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HEMOGRAMS AND HEMATOZOA OF SHARP-SHINNED (*ACCIPITER STRIATUS*) AND COOPER'S HAWKS (*ACCIPITER COOPERII*) CAPTURED DURING SPRING MIGRATION IN NORTHERN NEW YORK

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ABSTRACT: Hemograms were determined for 26 Cooper's (*Accipiter cooperii*) and 55 sharp-shinned hawks (*Accipiter striatus*) captured during spring migration (27 March to 12 May 1987) on the south shore of Lake Ontario, New York (USA). No significant differences were noted in packed cell volume and estimated total solids between the species. However, Cooper's hawks had significantly higher total white blood cell counts and higher concentrations of heterophils, monocytes, and eosinophils. Proportionally, lymphocytes made up a smaller percentage of the differential count in the Cooper's hawk. Eosinophil concentrations and percentages of the differential count were significantly higher in the females of both species. Both species had a high prevalence of *Leucocytozoon toddi* and *Haemoproteus* spp. infection. *Haemoproteus nisi* and *H. elani* were identified in both hawks. *Trypanosoma avium* was identified in a single Cooper's hawk and *Plasmodium circumflexum* was identified in a sharp-shinned hawk. Prevalence of *Leucocytozoon toddi* and *Haemoproteus* spp. infections were significantly higher in the birds caught late in the spring as compared to those caught earlier in the spring; this was evidence for a spring recrudescence of patent parasite infections.

Key words: Cooper's hawk, *Accipiter cooperii*, sharp-shinned hawk, *Accipiter striatus*, hemograms, hematozoa.

INTRODUCTION

Hematology is a valuable tool for the evaluation of blood loss (Palmer et al., 1978), disease status (Reiser and Temple, 1981), and responsiveness to therapy of sick and injured birds of prey (Campbell, 1988). For hematology to be maximally beneficial, however, it is necessary to know the expected hemograms for the species being evaluated. Sick and injured sharp-shinned (*Accipiter striatus*) and Cooper's hawks (*Accipiter cooperii*) make up a significant percentage of raptors presented to wildlife care facilities in North America (Redig, 1981). However, little published data on the hemograms of these species exist (Smith and Bush, 1978; Hunter and Powers, 1980).

Blood parasites are a common finding in blood smears obtained from wild birds (Wood and Herman, 1943; Barnard and Bair, 1986). Detailed information exists on the species of hematozoa that infect the Cooper's and sharp-shinned hawks (Kirkpatrick and Lauer, 1985; Peirce et al., 1990). Also, the prevalence of patent hematozoan infections in the Cooper's and sharp-shinned hawks has been reported (Kirkpatrick and Lauer, 1985). However, most birds sampled to date were trapped in fall migration. As seasonal fluctuations in parasite activity occur in many species (Alverson and Noblet, 1977), the prevalence of parasitemia and concentration of blood parasites might be expected to differ in birds sampled during the spring.

To establish hematological reference ranges and to further define the prevalence of hematozoan infections in these two avian species, hemograms were determined for 26 Cooper's and 55 sharp-shinned hawks captured for banding during spring migration.

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MATERIALS AND METHODS

Birds sampled in this survey were trapped for banding during spring migration at Braddock Bay, along the south shore of Lake Ontario, New York (USA) (43°18'N, 77°42'W) between 25 March and 12 May 1987. Age was assessed by plumage (Clark and Wheeler, 1987). Birds hatched in 1986 were classified as juveniles. Adult birds were defined as those birds hatched in 1985 or earlier. Blood was collected from the medial metatarsal vein for each bird and stored in two 100 μl heparinized capillary tubes. Blood smears were made from blood remaining in the needle hub and air dried. Birds were bled within 10 min of capture. Before release, birds, were weighed and examined for signs of disease.

Packed cell volume (PCV) was determined by centrifugation of one sealed capillary tube. A refractometer (Reichert Scientific Industries, Buffalo, New York) was used to estimate the total solids of the plasma layer. Blood in the second tube was mixed by repeated inversion and the combined heterophil and eosinophil count was obtained by diluting the blood with a phloxine stain (Eosinophil Unopette, Becton-Dickinson, Rutherford, New Jersey, USA) and counting the number of stained cells within the nine large squares in each of the two chambers in a hemocytometer (Reichert Scientific Industries). The concentration of combined heterophils and eosinophils per microliter was calculated by adding 10% to the stained cell count and multiplying this value by 16. Blood smears were stained with Diff Quik® (American Scientific Products, McGraw Park, Illinois, USA). The white blood cell composition for each bird was estimated by classifying 200 leucocytes for each bird's blood smear. White blood cells (WBC) which could not be clearly identified were included in the differential count as unidentified cells. Corrected WBC counts were obtained by dividing the absolute combined heterophil and eosinophil count obtained from the hemacytometer by the combined heterophil and eosinophil fraction determined by the differential count.

Insufficient blood was obtained from six birds for both PCV determination and WBC counts. In these cases, only one of these values was determined, resulting in variable sample sizes for each hematological parameter.

Leucocytozoon toddi and *Trypanosoma* spp. were detected by scanning 50 fields of each blood smear with a 40× objective. *Haemoproteus* spp. or *Plasmodium* spp. were detected by screening 10,000 red blood cells with a 100× objective. When hematozoa were present, their concentration per microliter of blood was estimated by determining the number of organisms present per 100 WBC's. If less than one parasite was found per 100 WBC's, the bird was considered infected, but the concentration of organisms was not calculated. Parasites were identified by the taxonomic keys of Bennett et al. (1993, 1994).

Hematological parameters were compared between male and female hawks, and adult and immature hawks, within each species, and between overall values of the two species using the Wilcoxon rank sum test (Rosner, 1986). Differences were considered significant at $P < 0.05$. The possibility of a spring recrudescence in patent hematozoan infections was evaluated by comparing the prevalence of infection and concentration of blood parasites in birds caught early in the spring (25 March to 24 April 1987) to those caught in the later portion of the migration (25 April to 12 May 1987) using the chi-square test of independence (Ott, 1988). This division was chosen so that there would be an equal number of birds in each group. Prevalences of *Haemoproteus* spp. and *Leucocytozoon* spp. infection in the sharp-shinned hawks reported here also were compared to those reported by Kirkpatrick and Lauer (1985) using the chi-square test of independence.

Representative blood smears were deposited in the International Reference Center for Avian Hematozoa, Harold Manter Laboratory of Parasitology, University of Nebraska State Museum, Lincoln, Nebraska (USA) (accession numbers 121221 to 121297).

RESULTS

Twenty-six Cooper's hawks were bled: five adult females, six immature females, six adult males, and nine immature males. Fifty-five sharp-shinned hawks were bled: 16 adult females, 18 immature females, five adult males, and 16 immature males. Feather quality on all birds was judged to be good to excellent and no birds had evidence of pectoral muscle atrophy, malnutrition, or other disease. There was a cutaneous lesion, interpreted to be a healing laceration, on the neck of one sharp-shinned hawk and a similar healing wound on the leg of one Cooper's hawk.

Cooper's hawks had significantly ($P < 0.05$) higher total white blood cell counts and higher concentrations of circulating heterophils, monocytes, and eosinophils than sharp-shinned hawks (Table 1). Lymphocytes made up the greatest percentage

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TABLE 1. Absolute and differential white blood cell (WBC) counts of Cooper's and sharp-shinned hawks, in northern New York, 1987.

Species	Evaluation	Total WBC count	Heterophils	Lymphocytes	Monocytes	Eosinophils
Cooper's hawks	Absolute cell counts (n = 21)[a]	11,904 (3,024–32,727)[b]	4,795 (574–12,436)[b]	5,595 (1,542–16,036)	995 (272–3,693)[b]	1,344 (351–4,255)[b]
	Differential cell counts (n = 21)[c]	NA[d]	38% (19–59%)[b]	42% (20–61%)[b]	9% (3–19%)[b]	12% (2–25%)[b]
Sharp-shinned hawks	Absolute cell counts (n = 48)[a]	6,594 (2,647–17,331)	1,875 (283–5,544)	4,019 (2,771–13,443)	502 (0–1,604)	391 (55–1,733)
	Differential cell counts (n = 51)[c]	NA[d]	28% (7–63%)	55% (32–81%)	6% (0–12%)	6% (2–13%)

[a] Median number of cells per μl (range).
[b] Values differed significantly ($P < 0.05$) from those of sharp-shinned hawks.
[c] Median percentages of each cell followed by the range.
[d] Not applicable.

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TABLE 2. Absolute and differential eosinophils counts for female and male Cooper's and sharp-shinned hawks, northern New York, 1987.

		Females	Males
Cooper's hawk	Absolute cell counts[a]	1,855 (n = 10)[b] 955–2,976	1,164 (n = 11) 350–4,254
	Differential cell counts[c]	14% (n = 10)[b] 9–25%	9% (n = 11) 2–17%
Sharp-shinned hawk	Absolute cell counts[a]	435 (n = 30)[b] 98–1,733	267 (n = 18) 55–990
	Differential cell counts[c]	7% (n = 31)[b] 2–12%	4% (n = 20) 2–13%

[a] Median number of cells per μl (sample size) range.
[b] Values differ significantly ($P < 0.05$) from those determined for the male birds of the same species.
[c] Median percentages of each cell (sample size) range.

of circulating leukocytes in both species. Lymphocyte to heterophil ratios also differed significantly between the species. Whereas the median ratio was 2.04 for the sharp-shinned hawks, concentrations were more nearly equal in the Cooper's hawk where the median lymphocyte to heterophil ratio was 1.22.

With each species, no significant differences were found between the hematological values obtained for the adult and immature birds (data not shown). Hematological values for male and female birds also were similar. Significant differences were detected only in the median absolute and differential eosinophil counts. Although the ranges of these values overlapped, median values were significantly higher in the female birds of both species (Table 2).

One adult male Cooper's hawk had a total white cell count of 32,727 cells/μl and one immature male Cooper's hawk had total white blood cell count 27,106 cells/μl. These values were respectively 13,287 and 7,666 cells/μl higher than the next highest total white blood cell count. Additionally, the adult male had *L. toddi* and *H. nisi* concentrations in excess of 3,200 parasites/μl of blood and the immature male had the highest percentage of parasitized cells with 24,395 *H. nisi*/μl of blood.

There was little variation for the PCV and estimated total solids among male and female and adult and immature birds (data not shown). Packed cell volumes for the Cooper's hawks (median value 52%, range 45 to 60%, n = 24) were nearly identical to those of the sharp-shinned hawks (median value 52%, range 45 to 65%, n = 48). Median estimated total solids for each species was 3.8 gm/dl. The range for the Cooper's hawks was 3.0 to 5.0 gm/dl (n = 24), for the sharp-shinned hawks 2.8 to 6.4 gm/dl (n = 48). A single female sharp-shinned hawk had severely lipemic serum. The estimated total solid reading for this individual was 6.4 g/dl, nearly double the median value.

No basophils were identified in the blood smears from either species. Unidentified cells ranged from 0 to 5% of the differential count. The largest percentage of these cells were round cells of similar size to the eosinophils and their cytoplasm contained densely packed clear vacuoles. The nuclei of most of these cells were mononuclear, although, infrequently, some were segmented.

Hematozoa were found in the blood smears from 18 Cooper's hawks (72%) and 43 sharp-shinned hawks (81%) (Table 4). The prevalence of infection ranged from

TABLE 3. Packed cell volume and estimated plasma total solids for Cooper's and sharp-shinned hawks in northern New York, 1987.

Species	Number sampled	Packed cell volume[a] (%)	Estimated total solids[a] (g/dl)
Cooper's hawk	24	52 (45–60)	3.8 (3.0–5.0)
Sharp-shinned hawk	48	52 (45–65)	3.8 (2.8–6.4)

[a] Values reported are the median (range).

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TABLE 4. Prevalence of *Leucocytozoon toddi* and *Hemoproteus* spp. in Cooper's and sharp-shinned hawks in northern New York, 1987.

Species	Number sampled	*L. toddi*	*H. nisi*	*H. elani*	*Haemoproteus* sp.[a]
Cooper's hawk	25	18 (72%)[b]	7 (28%)	2 (8%)	1 (4%)
Sharp-shinned hawk	53	43 (18%)	9 (19%)	5 (9%)	6 (11%)

[a] *Hemoproteus* was identified in these birds but could not be identified to species.
[b] Number infected with each respective parasite (prevalence).

40% of those obtained from adult female Cooper's hawks to 100% of those obtained from the immature male Cooper's and immature male sharp-shinned hawks. With the exception of a single sharp-shinned hawk in which only *H. elani* was found, all other birds infected with a *Haemoproteus* sp. also were concurrently infected with *L. toddi*. *Plasmodium circumflexum* was identified in a single immature male sharp-shinned hawk. *Trypanosoma avium* was identified in an immature male Cooper's hawk. Microfilariae were not observed.

There was a significantly higher prevalence of patent blood parasite infections and parasite concentrations were higher in the blood of birds captured after 24 April, compared to before 24 April 1987.

DISCUSSION

Although the two *Accipiter* species examined in this study are phylogenetically closely related, they had significant differences in some of their hematological parameters. In particular, sharp-shinned hawks had lower total white blood cell counts that were predominantly associated with lower heterophil concentrations. Absolute monocyte counts and eosinophil counts were also significantly lower in the sharp-shinned hawks, compared to Cooper's hawks. Statistically significant differences also were noted between the absolute and relative eosinophil counts of female and male birds in each species. While possibly evidence for physiological differences between the male and female cohorts, the differences were small enough as to have little significance in the evaluation of an individual bird.

Heterophils and eosinophils were easily differentiated. Eosinophil morphology was identical to that reported by Lind et al. (1990). The identity of the cells containing large clear cytoplasmic vacuoles remains unknown. Similar cells have been previously described (Lucas and Jamroz, 1961; Campbell, 1988) in other avian species. Campbell (1988) proposed that they are plasma cells containing Russell bodies, or unstained basophils or eosinophils. Lucas and Jamroz (1961) note that the granules contained with the avian basophil are water soluble and thus often are lost during staining. Based on their description of the degranulated basophil, it appears that many of the observed unidentified cells were basophils.

The median PCV of the two species examined in this study were nearly identical, but generally were higher than values reported for captive Falconiformes (Smith and Bush, 1978; Rehder et al., 1982). This discrepancy could be explained by a migration-induced dehydration, a stress-induced release of red blood cells into the circulation as occurs in some mammalian species (Jain, 1988), or elevated hemopoietic activity of free-flighted and migrating birds.

A high prevalence of *Leucocytozoon toddi* infection was found in both species examined, and a high prevalence of *Haemoproteus* spp. infection was found in the sharp-shinned hawk. The higher prevalence of infection and higher concentrations of parasites in birds captured after 24 April could be explained either by a

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spring recrudescence of patent infection similar to that observed in other avian species (Bennett and Cameron, 1974; Alverson and Noblet, 1977) or differences in parasite prevalence in adult and immature hawks. The finding that there was a higher prevalence of *Haemoproteus* spp. and *L. toddi* infections in spring captured sharp-shinned hawks as compared to in the fall and winter captured birds provides support to the spring recrudescence hypothesis.

It is unlikely that the low prevalence of *Plasmodium* sp. and *Trypanosoma* sp. and the absence of microfilariae represent the true prevalence of infection of these parasites in the birds sampled. Because of seasonal (Applegate, 1970) and diurnal (Noblet and Noblet, 1977) variation in parasite concentrations, or because of very low blood concentrations, examination of a single stained blood smear is a very insensitive assay for these parasites (Stabler et al., 1966). Specialized technique such as blood culture (Kirkpatrick and Lauer, 1985), examination of the buffy coat, and blood inoculation into a susceptible species (Herman et al., 1966) would be necessary to more accurately determine the true infection prevalence.

The finding that nearly all birds with circulating *Hemoproteus* spp. also had concurrent *L. toddi* parasitemias is evidence that specific host immunologic or physiological factors may similarly influence the replication of both of these parasites. The prevalences of *Leucocytozoon toddi* (79%) and *Haemoproteus* spp. (41%) infection in the sharp-shinned hawks reported here were significantly higher than those reported for sharp-shinned hawks (59% and 17%, respectively) captured in the fall and winter (Kirkpatrick and Lauer, 1985).

In conclusion, the hematological parameters determined in this study were obtained from birds who had successfully survived the winter, had completed much of their spring migration, and were found to be in good physical condition. While hematological values obtained from these birds may have been influenced by parasite burdens and the physiologic stress of migration and capture, we believe that these data represent a valuable reference point for the evaluation of sick and injured Cooper's and sharp-shinned hawks.

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