Some Parasites (Apicomplexa, Trematoda, Nematoda, Acanthocephala, Phthiraptera) of the Common Great Horned Owl, *Bubo virginianus virginianus* (Aves: Strigiformes: Strigidae), from Southeastern Oklahoma

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Abstract: A common great horned owl, *Bubo virginianus virginianus* was found dead on the road near Idabel, McCurtain County, Oklahoma; it was salvaged and examined for parasites. The following parasites were found: a coccidian, *Eimeria megabubonis*, a trematode, *Neodiplostomulum* sp., ova of a nematode, *Capillaria* sp., an acanthocephalan, *Centrorhynchus spinosus*, and a chewing louse, *Colpocephalum brachysomum*. All parasites are reported from Oklahoma for the first time. More importantly, it shows that utilizing select road-killed specimens that could otherwise not be collected alive because of state and federal regulations is worthwhile for parasitological examination, and can yield new scientific information.

Introduction

The common great horned owl, *Bubo virginianus virginianus* (Gmelin) is one of the largest strigiform birds and possesses an extremely large range, being the most widely distributed owl in the Americas (Lynch 2007). It ranges from the tree limit in Alaska and two-thirds of Canada to Tierra del Fuego, Chile (Peterson and Peterson 2002). In Oklahoma, according to Sutton (1967), it is a largely non-migratory owl, but some shifting of populations probably takes place in winter. This owl has been reported to feed on a great diversity of prey in Oklahoma, including insects, crayfish, snakes, a lizard, and 16 bird and 30 mammal species (Kittridge et al. 2006).

Although there are several reports on the coccidian (Cawthorn and Stockdale 198; Upton et al. 1990) and helminth parasites of *B. virginianus* (Ramaligam and Samuel 1978; Nickol 1983; Richardson and Nickol 1995; Kinsella et al. 2001; Bolette 2007; Richardon and Kinsella 2010), no parasites, to our knowledge, have been reported from this owl in Oklahoma. Here, we document several parasites from a salvaged *B. v. virginianus* from southeastern

Oklahoma and document new distributional records for its parasites.

Methods

An adult *B. v. virginianus* was found dead on 12 September 2017 off the road at the intersection of US 259 and Farley Road in Idabel, McCurtain County (33° 54' 36.3672"N, 94° 46' 48.6408"W) and brought to the laboratory for parasitic examination; it was not examined for subcutaneous helminths or *Trichinella* sp. The specimen appeared to be recently killed and the body showed no sign of petrification. The feathers were brushed for ectoparasites and those found were placed in a vial of 70% (v/v) ethanol; specimens were cleared in 10% potassium hydroxide, dehydrated through an ethanol series, further cleared in xylene, and slide-mounted in Canada balsam (Price et al. 2003). A midventral incision was made to expose the viscera and a blood sample was taken from the heart, smeared onto a microscopic slide and allowed to dry, then fixed in absolute methanol, stained in Giemsa, and rinsed in neutral buffered phosphate saline. The gastrointestinal (GI) tract from the throat to cloaca was removed, rinsed in 0.9% saline, and placed in a Petri dish. Feces from the rectum was collected and placed in a vial containing 2.5% (w/v) potassium dichromate ($K_2Cr_2O_7$) and, after flotation in Sheather's sugar solution (sp. gr. 1.30), examined for coccidia by brightfield microscopy. The sample contained unsporulated oocysts which were allowed to complete sporulation in a Petri dish containing a shallow layer of 2.5% $K_2Cr_2O_7$ for five days at room temperature (23°C). Sporulated oocysts were again isolated by flotation (as above) and measured using a calibrated ocular micrometer. Measurements on 10 oocysts/sporocysts are reported in micrometers (µm) with the means followed by ranges in parentheses; photographs were taken using brightfield optics. Oocysts were 14 days old when measured and photographed. Several 100 mm sections of the GI tract were cut, split lengthwise, and examined under a stereomicroscope for endoparasites. Trematodes were rinsed in saline, fixed in hot tap water without coverslip pressure, preserved in 70% ethanol, stained in acetocarmine, cleared in methyl salicylate, and coverslip mounted in kleermount. Nematode ova from the fecal flotation was placed on a microscopic slide, coverslip mounted, and photographed as above. Acanthocephalans were prepared, examined, and measurements (provided in µm) were made as described by Richardson (2005, 2006).

A photovoucher host was deposited in the Henderson State University (HSU) collection, Arkadelphia, Arkansas. Voucher specimens of ectoparasites were deposited in the General Ectoparasite Collection in the Department of Biology at Georgia Southern University, Statesboro, Georgia under accession no. L-3809. The coccidian (photovoucher), trematodes, and nematodes (photovoucher) were deposited in the Harold W. Manter Laboratory (HWML) of Parasitology, University of Nebraska, Lincoln, Nebraska. Voucher specimens of acanthocephalans were deposited in the Sam Houston State University Invertebrate Collection (SHSU), Sam Houston State University, Huntsville, Texas, and assigned accession numbers (SHSUINVRT000240 - SHSUINVRT000247).

Results and Discussion

The owl was infected or infested with several parasites, including a coccidian, trematode, nematode ova, an acanthocephalan, and a chewing louse; the blood smear was negative. The parasites recovered are presented below in annotated format.

Apicomplexa: Eimeriidae

***Eimeria megabubonis* Upton, Campbell, Weigel, and McKown, 1990.**–Oocysts (Figs. 1A–C, HWML 139369) matching the description of *E. megabubonis* were found to be passing in feces of *B. virginianus*. Measurements are as follows: subspheroidal oocysts, 28.6 × 25.0 (26–30 × 23–26), L/W ratio = 1.2 (1.0–1.3), bilayered (smooth outer) wall 1.2 (1.0–1.5), micropyle and oocyst residuum absent, 1–4 polar granules present; ellipsoidal-elongate sporocysts, 15.7 × 8.7 (14–17 × 8–10), L/W = 1.8 (1.5–2.1), Stieda and substieda bodies present, parastieda body absent, large sporocyst residuum present; each

sporozoite with three residual bodies.

This eimerian was originally described by Upton et al. (1990) from a single *B. virginianus* from Kansas. Another eimerian, the considerably smaller *E. bubonis* Cawthorn and Stockdale, has been reported from *B. virginianus* from Saskatchewan, Canada (Cawthorn and Stockdale 1981). We document a new distributional record for *E. megabubonis* as well as only the second time in over 25 years that this coccidian has been reported from a great horned owl.

Trematoda: Digenea: Neodiplostomidae

***Neodiplostomum* sp.–**Three specimens (HWML 139370) of an unknown *Neodiplostomum* (Fig. 1D) were found in the small intestine of this host. These digeneans possessed vitelline follicles restricted to the forebody (Fig. 1D) but lacked a genital cone, both morphological features of the Neodiplostominae (Dronen et al. 1995). Five species of *Neodiplostomum* have been previously reported from *B. virginianus*, including *N. cochleare* (Krause) La Rue, from Minnesota (Penrod 1947) and Wisconsin (Chandler and Rausch 1947), *N. americanum* Chandler and Rausch from Connecticut (Richardon and Kinsella 2010), Florida and Mississippi (Kinsella et al. 2001; Woodyard et al. 2017), *N. delicatum* Chandler and Rausch from Wisconsin (Chandler and Rausch 1947), *N. reflexum* Chandler and Rausch from Florida (Kinsella et al. 2001), Michigan (Chandler and Rausch 1947), and Brazil (Gallas and Silveira 2013), and *N. buteonis* Dubois and Rausch from owls from Ontario, Canada (Pearson 1960). Species of *Neodiplostomum* parasitize avian definitive hosts with the majority in the orders Falconiformes

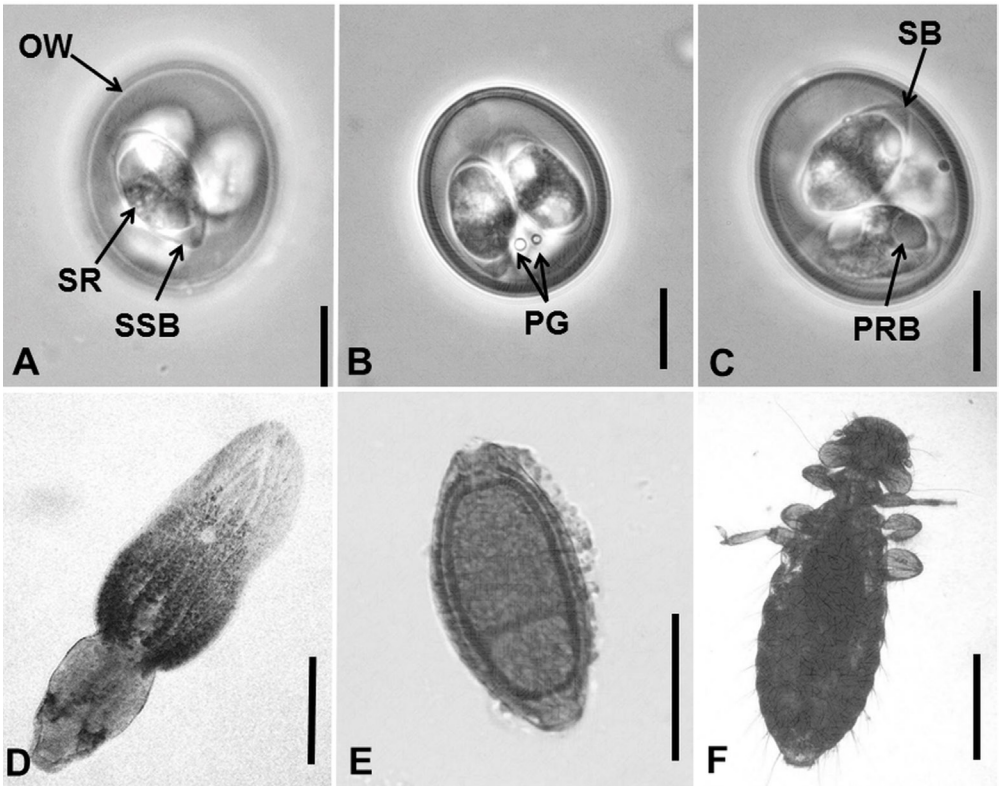

Figure 1. Parasites collected from *Bubo virginianus* from Oklahoma. A–C. Brightfield photomicrographs of sporulated oocysts of *Eimeria megabubonis*. Scale bars = 10 µm. D. *Neodiplostomum* sp. Scale bar = 25 µm. E. Ova of *Capillaria* sp. Scale bar = 25 µm. F. *Colpocephalum brachysomum* female. Scale bar = 500 µm. Abbreviations: OW (oocyst wall), PG (polar granule), PRB (posterior refractile body), SB (Stieda body), SSB (substieda body), SR (sporocyst residuum).

or Strigiformes but the orders Ciconiiformes, Coarciiformes, Cuculiformes, Passeriformes, and Piciformes are also represented (Dubois 1938, 1944, 1962). This is the first time this parasite has been reported from Oklahoma.

Nematoda: Trichurida: Capillariidae

***Capillaria* sp.**–Ova of a *Capillaria* sp. (Fig. 1E, HWML 139369) were recovered from the feces of *B. v. virginianus*. Two capillarid species have previously been reported from great horned owls, including *C. falconis* (Rudolphi) from Florida and Alberta, Canada, and *C. tennissima* (Rudolphi) from Florida (Ramalingam and Samuel 1978; Kinsella et al. 2001) and Connecticut (Richardson and Kinsella 2010). *Capillaria* spp. have also been previously reported from other vertebrates of Oklahoma (Williams 1953; Morrison and Gier 1979; Criffield et al. 2009); however, this is the first time ova of *Capillaria* sp. has been reported from an owl from the state.

Acanthocephala: Paleoacanthocephala: Polymorphida: Centrorhynchidae

***Centrorhynchus spinosus* (Kaiser, 1893) Van Cleave, 1924.**–Ten specimens (3 males, 7 females) of *C. spinosus* were collected from the small intestine representing a new geographic distribution record for this parasite. In accordance with Nickol (1983), the specimens were identified as *C. spinosus* based primarily on trunk shape, the most important character distinguishing individuals of this species from the morphologically similar congener, *Centrorhynchus kuntzi* Nickol, 1983. All specimens lacked the characteristic anterior inflation of the trunk associated with *C. kuntzi*.

Centrorhynchus spinosus was originally described as *Echinorhynchus spinosus* by Kaiser (1893) from an unknown host in Florida. Van Cleave (1916) described *C. spinosus* from a great blue heron, *Ardea herodias* (as *Herodias egretta*) from an undetermined locality. Van Cleave (1924) examined specimens from the Leidy collections supposedly collected from a great gray owl (*Strix nebulosa*) from Florida. Van Cleave (1924) concluded that the specimens were conspecific with *C. spinosus*, ascribed them as *C. spinosus* (Kaiser, 1893) and listed *C. spinosus* Van Cleave, 1916 as a junior synonym, thus establishing the current correct taxonomic designation *C. spinosus* (Kaiser, 1893) Van Cleave, 1916. The known range of *S. nebulosa* lies far north of Florida (Johnsgard 1988). Thus, the host of the Leidy specimens listed as being from *S. nebulosa* should be considered to be from an "unknown host," although it is likely that it was some kind of owl. Van Cleave (1940) reported acanthocephalans collected from an undetermined "Galapagos hawk" collected from the Galapagos Islands that were morphologically similar to *C. spinosus* but they possessed a longer proboscis and exhibited an anterior inflation of the trunk. Subsequently, Schmidt and Neiland (1966) described *C. kuntzi* from a roadside hawk, *Rupornis magnirostris* (as *Buteo magnirostris*), collected in El Recreo, Rio Escondido, Nicaragua. *Centrorhynchus kuntzi* was not reported from North America until Nickol (1983) reported *C. kuntzi* from three species of hawks in southern Louisiana. Nickol (1983) observed the fact that Van Cleave did not recognize the Galapagos specimens as distinct from *C. spinosus,* and may have permitted the occurrence of *C. kuntzi* to go unnoticed in North America. Nickol (1983) reexamined all available specimens of *C. kuntzi* in an attempt to correct previous misidentifications.

Centrorhynchus spinosus has been reported only rather sporadically throughout North America with the majority of reports being from owls in Florida and Louisiana, *B. virginianus*, barred owls (*Strix varia*), and eastern screech owls (*Megascops asio*) (Nickol 1983; Kinsella et al. 2001). Kinsella et al. (2001) reported prevalences of 78, 82, and 22% for *C. spinosus* from barred owls, great horned owls, and eastern screech owls, respectively, with intensities as high as 340 individual worms. Likewise, Nickol (1983) found six of eight (75%) barred owls to be infected with *C. spinosus*. Aside from owls, small numbers of specimens identified as *C. spinosus* have been reported from a potpourri of other hosts. Nickol (1969) reported five specimens of *C. spinosus* from three of 53 (6%) red-bellied woodpeckers, *Melanerpes carolinus* (as *Centurus carolinus*) from southern

Louisiana. A single specimen conforming to the description of *C. spinosus* was collected from a Cooper's hawk (*Accipiter cooperi*) in Connecticut (Nickol, 1983). Taft et al. (1993) reported two of 16 (13%) broad-winged hawks (*Buteo platypterus*) to be infected with *C. spinosus,* although these specimens have not been subsequently confirmed. Coulson et al. (2010) reported one salvaged nestling swallow-tailed kite (*Elanoides forficatus*) from southern Mississippi or Louisiana to be infected with *C. spinosus*, although voucher specimens were not referenced. Single immature individuals of *C. spinosus* were reported from an opossum (*Didelphis virginiana*) and raccoon (*Procyon lotor*) in Georgia (Ellis et al. 1999; Richardson 2014). Interestingly, Read (1950) demonstrated that *C. spinosus* did mature in an experimentally infected rat (*Rattus*), which had been fed cystacanths taken from the body cavity of an eastern garter snake (*Thamnophis sirtalis*) collected in Texas. Additionally, Vogel and Bundy (1987) indicated that cystacanths of *C. spinosus* were common in Jamaican gray anoles (*Anolis lineatopus*). The relatively high prevalence and intensity of *C. spinosus* among owls in Florida and Louisiana supports the assertion of Richardson and Nickol (1995) that *C. spinosus* is primarily a parasite of raptors of the order Strigiformes (owls).

Morphological analysis of the collected specimens revealed some apparent ambiguities, suggesting that the morphological distinctions previously used to distinguish between *C. spinosus* and *C. kuntzi* may not be as pronounced as previously thought. Nickol (1983) found that the number of large rooted hooks on the anterior proboscis differed between *C. spinosus* and *C. kuntzi*. Nickol (1983) also reported that *C. spinosus* bears eight to 11 large rooted hooks whereas *C. kuntzi* bears seven to nine, with the usual numbers of large rooted hooks being nine and 10 in females and males of *C. spinosus*, respectively, and eight in both sexes for *C. kuntzi*. The three males collected in this study that provided good hook counts exhibited nine to 10 (usually nine), large rooted hooks and the four females that provided reliable hook counts exhibited seven to nine large rooted hooks. Nickol (1983) further indicated that the proboscis of *C. kuntzi* is longer than that of *C. spinosus*, in contrast to the report of Schmidt and Neiland (1966) who stated that *C. spinosus* possesses a longer proboscis. The proboscides of female *C. spinosus* were frequently less than one mm long and averaged only 1,014 in length and that the proboscides of males averaged only 963 long according to Nickol (1983). In the current study, the proboscides of three males measured 1,050, 1,110, and 1,130 long × 300, 330, and 300 wide, respectively. The proboscides of three females measured 1,220, 1,250, and 1,250 long × 450, 440, and 410 wide, respectively. Nickol (1983) also indicated and figured a difference in proboscis shape between the two species with *C. kuntzi* exhibiting a pronounced swelling at the level of insertion of the proboscis receptacle that is lacking in *C. spinosus*. One specimen in the current study exhibited a pronounced swelling of the proboscis at the level of the insertion of the proboscis receptacle, similar to that figured by Nickol (1983) for *C. kuntzi*. The other proboscides either exhibited a proboscis shape like that figured by Nickol (1983) for *C. kuntzi*, or a slight swelling that was intermediate between the two species.

Centrorhynchus kuntzi has been reported from hawks throughout North America (Richardson and Kinsella 2010), as well as two *H. leucocephalus* from Florida (Kinsella et al. 1998), a groove-billed ani (*Crotophaga sulcirostris*) in Veracruz, México (Richardson et al. 2010), and in low numbers from great horned owls in Florida (Nickol 1983; Kinsella et al. 2001).

Although there appears to be some ambiguity in morphological characters previously thought to be unambiguous between *C. kuntzi* and *C. spinosus*, trunk shape, along with host affiliation, clearly support the validity of the view that *C. kuntzi* and *C. spinosus* are distinct species. Nevertheless, given the ambiguities revealed by the present specimens, further analysis, including molecular characterization of both species, is warranted as new material becomes available.

Arthropoda: Insecta: Phthiraptera: Menoponidae

***Colpocephalum brachysomum* Kellogg and Chapman, 1902.–**One male, 17 females, and one nymph of *C. brachysomum* (Fig. 1F) were found on the road-killed *B. virginianus*. Emerson (1940) did not include this species in his list of chewing lice recorded from Oklahoma. Peters (1936) reported three species of chewing lice from *B. virginianus* in the eastern United States, namely, *Strigiphilus oculatus* (Rudow) (listed under the synonym *Eustrigiphilus bubonis* Osborn), *Strigiphilus cursor* (Burmeister) (listed under the synonym *Philopterus cursor* Burmeister), which typically parasitizes the short-eared owl, *Asio flammeus*, and *Kurodaia subpachygaster* (Piaget), which typically parasitizes the barn owl, *Tyto alba.* Emerson (1972) listed the following four species of chewing lice as ectoparasites of *B. virginianus* in North America: *C. brachysomum, Kurodaia magna* Emerson, *Strigiphilus syrnii* (Packard) (listed under the synonym *Strigiphilus acutifrons* Emerson) and *S. oculatus*. Price et al. (2003) listed six species of chewing lice as true ectoparasites of *B. virginianus* namely *C. brachysomum, K. magna, S. oculatus, S. syrnii, Strigiphilus elutus* Carricker, and *Strigiphilus chilensis* Carricker, with the last two listed species apparently being restricted to the neotropical part of the range of *B. virginianus.*

Peters (1936) also recorded the ectoparasitic louse fly *Lynchia americana* Leach from *B. virginianus* in six eastern states, and Boyd (1951) reported a single great horned owl with more than 100 louse flies (which were not identified to species).

In conclusion, we document several new distributional records for parasites of *B. virginianus*. This survey, albeit based on a single host specimen, illustrates the significance of salvaging road-killed owls which can yield knowledge on their parasites that could not be obtained otherwise because of state and federal restrictions on collecting and euthanizing live birds, all in the spirit of conservation.

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