LOW INCIDENCE OF HELMINTH PARASITES IN BROWN-HEADED COWBIRDS (*MOLOTHRUS ATER*) FROM EAST TEXAS

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The brown-headed cowbird (*Molothrus ater*) is a common species of brood parasite in the continental US and Canada that reproduces by laying its eggs within the nests of other bird species (Lowther 2020). Because they are ground feeders, gregarious, obligate brood parasites, cowbirds have the potential to be exposed to a wide variety of diseases and parasites throughout their life cycle (Cooper & Crites 1976; Reisin & Hahn 2007). Surprisingly few studies have examined the incidence of helminths, i.e., parasitic worms, in cowbirds. Cooper et al. (1973) surveyed the helminths of brown-headed cowbirds from a cowbird control program in two counties in Ohio; each during a one year period. They found 45% of the 166 individuals were infected with helminths. Since Cooper et al. (1973) there have been no other published reports on the prevalence of helminths in brown-headed cowbirds to our knowledge. However, studies of another icterid with similarities to cowbirds in foraging and diet, the common grackle (*Quiscalus quiscula*), have found 100% of individuals infected with helminths (Welker 1962; Johnson 1984). The purpose of our study was to determine the incidence of helminth parasites in brown-headed cowbirds and the relationship between parasitic infection and body condition.

We examined 26 specimens of cowbirds (nine females and 17 males). We obtained cowbird specimens in April of 2021 and 2022 from a private landowner participating in a cowbird control program as part of a state of Texas Wildlife Exemption in Gilmer, TX. The birds were placed in plastic bags, frozen and transported to the biology department at Sam Houston State University (salvage permit MB126180-0), where they were stored in a standard freezer (2021) or an ultracold freezer (2022).

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To necropsy the birds, each individual was defrosted in a plastic bag that was placed in room temperature water. The mass of each bird was determined using an Ohaus portable electronic scale and the length of the right tarsus was measured in mm with dial calipers. We calculated body condition as body mass/tarsus length. Values are presented as mean with standard error. The 17 male birds had an average body mass of 42.7 ± 0.70 g, and the nine females averaged 34.3 ± 1.49 g. Body condition of males was 1.5 ± 0.04 and for females it was 1.2 ± 0.06.

Each bird was necropsied by using dissecting shears to cut from the cloaca, through the pectorialis muscle and keel. The organs were individually removed using fine-tipped forceps and placed in glass dishes covered with tap water to keep them moist. The liver and lungs were macerated first. The entire intestine was split lengthwise and gently scraped along its inner wall to dislodge any attached helminths before being placed into a glass dish with tap water for the contents to settle. After any particles in the water had settled, the contents of the dishes were examined under a dissecting microscope. All nematodes found were fixed in glacial acetic acid and permanently stored in 7% glycerol for future study. Nematode diagnosis was verified through original taxonomic descriptions.

Nineteen cowbird specimens were collected in 2021, and we examined an additional seven birds from 2022. Of the 26 cowbird specimens necropsied, only three individuals (11.5%), were infected. The three cowbirds were all males from 2021 and they appeared to be infected with the same nematode parasite. We identified the parasites as belonging to the genus *Tetrameres*, a blood nematode, and the seven *Tetrameres* sp. we observed were embedded in the proventriculi of the male brown-headed cowbirds. All the *Tetrameres* sp. found were females, which tend to be the larger and more readily observed sex (Raggi & Baker 1957). Although we predicted that cowbirds would be infected with several parasitic helminth species, we found no sign of endoparasites except *Tetrameres* sp. in these specimens. Furthermore, there was no significant difference between the body condition of the three parasitized males and the non-infected males (Mann Whitney U

test, U = 33, P = 0.13) but the low number of parasitized birds makes it hard to draw firm conclusions from this result.

Cooper et al. (1973) found that 45% of the brown-headed cowbirds they examined from Ohio were infected with helminths compared to our 11.5% of individuals infected. We questioned if the storage process we used in 2021 led to deterioration of the parasites over time and could account for the low numbers of helminth parasites we observed. The cowbird specimens we used in 2021 were stored in a standard freezer for five to seven months before they were necropsied. Thus in 2022, we obtained cowbird specimens that were freshly euthanized, transported on ice and frozen immediately in an -40° C ultracold freezer. We necropsied them within one to four weeks and found no parasites in the seven specimens we examined. The lack of parasites observed in the 2022 cowbird specimens infers that specimen storage methods in 2021 did not contribute to the low incidence of parasites detected.

Our findings align with those of Mena et al. (2020) who found that shiny cowbirds (*Molothrus bonariensis*), another generalist brood parasite, also have a low incidence of helminth infection (10% of 20 individuals sampled) although more parasitic species were found in each individual. Due to their brood parasitic nature, cowbirds are exposed to many potential pathogens and parasites while interacting with host species. Selection has favored heightened immunity in cowbirds toward viruses (Reisen & Hahn 2007) which may also account for the lower incidence of parasitism we reported, but this requires further investigation. Another likely explanation for the low helminth numbers in our study may be a result of the time of year the birds were collected. If birds are feeding mostly on seeds or grain in the winter, they may have less opportunity to become infected with parasites than later in the season when they eat more insects. Cooper et al. (1973) collected cowbirds throughout the year whereas our specimens were collected only in spring. There is evidence for seasonal effects on infection rate from other icterid species, with lower infection occurring in wintering and premigratory birds (e.g., red-winged blackbird, *Agelaius phoneniceus;* Hood & Welch 1980). Further study of helminth infections in cowbirds are warranted to determine how

seasonality and geography may impact parasite infections and whether infection influences body condition.

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