriformes)

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(Price *et al.* 2003). There are six species of *Anatoecus* recorded from Anseriformes worldwide; however, two of these species, *Anatoecus clayae* (Kéler) and *Anatoecus regina* Ansari, are only recorded from hosts found outside of North America. The four species found on hosts that have at least part of their distribution in North America are *Anatoecus cygni* (Denny), *Anatoecus dentatus* (Scopoli), *Anatoecus icterodes* (Nitzsch), and *Anatoecus penicillatus* Kéler.

The majority of *Anatoecus* species infest one to three closely related host species. For example, *A. penicillatus* infests mute swans (*Cygnus olor* (Gmelin)), and *A. cygni* infests tundra swans (*Cygnus columbianus* (Ord)), trumpeter swans (*Cygnus buccinator* Richardson), and whooper swans (*Cygnus cygnus* (Linnaeus)). *Anatoecus dentatus* and *A. icterodes* are found on at least 67 and 70 host species, respectively, and co-infest at least 55 of the same host species worldwide (Price *et al.* 2003). Co-infestations among species of lice from the same genus do occur; however, co-infestations usually only occur on one or a small number of host species (Price *et al.* 2003; Galloway 2004). *Anatoecus dentatus* and *A. icterodes* are extraordinary in the large number of Anseriformes host species they co-infest.

Females of all species of *Anatoecus* can be identified by the shape of the clypeal plate and characteristics of the terminal segments, except *A. dentatus* and *A. icterodes* (Kéler 1960). Interestingly, females of *A. dentatus* and *A. icterodes* are morphologically indistinguishable. However, males of all *Anatoecus* species are separable by characteristics of the genitalia (Fig. 1). Male *A. dentatus* have an effractor, which is an oval piece of dark-shiny chitin on the posterior margin of the endomeral plate, described by Cummings (1916) as resembling a "tin-opener without the handle" (Fig. 1A). Males of *A. dentatus* also have a conspicuous reticular comb on the hypoineral area (Cummings 1916) (Fig. 1A). *Anatoecus icterodes* lacks both the effractor and the reticular comb (compare Figs. 1B to 1A). The lack of distinction between females of *A. dentatus* and *A. icterodes*, coupled with both species co-infesting the head and neck of their host (Ash 1960; Strilchuck 1976) and being found together on at least 55 species of hosts, has raised questions about the validity of these two species (Emerson 1972).

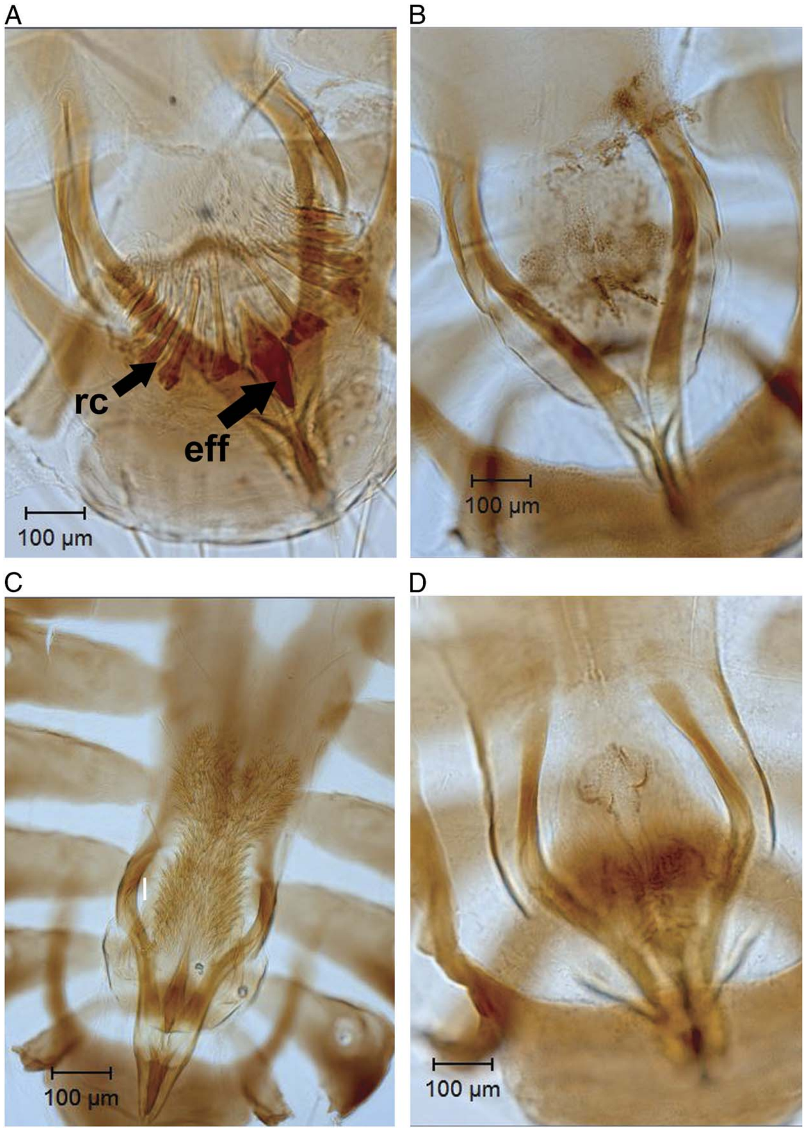
DNA barcoding using the mitochondrial gene cytochrome c oxidase subunit I (COI) has been developed to assign specimens to species as well as to identify cryptic species in insects (Hebert *et al.* 2003). For genetically well-defined lineages, specimens can be assigned to different species by using neighbour-joining analyses in which species form clustered groups on a tree and by the presence of a barcoding gap between species that forms when interspecific and intraspecific genetic distances are compared. The barcoding gap appears when interspecific genetic variation exceeds intraspecific genetic variation (Hebert *et al.* 2004; Barrett and Hebert 2005). A barcoding gap threshold of the interspecific genetic distance being at least 10 times greater than the distance within samples has been proposed; therefore, if the interspecific genetic divergence is below the threshold, this would support the taxonomic similarity among the samples and if the interspecific genetic divergence is above the threshold, the samples are distinct (Hebert *et al.* 2004). In the past, a 383 base pair (bp) portion of COI has been successfully used to differentiate species of chewing lice (Johnson *et al.* 2001; Clayton *et al.* 2006; Price *et al.* 2008; Price and Johnson 2009; Valim *et al.* 2011). Here we compare DNA sequence divergence of COI of *A. cygni*, *A. dentatus*, *A. icterodes*, and *A. penicillatus* to assess the taxonomic status of *A. icterodes* and *A. dentatus.*

Materials and methods

Lice were collected from salvaged birds collected in Manitoba, Canada, from 1994 to 2012. The majority of birds came from wildlife rehabilitation centres where they were either euthanised or died in their care. A few birds were provided by Manitoba Conservation and by hunters. All birds have been salvaged under a Wildlife Scientific Permit (WB12483) issued by Environment Canada, Canadian Wildlife Service.

Each bird was individually bagged as soon as possible after death and frozen at −20 °C for a minimum of 48 hours in order to kill all ectoparasites. Birds were allowed to thaw at room temperature until the wings and legs were movable. To remove ectoparasites, birds were washed twice with warm water and liquid soap and once with just warm water. The contents of each washing were poured through a 90 μm mesh

Fig. 1. The male genitalia of (A) *Anatoecus dentatus*, (B) *Anatoecus icterodes*, (C) *Anatoecus penicillatus*, (D) *Anatoecus cygni*. (eff, effractor; rc, reticular comb).



screen and rinsed into a plastic 400 mL storage container with either 70% or 95% ethanol to preserve the contents. Specimens were originally stored in 70% ethanol but were subsequently switched to 95% ethanol when the necessity of DNA analysis became apparent. Lice from each sample were sorted under a dissecting microscope.

In order to see the genitalia clearly, lice were cleared and mounted onto microscope slides. Mounting can be done after DNA extraction; however, this would have lead to hundreds of unnecessary extractions. Instead, half the louse was cleared and mounted onto a slide. If the louse was male the abdomen was mounted and if the louse was female the head was mounted; the other half of the louse was placed into an Eppendorf tube for DNA extraction. From this procedure, specimens were identified to species and selected for DNA extraction. All slides were prepared using the method described by Richards (1964) and vouchers were deposited in the J.B. Wallis – R.E. Roughley Museum of Entomology, University of Manitoba, Winnipeg, Manitoba, Canada.

DNA was extracted from the portion of the louse placed in the Eppendorf tube using the DNeasy™ Tissue Kit (Qiagen, Valencia, California, United States of America) following the manufacturer's protocol for animal tissue using spin columns. The DNA extract was used for PCR amplification of the COI gene using primers LCO1490 and HCO2198 (Folmer *et al.* 1994). Each PCR reaction mixture included 20.0–50.0 ng/μL of DNA template, 1X Buffer (0 mM Tris-HCl, 550 mM KCL, 1.5 mM $MgCl_2$, pH 8.3 at 25 °C) (New England Biolabs (NEB)), 0.2 mM dNTP (NEB), 4.0 μM MgSO4, 0.4 μM of each primer, one unit of *Taq* (NEB) and enough purified water to reach a final volume of 25 μL. Amplification was carried out using a MyCycler Thermal Cycler (BioRad – 170-9701EDU). The thermal regime consisted of an initial denaturation of one minute at 95 °C followed by 35 cycles of 15 seconds at 95 °C, 15 seconds at 46 °C, and 45 seconds at 72 °C, with a final extension of 4 minutes at 72 °C. Sequences were obtained using an ABI3730 sequencer from the University of Kentucky, Advanced Genetics Technology Center (refer to Table 2 for accession numbers).

Sequences were aligned by eye using BioEdit 7.2.0 (Hall 1999). As there were no indels, alignment was trivial and the ends were trimmed to minimise missing data across taxa. Genetic distances were calculated using Kimura's two-parameter model (K2P) (Kimura 1980) for base substitutions in MEGA v. 5.1 software (Kumar *et al.* 2004), following the barcode of life data system (BOLD) protocol (Ratnasingham and Hebert 2007). A neighbour-joining (NJ) analysis was performed based on these distances using MEGA software. MEGA was also used to perform bootstrap analysis for the NJ analysis (1000 replications) (Kumar *et al.* 2004). *Anaticola crassicornis* (Scopoli), a common chewing louse infesting ducks, was also used in the analysis and sequences for this species were obtained from GenBank (Accession numbers: NC015998.1 and DQ007339).

Results

In the 19 years that birds were examined for ectoparasites, 28 species of anseriform birds were examined and 17 of them were infested with *Anatoecus* species in Manitoba, Canada (Table 1). In all host species examined in which *A. dentatus* and *A. icterodes* were present, *A. icterodes* was more frequently observed than *A. dentatus* with an average of nine *A. icterodes* to one *A. dentatus*.

DNA was sequenced from 60 specimens of *Anatoecus,* which were collected from 18 hosts, representing seven species (Table 2). Hosts from which lice were acquired for sequencing were salvaged from 1999 to 2012; 10 were from Winnipeg, four were from within 360 km of Winnipeg, and four had no locality data, but all came from within the province of Manitoba, Canada.

The final alignment of COI was 519 bp in length of which 189 sites were variable, 189 were parsimony informative. The genetic distances of all specimens are presented in Table 3. The average interspecific genetic distance between *A. icterodes* and *A. dentatus* was 0.04%, which equates to <1 substitution per 519 bp. The next lowest interspecific genetic distance was 18.2% between *A. icterodes* and *A. cygni*. The average intraspecific genetic distances for *A. icterodes* and *A. dentatus* were 0.05% and 0.02%, respectively. These average intraspecific genetic distances are comparable to the other *Anatoecus* species, which range from 0% to 0.1%. The average interspecific genetic distance between *A. icterodes* and *A. dentatus* does not meet the barcoding gap threshold of being 10 times greater than the average intraspecific genetic distances. The average interspecific genetic differences between all other combinations of *Anatoecus* species exceed the 10 times barcoding gap threshold for the intraspecific genetic differences of those species and therefore support specific distinctness for *A. penicillatus* and *A. cygni.* All specimens of *A. icterodes* and *A. dentatus* were recovered in a well-supported cluster (bootstrap support = 100%) regardless of host (Fig. 2). However, *A. dentatus* and *A. icterodes* were mixed within the same cluster, which is not surprising given the extremely low interspecific distances between these species.

We have also observed two new host records for *A. penicillatus.* In 1995, *A. penicillatus* was first collected from a Canada goose (*Branta canadensis* (Linnaeus) (Anseriformes: Anatidae)). Since then, it has been recorded from 27 Canada geese. *Anatoecus penicillatus* was found on a snow goose

Table 1. Male *Anatoecus dentatus* and *Anatoecus icterodes* collected from Anseriformes birds from 1994 to 2012 in Manitoba, Canada.

Host species	Number of hosts examined	Number of hosts with only *A. dentatus*	Number of hosts with only *A. icterodes*	Number of hosts with *A. dentatus* and *A. icterodes*
Anatinae				
Anas crecca	9	0	2 (4)	1 (1;1)
Anas clypeata	3	0	0	1 (2; 31)
Anas discors	15	1 (1)	2 (3)	3 (4; 9)
Anas platyrhynchos	295	10 (13)	30 (149)	28 (104; 296)
Aix sponsa	38	0	6 (7)	4 (10; 15)
Branta bernicla	2	0	0	1 (1; 9)
Branta canadensis	293	5 (5)	87 (714)	19 (53; 691)
Branta hutchinsii	11	0	4 (67)	0
Chen caerulescens	20	0	5 (11)	3 (16; 114)
Chen rossii	3	0	1 (3)	0
Aythyinae				
Aythya affinis	4	0	2 (10)	0
Aythya americana	5	0	2 (6)	0
Aythya collaris	1	0	1 (1)	0
Aythya valisineria	14	0	2 (2)	2 (15; 71)
Merginae				
Bucephala clangula	13	1 (1)	0	1 (6; 11)
Lophodytes cucullatus	26	0	1 (1)	1 (2; 11)
Oxyurinae				
Oxyura jamaicensis	5	0	0	1 (2; 7)
Total	757	17 (20)	145 (978)	65 (216; 1266)

Note: The numbers in brackets represent total lice collected. Where both species were collected from the same bird, the first number in brackets is *A. dentatus* and the second is *A. icterodes.*

(*Chen caerulescens* (Linnaeus) (Anseriformes: Anatidae)) in 1998 and has been recorded from two additional snow geese. Specimens were sent to Ricardo Palma (Museum New Zealand Te Papa Tongarewa, Wellington, New Zealand) for identification and we compared our specimens to a syntype of *A. penicillatus,* which was borrowed from the K.C. Emerson Entomology Museum, Oklahoma State University, Stillwater, Oklahoma, United States of America. **Material Collected:** 113 ♂, 211♀, *ex* Canada geese from Winnipeg, Stony Mountain, Selkirk, Headingley, Hecla Island, Pine Falls, Oakbank, and East St. Paul, Manitoba, Canada; 18 ♂, 7♀, *ex* snow geese, from Winnipeg and Winnipeg Beach, Manitoba, Canada. Some vouchers of *A. penicillatus* (not specimens used for this study) were also deposited in the Canadian National Collection of Insects, Arachnids and Nematodes, Ottawa, Ontario, Canada.

Anatoecus penicillatus has been collected from hosts that were also infested with *A. dentatus* and *A. icterodes.* Of the 30 geese from which *A. penicillatus* was collected, 14 Canada geese also had *A. icterodes*, one Canada goose was infested with both *A. dentatus* and *A. icterodes*, and one snow goose was also infested with both *A. dentatus* and *A. icterodes.*

Discussion

There is good genetic evidence based on the lack of a barcoding gap (Table 3), that *A. dentatus* and *A. icterodes* are conspecific. The average interspecific genetic distance between *A. dentatus* and *A. icterodes* was 0.04%, considerably smaller than the next smallest interspecific distance of 18.2% between *A. icterodes* and *A. cygni.* In other studies on lice, the genetic divergences between species were >14.5% for *Myrsidea* Waterston species (Phthiraptera: Menoponidae) (Price and Johnson 2009) and >7.0% for *Dennyus* Neumann

Table 2. *Anatoecus* specimens chosen for sequencing with corresponding GenBank accession numbers and host from which they were collected from in Manitoba, Canada.

Host	Case number	Louse sp.	Sex	Voucher number	Accession number
Anas crecca	AGWT/144/CEN/11	*A. dentatus*	♂	954	KF754407
		A. icterodes	♂	955	KF754415
Anas discors	BWTE/380/PWRC/11	*A. dentatus*	♂	957	KF754416
				005	KF754376
Anas platyrhynchos	MALL/405/PWRC/11	*A. dentatus*	♂	006	KF754391
				010	KF754393
				011	KF754380
		A. icterodes	♂	001	KF754394
				002	KF754381
				003	KF754382
				004	KF754383
				009	KF754395
				012	KF754384
		A. dentatus or	♀	013	KF754385
		A. icterodes		015	KF754386
				016	KF754388
				017	KF754390
				018	KF754387
				019	KF754392
				020	KF754389
	MALL/1055/CEN/11	*A. dentatus*	♂	110	KF754378
				111	KF754376
		A. icterodes	♂	118	KF754409
				124	KF754410
	MALL/1543/CEN/11	*A. dentatus*	♂	367	KF754374
				375	KF754372
				376	KF754396
		A. icterodes	♂	369	KF754411
				370	KF754412
				373	KF754399
Aythya affinis	LESC/1/MC/11	*A. icterodes*	♂	951	KF754408
Branta canadensis	CAGO/1130/CEN/11	*A. dentatus*	♂	580	KF754413
				604	KF754402
				609	KF754375
				613	KF754400
		A. icterodes	♂	568	KF754406
				589	KF754414
	CAGO/1611/CEN/11	*A. dentatus*	♂	782	KF754398
				790	KF754373
		A. icterodes	♂	792	KF754404
				794	KF754403
				795	KF754405
	CAGO/41/PWRC/11	*A. icterodes*	♂	884	KF754401
		A. penicillatus	♂	882	KF754363
				883	KF754360
				885	KF754364
	CAGO/001/CEN/12	*A. icterodes*	♂	552	KF754379
	CAGO/1562/CEN/09	*A. penicillatus*	♂	913	KF754365
				914	KF754366
	CAGO/1557/CEN/09	*A. penicillatus*	♂	910	KF754358
				911	KF754359

Table 2. *Continued*

Host	Case number	Louse sp.	Sex	Voucher number	Accession number
	CAGO/381/PWRC/11	*A. penicillatus*	♂	880	KF754361
				881	KF754362
				879	KF754367
	CAGO/397/PWRC/11	*A. icterodes*	♂	246	KF754397
Bucephala clangula	COGO/1/MC/11	*A. icterodes*	♂	963	KF754417
Cygnus columbianus	TUSW/211/CEN/09	*A. cygni*	♂	895	KF754368
				905	KF754371
	TUSW/1/MC/10	*A. cygni*	♂	888	KF754370
				900	KF754369

Note: Case number: the first four letters are the bird species, the number indicates the hospital case number, the next letters represent the source from which the host came (CEN, Wildlife Haven; PWRC, Prairie Wildlife Rehabilitation Centre; MC, Manitoba Conservation) and the last two numbers represent the year the bird was collected.

Table 3. Average interspecific (plain text) and intraspecific genetic distances (in bold, standard error in brackets) under the K2P model for CO1 (519 bp).

	Anatoecus icterodes	*Anatoecus dentatus*	Females	*Anatoecus penicillatus*	*Anatoecus cygni*	*Anaticola crassicornis*
Anatoecus icterodes ($n = 22$)	**0.005 (0.0003)**	0.00018	0.00015	0.02085	0.01989	0.02542
Anatoecus dentatus ($n = 17$)	0.00036	**0.0002 (0.0002)**	0.00011	0.02104	0.01994	0.02548
Females ($n = 08$)*	0.00027	0.00011	**0.000 (0.000)**	0.02102	0.01988	0.02546
Anatoecus penicillatus ($n = 10$)	0.18363	0.18566	0.18585	**0.0008 (0.0007)**	0.02295	0.03104
Anatoecus cygni ($n = 04$)	0.18198	0.18335	0.18291	0.21289	**0.001 (0.001)**	0.02860
Anaticola crassicornis ($n = 02$)	0.26961	0.27134	0.27062	0.34107	0.33570	**0.000 (0.000)**

Notes: Standard errors are based on 1000 bootstrap replications. Data are based on 62 sequences from five species of lice collected from seven species of Anseriformes in Manitoba, Canada. Two sequences of *Anaticola crassicornis* were downloaded from GenBank (NC015998.1 and DQ007339).

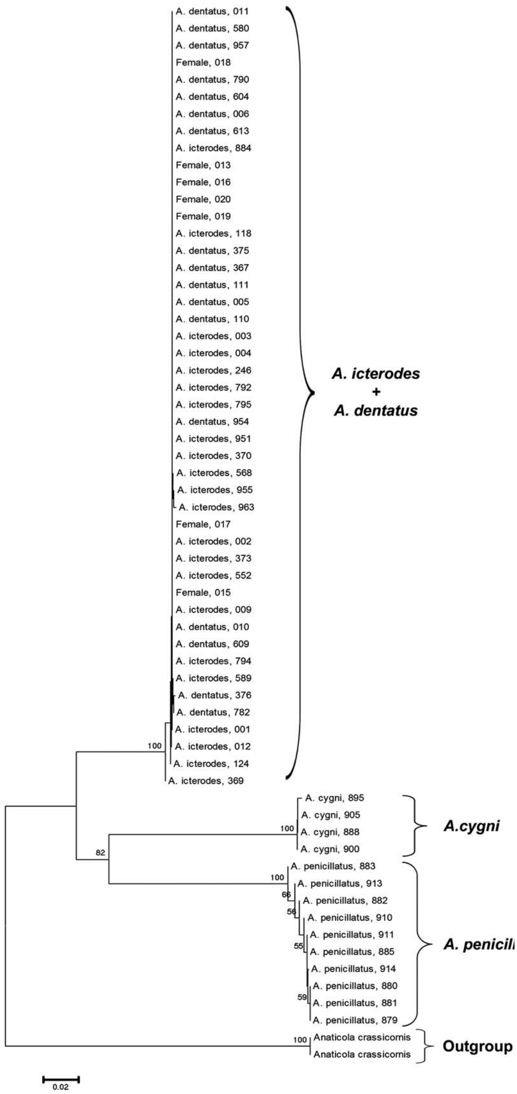
*Females are either *A. icterodes* or *A. dentatus* since they are indistinguishable. All other lice listed are males.

species (Phthiraptera: Menoponidae) (Clayton *et al.* 2006).

Hosts of *A. dentatus* and *A. icterodes* are found around the world; however, we have only sampled birds within Manitoba. Since these hosts are migratory in North America, and some, such as the Canada goose, may travel from Canada to northern Mexico to overwinter (Saunders and Saunders 1981), this allows different populations to interact. Although only birds collected from Manitoba were examined, because of their migratory behaviour, they should contain a reasonable representation of the population diversity of louse fauna that occurs over a wide area of North America. Therefore we formally propose the synonymy of *A. icterodes* and *A. dentatus*, as *Anatoecus dentatus* (Scopoli) (**new synonymy**).

The effractor and reticular comb are fascinating structures, although their function is unknown. *Anatoecus dentatus* without the effractor and reticular comb outnumbered those with the effractor and reticular comb nine times to one. In the course of this study, hundreds of male *A. dentatus* were mounted onto slides. All of the lice examined, with the exception of one, were clearly distinguishable as either expressing the effractor and reticular comb or not. In one louse from a brant goose (*Branta bernicla*

Fig. 2. Neighbour-joining tree under the K2P model based on cytochrome c oxidase subunit I (COI) with bootstrap values over 50% (1000 replications) indicated at the relevant nodes. Data based on 63 sequences from four *Anatoecus* species. Female specimens that are not distinguishable as *A. dentatus* or *A. icterodes* are labelled “Female”. After the louse species name is the voucher number.



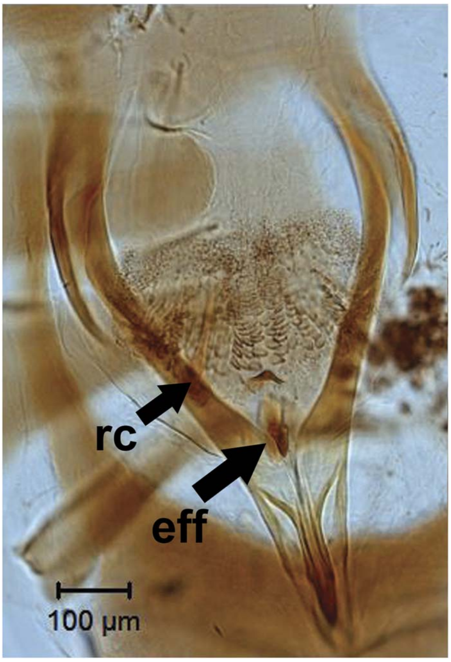
(Linnaeus) (Anseriformes: Anatidae)), the reticular comb was not completely developed (Fig. 3). However, an effractor was present so it was initially identified as *A. dentatus.* This was not the only male *Anatoecus* on this host; it was also infested with nine males that lacked an effractor and reticular comb.

The effractor and the reticular comb are always expressed together; however, it is not known what triggers the expression of these structures or how many genes are involved. Expression could also be based on environmental cues, or stress caused by resource availability or competition. Since the effractor and reticular comb are located in the same region as the male genitalia, they could possibly serve some function during mating or reproduction. If would be interesting to see if females are biased when offered a choice between a male with an effractor and reticular comb versus a male without.

Anatoecus penicillatus was previously recorded only from the mute swan (Price *et al.* 2003). Mute swans are native to Eurasia and were introduced to North America from the mid-1800s through the early 1900s as embellishments to parks and large estates as well as exhibits in zoos (Ciaranca *et al.* 1997). Since then, the mute swan has escaped captivity and the greatest concentration of them can be found along the Atlantic coast from Maine all the way to South Carolina, United States of America (Ciaranca *et al.* 1997). There are also populations of mute swans along the northern shores of Lake Erie and Lake Ontario as well as on Vancouver Island and the Fraser River delta of British Columbia, Canada (Cadman *et al.* 1987; Campbell *et al.* 1990). There are no established populations of mute swans in Manitoba (Parsons 2003) and none have come through the rehabilitation centres in Manitoba during the time this study was conducted. For this reason, it is unlikely that the specimens of *A. penicillatus* found on Canada geese and snow geese were the result of contamination in the rehabilitation centres.

Anatoecus penicillatus has invaded hosts that have a smaller body size than its native host. This is opposite to what Bush and Clayton (2006) found when they transferred *Columbicola columbae* (Linnaeus) (Phthiraptera: Philopteridae) and *Campanulotes compare* (Burmeister) (Phthiraptera: Philopteridae) from rock pigeons (*Columba livia* Gmelin (Columbiformes: Columbidae)) to smaller, novel Columbiformes. When birds were able to preen normally, introduced louse populations were reduced to near zero. *Columbicola columbae,* a wing louse, was not able to insert itself fully between the furrows in

Fig. 3. *Anatoecus dentatus* from a brant goose (*Branta bernicla*) with an incompletely formed reticular comb (eff, effractor; rc, reticular comb).



the barbs of the contour feathers. *Campanulotes compar*, a body louse, was not able to burrow into the downy feathers as successfully when on the smaller exotic hosts due to the smaller amount of downy feathers. Therefore, *C. columbae* and *C. compar* were more susceptible to preening by smaller, novel host birds. However, *A. penicillatus* is found on the head of its hosts (Grossi and Galloway, personal observation), where preening may be less efficient, therefore feather size may not be a restricting factor.

Over a period of seven years (2006–2012), *A. penicillatus* has been found regularly on Canada geese and snow geese. Therefore, it appears *A. penicillatus* has expanded its host range and has established populations on these novel hosts. Canada geese have also been introduced to Europe, where the mute swan is native. It would be very interesting to see if *A. penicillatus* has also made this transition to Canada geese in Europe.

The mute swan, in addition to being host to *A. penicillatus,* is host to *A. dentatus,* which is also recorded from Canada geese and snow geese (Price *et al.* 2003). In our study, *A. dentatus* and *A. penicillatus* were recorded co-infesting 15 Canada geese and have been observed together on the head and neck of their hosts (Grossi and Galloway, personal observation).

Finding two species of louse from the same genus recorded from the same host is not uncommon. For example, *Columbicola baculoides* (Paine) and *Columbicola macrourae* (Wilson) can be found together on mourning dove, *Zenaida macroura* (Linnaeus) (Columbiformes: Columbidae) (Galloway and Palma 2008). There is even one bird, the sora (*Porzana carolina* (Linnaeus) (Gruiformes: Rallidae)), which is host to two pairs of species of lice within the same genus (Galloway 2004): *Fulicoffula americana* Emerson and *Fulicoffula distincta* Emerson and *Rallicola mystax* (Giebel) and *Rallicola subporzanae* Emerson. The ability for two species of the same genus to co-exist on one host seems counter-intuitive to the competitive exclusion principle (Gause's law), where two species with the same resource requirements cannot occupy the same niche (Gause 1934; Hardin 1960). By this principle, either competition will drive one species to extinction or the species will evolve adaptations that allow them to exploit non-overlapping niches. The majority of sampling techniques only capture a snapshot of the louse population and do not monitor its change over time. It seems *A. penicillatus* and *A. dentatus* are able to co-exist, suggesting they inhabit at least, in part, non-overlapping niches.

To conclude, *A. icterodes* and *A. dentatus* are formally synonymised as *Anatoecus dentatus* (**new synonymy**), and Canada geese and snow geese are identified as new hosts for *A. penicillatus.* Nevertheless, we encourage others to replicate this study in other locations to further test this synonymy among additional populations of lice and former subspecies of *Anatoecus dentatus* (Price *et al.* 2003). We also encourage louse workers to examine anseriform birds to see if *A. penicillatus* has established populations on novel hosts elsewhere.

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