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## ***Strongyloides robustus* and the Northern Sympatric Populations of Northern (*Glaucomys sabrinus*) and Southern (*G. volans*) Flying Squirrels**

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**ABSTRACT:** Within North America, northern (*Glaucomys sabrinus*) and southern (*Glaucomys volans*) flying squirrels occupy distinct ranges with limited overlap. Sympatry in northern latitudes coincides with northern hardwood vegetation from Minnesota to New England. *Strongyloides robustus* is an intestinal parasite that infects both species but appears to be deleterious only to northern flying squirrels. As a result, *S. robustus* could be a critical determinant of flying squirrel population characteristics in at least some areas of sympatry. However, cold weather could potentially limit the distribution of *S. robustus* in northern climates. Therefore, we assessed fecal samples from both flying squirrel species to determine the presence of the nematode in Wisconsin. *Strongyloides robustus* was found in 12 flying squirrel scat samples and infected 52% of southern flying squirrels and 11% of northern flying squirrels. Prevalence of *S. robustus* infection for northern flying squirrels was substantially lower than previously reported from more southern regions. This is the northernmost documentation of *S. robustus* in flying squirrels and the first documentation of *S. robustus* parasitizing flying squirrels in Wisconsin.

**Key words:** Flying squirrels, *Glaucomys sabrinus*, *Glaucomys volans*, sciuridae, *Strongyloides robustus*, Strongyloididae, sympatry, Wisconsin.

Two flying squirrel species are recognized in North America: the northern, *Glaucomys sabrinus*, and southern, *Glaucomys volans* (Dolan and Carter, 1977; Wells-Gosling and Heaney, 1984). As their common names imply, the two species have distinct ranges with only limited overlap. They are ecologically similar and are irregularly sympatric in the southern Appalachians and throughout a relatively narrow band of northern hardwood vegetation from Minnesota eastward through south-

ern Canada and New England (Muul, 1968; Weigl, 1978). Weigl et al. (1999) suggested that regions of overlap supporting roughly equal numbers of both species have low and fluctuating flying squirrel populations (Weigl et al., 1999), although the source of population oscillations is unknown.

In overlap regions, northern and southern species seem to remain segregated within microenvironments (Weigl, 1978). Greater aggression of southern flying squirrels, differential adaptation to altitude and climate, vegetation preferences, diet availability, and differential susceptibility to parasitic diseases have been proposed to explain species segregation (Muul, 1968; Weigl, 1978).

*Strongyloides robustus* is an intestinal nematode that infects both species, but infection is more prevalent in the southern species (Wetzel and Weigl, 1994). Eggs are produced within the hosts' gastrointestinal tract, are shed in feces, and develop into larvae, which, after two molts, transform into an infective form (Anderson, 1992). *Strongyloides robustus* is pathogenic to most sciurids and can cause severe hemorrhagic enteritis (Davidson, 1976).

*Strongyloides robustus* might limit northern flying squirrels in areas of overlap because northern flying squirrels represent a susceptible "naïve" host compared with southern flying squirrels (Weigl, 1968; Weigl et al., 1999). This postulate was based on observations that when both species were infected with *S. robustus*, northern flying squirrels were detrimentally af-

ected by infection and experienced high rates of mortality, whereas southern flying squirrels appeared unaffected by infection and even produced several litters (Weigl, 1968). Although *S. robustus* is a significant cause of mortality in captive northern flying squirrels, there is little information on the influence this parasite might have on the ecology of flying squirrels and its role in mediating competition between the two species in regions of sympatry.

Rausch and Tiner (1948) documented *S. robustus* in three species of tree squirrels in the Upper Great Lakes region: red squirrels (*Tamiasciurus hudsonicus*), gray squirrels (*Sciurus carolinensis*), and fox squirrels (*Sciurus niger*). However, they did not detect *S. robustus* in either species of flying squirrel. Previous studies in more southern regions have shown that harsh winters markedly decrease the probability of *S. robustus* survival (Wetzel and Weigl, 1994). Therefore, it was conceivable that at higher latitudes, *S. robustus* might not even exist in flying squirrels. The goal of this study was to assess the presence, species distribution, and intensity of *S. robustus* in both flying squirrel species inhabiting the 83-ha Schmeckle Reserve located in Portage County, Wisconsin, USA (89°34'3"W, 44°32'25"N).

The reserve lies in a vegetative tension zone separating the southern prairie-forest region from the northern hardwood region (Curtis and McIntosh, 1951). Primary overstory vegetation includes red maple (*Acer rubrum*), red oak (*Quercus rubra*), paper birch (*Betula papyrifera*), quaking aspen (*Populus tremuloides*), hop-hornbeam (*Caprinus caroliniana*), white pine (*Pinus strobus*), jack pine (*Pinus banksiana*), and red pine (*Pinus resinosa*).

Fecal samples from free-ranging flying squirrels were collected from sympatric populations located in Schmeckle Reserve. Three sets of flying squirrel scat samples were used: seventeen scat samples were obtained in 1994 (seven northern and 10 southern flying squirrels of unknown sex), five samples were collected in

2000 (two northern, one each from a male and female, and three southern flying squirrels, all male), and eight samples were collected in 2002 (eight southern flying squirrels, one female and seven males). Irrespective of the year, all scat samples were collected from flying squirrels live-trapped between 1 September and 30 November and preserved in 10% formalin.

Three scat pellets were randomly selected from each sample and prepared with the use of a modified formalin-ethyl acetate sedimentation technique (Ritchie, 1948; Pritchard and Kruse, 1982). Scat pellets were broken apart and strained through double-layered wide-mesh cheesecloth with 10% formalin. The cheesecloth containing large unstrained material was discarded. Fecal material suspended in solution was centrifuged at  $1,500 \times G$  for 1 min. The sediment was resuspended in 10 ml of fresh formalin. Ethyl acetate (3 ml) was added to the suspension and shaken for 1 min. The solution was recentrifuged at  $1,380 \times G$  for 1 min, and three drops of iodine were added to the sediment. This was placed on a slide and examined by light microscopy for eggs and larvae of *S. robustus*, identified by size and morphologic criteria (Chandler, 1942; Ecklerlin, 1974; Wetzel, 1992; Bartlett, 1995); eggs in each sample were counted. Prevalences of infection for northern and southern flying squirrels were compared by Fisher's exact test (Zar, 1996). Pellets that were not microscopically screened for parasites were dried and weighed to estimate the mean mass of flying squirrel fecal pellets.

*Strongyloides robustus* was present in 40% of flying squirrel scat samples (Table 1). Prevalence was significantly higher in southern flying squirrels than in northern flying squirrels ( $P=0.049$ , Fisher exact test). Numbers of eggs per fecal pellet for flying squirrels was highly variable (Table 1).

Prevalence of southern flying squirrels infected with *S. robustus* varied from 24% to 100% in more southern portions of their range (Patrick, 1991; Pung et al., 2000). Weigl et al. (1999) reported 80% preva-

TABLE 1. Prevalence and intensity of *Strongyloides robustus* eggs/fecal pellet from nine northern (*Glaucomys sabrinus*) and 21 southern (*Glaucomys volans*) flying squirrels inhabiting central Wisconsin.

| Host                      | Prevalence (%) | Intensity       |                 |                 |
|---------------------------|----------------|-----------------|-----------------|-----------------|
|                           |                | $\bar{x}$       | SD              | Range           |
| <i>Glaucomys sabrinus</i> | 11             | 16 <sup>a</sup> | NA <sup>a</sup> | NA <sup>a</sup> |
| <i>G. volans</i>          | 52             | 14              | 13.5            | 1–20            |
| Total                     | 40             | 14              | 12.9            | 1–20            |

<sup>a</sup> Mean, standard deviation, and range were incalculable because  $n=1$ .

lence in northern flying squirrels in North Carolina and Tennessee (USA). Our estimated prevalence of *S. robustus* infection for southern flying squirrels is similar to prevalence of infection documented at lower latitudes. Prevalence of infection in northern flying squirrels in central Wisconsin appears to be substantially lower than reported elsewhere (Weigl et al., 1999).

Previous research has shown that *S. robustus* egg counts are highly and significantly correlated with the number of adult *S. robustus* parasitizing flying squirrels (Wetzel, 1992). Thus, fecal egg counts for *S. robustus* provide at least a relative index of worm abundance in both species of flying squirrels. In North Carolina and Tennessee, egg counts for southern flying squirrels exceeded 1,000 eggs/g, and approximately 60% of infected northern flying squirrels had intensities exceeding 500 eggs/g (Weigl et al., 1999). Given a mean mass for one flying squirrel fecal pellet of 0.10 g (SD=0.024,  $n=22$ ) and given that each pellet had an average of 14 eggs, our reported intensities appeared to be considerably lower than those reported by Weigl et al. (1999).

*Strongyloides robustus* is susceptible to cold temperatures (Wetzel and Weigl, 1994), and its abundance might be reduced in northern flying squirrels in the Upper Great Lake region because of decreased survival from exposure to severe and prolonged winters. Survival of *S. robustus* outside of its host in cold climates presents a special challenge (Wetzel and Weigl, 1994). Both species of flying squir-

rels aggregate during the winter, but the southern flying squirrel forms large aggregations of up to 50 individuals (Merritt et al., 2001). Although these aggregations might both decrease thermoregulatory costs and serve a social function (Merritt et al., 2001), such aggregations could also create a more suitable thermal environment for *S. robustus* survival and dispersal. Furthermore, flying squirrels often line nest sites with various kinds of organic material. Moist organic material could provide enhanced microhabitat for parasites and increase *S. robustus* transmission. In contrast, although not optimal for winter survival, northern flying squirrels will nest in open-faced leaf nests known as drays (Jackson, 1961). In sympatric areas, where nest site competition with southern flying squirrels is high, winter use of drays by northern flying squirrels might be particularly prevalent (Jackson, 1961). Therefore, the lower *S. robustus* infection frequency in northern flying squirrels reported herein might result from *S. robustus* being exposed to severe winter weather because of the tendency for northern flying squirrels to use drays.

Weigl et al. (1999) suggested that *S. robustus* and the southern flying squirrel have had a longer association than *S. robustus* and the northern flying squirrel. Although unproven, this is reasonable because the life cycle of *S. robustus* seems particularly suited to the natural history of southern flying squirrels, which, unlike the northern flying squirrels, occur at lower latitudes and altitudes, strongly prefer tree cavities for nest sites, and defecate within nesting cav-

ities (Wetzel and Weigl, 1994). Feces deposited in nest cavities create an ideal microenvironment for *Strongyloides* eggs to develop and infect southern flying squirrels. Infection of northern flying squirrels appears to occur when they take over abandoned or secondary southern flying squirrel nests (Wetzel and Weigl, 1994).

This is the first documentation of *S. robustus* in flying squirrels from Wisconsin and the northernmost documentation of *S. robustus* infecting either species of flying squirrels. Because infection of northern flying squirrels by *S. robustus* possibly is more deleterious than in southern flying squirrels, it could play a role in variability of flying squirrel populations and in segregation of northern and southern flying squirrels in the northernmost portions of their overlapping range.

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