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HEMATOLOGY AND OCCURRENCE OF HEMOPARASITES IN MIGRATING SHARP-SHINNED HAWKS (*Accipiter striatus*) DURING FALL MIGRATION

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ABSTRACT.—Packed cell volume (%), total solids (g/dl), white blood cell count (cells/μl), differential and absolute white blood cell counts, and prevalence of hemoparasites were determined for 85 healthy sharp-shinned hawks (*Accipiter striatus*) during the 1991 fall migration. The packed cell volume (47.6 ± 6.73%), total solids (2.83 ± 0.58 g/dl) and white blood cell count (12 900 ± 7310 cells/μl) are within ranges reported previously for other raptors, both captive and wild. Immature birds showed a greater prevalence for the hemoparasites *Hemoproteus* and *Leukocytozoon* than adults but there was no significant difference in prevalence between males and females. These findings add to the small but growing data base on hematology of birds of prey.

KEY WORDS: *hematology*; *hemoparasites*; *migration*; *sharp-shinned hawk*; *stress.*

Hematología y ocurrencia de hemoparásitos en *Accipiter striatus* migratorios durante la migración otoñal de 1991

RESUMEN.—Se determinó el volumen celular (%), sólidos totales (g/dl), conteo de glóbulos blancos (células/μl), conteo diferencial y absoluto de glóbulos blancos y prevalencia de hemoparásitos para 85 individuos saludables de *A. striatus* durante la migración otoñal de 1991. El volúmen celular (47.6 ± 6.73%), los sólidos totales (2.83 ± 0.58 g/dl) y el conteo de glóbulos blancos (12 900 ± 7310 células/μl), están dentro de los rangos reportados previamente para otros rapaces, tanto cautivos como silvestres. Aves inmaduras mostraron una mayor prevalencia de los hemoparásitos *Hemoproteus* y *Leukocytozoon* que los adultos, pero no hubo una diferencia significativa en la prevalencia entre machos y hembras. Estos resultados se agregan a la pequeña pero creciente base de datos sobre la hematología de aves de presa.
[Traducción de Ivan Lazo]

Hematology is a useful tool in determining normal and pathological states in a variety of species including birds. The number of studies on raptor hematology is limited despite the large amount of information available for poultry. In addition, most reports are restricted to captive individuals or birds under rehabilitation and have a small sample size. Previous studies (Hunter and Powers 1980, Gessaman et al. 1986) have examined hematology from free-ranging raptors, which may vary from values obtained from captive birds. The purposes of this study were to add to the data base for hematology of wild raptors and to examine hematologic data for evidence of systemic disease.

MATERIALS AND METHODS

Subjects of our study were migrating sharp-shinned hawks (*Accipiter striatus*) captured for banding purposes at the Little Gap Banding Station near Hawk Mountain Sanctuary in eastern Pennsylvania. Birds were trapped from 9 September to 11 November 1991 between 0630 and 1635 H. Captured birds were weighed to the nearest gram. Each bird was classified as either immature (hatching year) or adult (second year or older) based on plumage characteristics.

Approximately 1 ml blood was collected from the jugular vein. Captured birds were injected with approximately 1 ml lactated Ringer's solution subcutaneously before release. A drop of whole blood was smeared onto a glass slide for measurement of the differential white blood cell count. A sample of whole blood was placed in a mi-

crohematocrit tube (approximately 70 μl) and spun at 10 000 rpm for 4 min to measure packed cell volume (PCV), or hematocrit. Total solids (TS), an estimate of the protein content, was measured from the plasma using a clinical refractometer.

Samples of whole blood were diluted and stained using a Unopette® Eosinophil staining kit (Becton-Dickinson and Co., Test No. 5877). Stained granulocytes (heterophils, eosinophils and basophils) were counted manually using a hematocytometer. Total white blood cell count (WBC) was calculated using methods described by Dein (1984): cells in 10 squares were counted and the result multiplied by 32 to adjust for dilution. This value was divided by the percentage of staining granulocytes in the differential white blood bell count to adjust for the lack of staining of lymphocytes and monocytes. Blood smears were dipped in the quick stain HEMA-3® (Biochemical Sciences, Inc.) for differential white blood cell counting. One hundred leukocytes were counted and the percent of each cell type was recorded. Differential counts were verified by having each slide read by three different analysts. Absolute white cell counts for each of the types of leukocyte were calculated by multiplying total white blood cell count by the percentage of each cell type in the differential count. Smears were also examined for the presence of blood parasites.

Mean, standard deviation, range, median, and percentiles are reported for the hematologic data. Analysis of variance (ANOVA) and Student *t*-tests were used to detect significant differences in selected parameters. An alpha level of less than 0.05 was considered significant. The Statistical Analysis System (SAS 1988) was used for analysis of data.

RESULTS

Results for packed cell volume (%), total solids (g/dl), white blood cell count (cells/μl), relative differential white blood cell count (% cell type), and absolute differential count (cells/μl) are presented in Table 1. We found that distributions of PCV, WBC, relative differential counts for heterophils, basophils, and monocytes, and absolute differential counts for all leukocyte types were not statistically normal (Kolmogorov statistic, W: Normal <95%). Therefore, percentiles may be more appropriate for accurate reporting of these data. Comparisons between males and females and between adults and immature birds found no significant differences in hematologic parameters, so results from all birds are compiled in Table 1. Prevalences (% affected) of the blood parasites *Hemoproteus* and *Leukocytozoon* detected from the smears are reported in Table 2. Comparison between males and females found no significant difference for either parasite. Comparison between adults and immature birds found that immature birds had a higher prevalence than adults for both *Hemoproteus* and *Leukocytozoon*. The hemo-

Table 1. Hematology of sharp-shinned hawks captured at Hawk Mountain Sanctuary.

	PCV (%)	TS (g/dl)	WBC (cells/μl)	Relative Differential WBC Count (%)						Absolute Differential WBC Count (cells/μl)				
				HET[a]	EO-SIN[b]	BASO[c]	LYMPH[d]	MONO[e]		HET[a]	EOSIN[b]	BASO[c]	LYMPH[d]	MONO[e]
N	85	85	72	81	81	81	81	81		72	72	72	72	72
Mean	47.6	2.83	12 900	27.0	7.67	0.40	63.4	1.60		3200	955	46	8480	187
SD	6.73	0.58	7310	14.2	4.18	0.79	14.4	1.99		2800	715	110	5350	296
Range	30–76	1.4–5.0	2400–37 600	5–69	0–18	0–4	28–90	0–10		240–18 800	0–3380	0–739	1770–23 700	0–1640
Median	47	2.8	11 520	26	7	0	65	1		2470	862	0	7930	111
50% percentile	44–52	2.4–3.2	7680–16 800	16–34	5–11	0–1	54–75	0–3		1810–3630	422–1270	0–22	4030–11 800	0–224
95% percentile	38–56	2.0–3.8	4240–25 400	8–52	1–15	0–2	35–83	0–4		820–9080	80–2520	0–241	1980–17 500	0–744

[a] Heterophils.
[b] Eosinophils.
[c] Basophils.
[d] Lymphocytes.
[e] Monocytes.

Table 2. Comparison of hemoparasite prevalence between immature and adult sharp-shinned hawks.

	TOTAL ($N = 83$)	IMMATURE vs. ADULT ($N = 60$)	($N = 23$)	t	df	P
Hemoproteus	20.5%	28.3%	0.00%	2.98	81	0.0038[a]
Leukocytozoon	16.9%	21.7%	4.35%	2.51	75	0.0143[a]

[a] Student's t-test. Probability <0.05 considered significant.

parasite *Plasmodium* was not detected on the blood smears.

Birds with white blood cell counts falling in the second and third quartiles are considered normal for this study. Those with a count above the third quartile are considered leukocytic. Those with a count below the first quartile are termed leukopenic. Comparisons of relative and absolute differential white blood cell counts between normal and leukocytic birds are presented in Table 3. Leukocytic birds had a significant absolute heterophilia, eosinophilia and lymphocytosis. However, differences were not significant between the two groups in relative differential white blood cell counts. Comparisons of relative and absolute differential white blood cell counts between normal and leukopenic birds are presented in Table 4. Leukopenic birds had a significant absolute heteropenia, eosinopenia, and lymphopenia and a significant relative heterophilia.

DISCUSSION

The mean hematocrit value of 47.6% falls within the range of reported values for trapped sharp-shinned hawks (49.5 ± 2.5%) given by Gessaman et al. (1986) and agrees with previously reported values for other falconiforms (Bond and Gilbert 1958, Elliott et al. 1974, Cooper 1975, Balasch et al. 1976, Smith and Bush 1978, Hunter and Powers 1980, Gee et al. 1981, Ferrer et al. 1987). Hematocrits of migrating, healthy birds of prey may be significantly greater than those in captivity. High altitude is known to increase the production of red blood cells due to the greater demand for efficient oxygen extraction from air. Hemoconcentration from dehydration may occur in migrating birds (Carpenter 1975, Gessaman et al. 1986, Perry et al. 1986) because of relative reduced intake of water. No significant difference was found between the hematocrits of males and females, although it has been suggested that male birds should have a higher hematocrit than females due to the erythropoietic effect of androgens (Gee et al. 1981, Sturkie 1986). Gessaman et al. (1986) found no significant difference in hematocrits between the sexes of trapped sharp-shinned hawks. Hunter and Powers (1980) and Snyder et al. (1980) also found no significant difference between the sexes in either

Table 3. Comparison of differential hematology between sharp-shinned hawks with white blood cell counts above the third quartile (leukocytic) and falling in the second and third quartile (normal).

	TOTAL ($N = 81$)	LEUKOCYTIC vs. ($N = 18$)	NORMAL ($N = 37$)	t	df	P
RELATIVE COUNTS (%)						
Heterophils	27.0	22.1	25.1	0.79	53.0	0.430
Eosinophils	7.67	6.44	8.54	1.82	53.0	0.074
Basophils	0.40	0.28	0.38	0.57	53.0	0.574
Lymphocytes	63.4	69.5	64.4	−1.37	53.0	0.176
Monocytes	1.60	1.61	1.57	−0.07	53.0	0.947
ABSOLUTE COUNTS (cells/ml)						
Heterophils	3200	5270	2880	−2.22	19.1	0.038[a]
Eosinophils	955	1420	1000	−2.11	53.0	0.039[a]
Basophils	46	72	43	−0.66	20.0	0.518
Lymphocytes	8480	15 400	7720	−6.88	24.4	0.0001[a]
Monocytes	187	321	163	−1.29	19.8	0.21

[a] Student's t-test. Probability <0.05 considered significant.

Table 4. Comparison of differential hematology between sharp-shinned hawks with white blood cell counts below the third quartile (leukopenic) and falling in the second and third quartile (normal).

	TOTAL	LEUKOPENIC vs.	NORMAL	t	df	P
RELATIVE COUNTS (%)	(N = 81)	(N = 26)	(N = 37)			
Heterophils	27.0	33.0	25.1	−2.27	61.0	0.027[a]
Eosinophills	7.67	7.27	8.54	1.13	61.0	0.263
Basophils	0.40	0.50	0.38	−0.52	37.5	0.607
Lymphocytes	63.4	57.8	64.4	1.90	61.0	0.063
Monocytes	1.60	1.65	1.57	−0.20	60.7	0.842
ABSOLUTE COUNTS (cells/ml)	(N = 72)	(N = 17)	(N = 37)			
Heterophils	3200	1720	2880	−3.39	48.7	0.001[a]
Eosinophils	955	361	1000	−5.24	51.9	0.0001[a]
Basophils	46	26	43	−0.75	52.0	0.454
Lymphocytes	8480	2840	7720	−9.88	46.7	0.0001[a]
Monocytes	187	98	163	−1.72	50.1	0.091

[a] Student's t-test. Probability <0.05 considered significant.

trapped or captive American kestrels (*Falco sparverius*). Age may also have an effect on hematocrit in that immature birds should have higher hematocrit values than adults (Rehder et al. 1982). No significant difference was found between immature birds and adults for hematocrit values in this study or that by Gessaman et al. (1986).

The mean total solids value of 2.83 g/dl was less than that reported previously for some falconiforms (Elliott et al. 1974, Halliwell et al. 1975, Smith and Bush 1978, Gee et al. 1981, Ferrer et al. 1987) but within the range reported by Balasch et al. (1976) and Snyder et al. (1980). Accurate comparisons with previously reported results may not be possible due to the different techniques in measuring protein levels in blood such as the Biuret and refractometric methods. Lumeij and de Bruijne (1985) showed that total solids measured with a refractometer have little correlation with serum protein levels measured by the Biuret method in rock doves (*Columba livia*). A low total solids value is often indicative of poor nutrition in raptors (Smith and Bush 1978, Ferrer et al. 1987). No significant difference in total solids was found between males and females or between adults and immature birds. These findings agree with those reported previously for some falconiforms (Snyder et al. 1980).

The prevalence of *Hemoproteus* in immature sharp-shinned hawks was 28.3%, yet no adults showed *Hemoproteus* parasitemia on the blood smears. On a similar note, 21.7% of immature birds had *Leukocytozoon* but only 4.35% of adults showed signs of infection. No significant difference in hemoparasite prevalence was found between males and females. This finding is in agreement with previous reports (Kirkpatrick and Suthers 1988, Davidar and Morton 1993). The greater prevalence of *Hemoproteus* and *Leukocytozoon* in immature sharp-shinned hawks from this study is in contrast to other reports. Kirkpatrick and Suthers (1988) found that hatching-year birds were infected at a lower rate than older birds representing 59 species from central New Jersey. Yearling purple martins (*Progne subis*) were significantly less infected with *Hemoproteus prognei* than adults (Davidar and Morton 1993). Ashford et al. (1990) reported a higher prevalence of *Leukocytozoon toddi* in adult sparrowhawks (*Accipiter nisus*). This study also suggested a vertical mode of transmission from adults to young in the nest through vector species (*Culicoides* for *Hemoproteus* and ornithophilic members of the family Simuliidae for *Leukocytozoon*). Relapses of hemoparasitemia as birds become stressed or begin breeding has been suggested (Peirce 1980). A loss of detectable levels of hemoparasite in the peripheral blood (latency) between the months of October and April with a spring relapse was reported by Ashford et al. (1990). It is not unreasonable to suggest that immature sharp-shinned hawks become infected in the nest and are less able to achieve latency of infection with hemoparasites due to their naive immunologic status and because of the stresses of first migration and incompletely developed hunting skills. The clinical effects of hemoparasites have not been completely deter-

Table 5. Total and relative differential white blood cell counts reported in various raptors.

			RELATIVE DIFFERENTIAL WBC COUNT (%)					
	N	WBC (cells/ μl $\times$ 10^3)	HETEROPHILS	EOSINOPHILS	BASOPHILS	LYMPHOCYTES	MONOCYTES	AUTHOR
Common buzzard (*Buteo buteo*)	11	14.0–49.0	20.5–39.8	5.5–19	0.25–8	35–65.5	0.25–3.75	a
Honey buzzard (*Pernis apivorus*)	1	10.5	29.8	9	4.8	55.3	1.25	a
Andean condor (*Vultur gryphus*)	1	13.5	42.8	11	2.5	42	1.8	a
Golden eagle (*Aquila chrysaetos*)	1	23.5	52.5	6	3.3	34	4.3	a
White-tailed sea eagle (*Haliaeetus albicilla*)	1	19.5	32.3	9.5	1.3	55	2	a
Imperial eagle (*Aquila heliaca*)	1	15.0	44	11.8	2.3	40.3	1.3	a
Tawny eagle (*Aquila rapax*)	1	42.5	57.3	10	2	30	0.8	a
Marsh harrier (*Circus aeruginosus*)	3	9.0–33.0	26.5–39.5	1.5–6.5	2.8–5.3	48–59.5	2.5–10.5	a
Common kestrel (*Falco tinnunculus*)	5	14.5–57.0	11.3–33	8.75–59.3	1.5–3.8	24-57.5	0.25–3.0	a
Black kite (*Milvus migrans*)	5	10.0–28.0	28.8–35.3	12.8–35.5	2.3–3.5	29.5–50.5	0–2	a
Red kite (*Milvus milvus*)	1	12.0	19.5	28.3	2.8	48.8	0.75	a
Egyptian vulture (*Neophron percnopterus*)	1	29.5	43.8	5.5	8.5	37.5	4.8	a

Table 5. Continued.

	N	WBC (cells/ $\mu l \times 10^3$)	Relative Differential WBC Count (%)					Author
			Heterophils	Eosinophils	Basophils	Lymphocytes	Monocytes	
King vulture (*Sarcoramphus papa*)	2	41.9						[b]
Hooded vulture (*Necrosyrtes monachus*)	1	22.4						[b]
Savannah hawk (*Heterospizias meridionalis*)	1	31.0						[b]
Ornate hawk-eagle (*Spizaetus ornatus*)	1	33.0						[b]
White-bellied sea eagle (*Haliaeetus leucogaster*)	1	22.0						[b]
Crested serpent eagle (*Spilornis cheela*)	1	22.0						[b]
Bald eagle (*Haliaeetus leucocephalus*)	1	11.3						[b]
Collared forest falcon (*Micrastur semitorquatus*)	1	26.4						[b]
Crested caracara (*Polyborus plancus*)	2	24.1						[b]
Red-tailed hawk (*Buteo jamaicensis*)	2	6–46						[c]
Harris' hawk (*Parabuteo unicinctus*)	2	12–14						[c]

[a] Christoph and Borowski (1961).
[b] Elliott et al. (1974).
[c] Halliwell et al. (1975).

mined. Hemoparasites have been shown to increase rehabilitation time in raptors (Olsen and Gaunt 1985). However, most studies on wild birds show no evidence of decreased longevity or reproductive ability in infected birds (Kirkpatrick and Suthers 1988, Ashford et al. 1990, Davidar and Morton 1993), although a negative effect on mate selection has been suggested (Kirkpatrick and Suthers 1988, Ashford et al. 1990, Davidar and Morton 1993). No significant differences in measured hematologic parameters were identified between hemoparasitemic and non-hemoparasitemic birds in our study, indicating no correlation between infection and other hemogram indicators of general health status.

The mean total WBC was 12 900 ± 7310 cells/μl. This value falls within range of normal counts for other falconiforms (Elliott et al. 1974, Halliwell et al. 1975, Smith and Bush 1978). The relative differential white cell counts also agree with those previously reported (Table 5). It is difficult, if not impossible, to measure normal white blood cell counts accurately in raptors since these birds are unavoidably stressed when handled and especially when captured in the wild. Migration is also a source of stress to a raptor. ACTH and corticosteroids have been shown to be elevated during periods of stress in birds (Wolford and Ringer 1962). In poultry the hematologic response to corticosteroids is leukocytosis with heterophilia and lymphopenia (Hublé 1955, Glick 1961, Bell and Freeman 1971). However, it must be emphasized that the hematologic response to stress varies from species to species. For example, Bhattacharyya and Sarkar (1968) found that in the rock dove, the house crow (*Corvus splendens*) and the cattle egret (*Bubulcus ibis*) the response to cortical stimulation by ACTH and unilateral adrenalectomy was heteropenia and lymphocytosis. Only the common myna (*Acridotheres tristis*) responded in a similar fashion to poultry with heterophilia and lymphopenia in that study.

Levels of epinephrine and norepinephrine have been shown to be elevated during migration of the common snipe (*Gallinago gallinago*) and the rose-colored starling (*Sturnus roseus*; Epple and Stetson 1980). Information about the effect of catecholamines on avian hematology is lacking, but these substances are known to cause leukocytosis with neutrophilia and lymphocytosis in mammals (Duncan and Prasse 1986). Without having measured serum levels of catecholamines and corticosteroids during this study, it is impossible to determine the relative level of stress for each bird. Birds with a total white blood cell count above the third quartile (leukocytic) had an absolute heterophilia, lymphocytosis and eosinophilia which may reflect more of a catecholamine-induced stress pattern rather than a corticosteroid-induced hemogram (which should present with lymphopenia). However, we caution against over-interpretation of the data since hormone levels were not measured and the effects of catecholamines and glucocorticoids on the hemogram are not known. Leukocytosis is often present with bacterial, fungal or parasitic infections, whereas leukopenia may accompany viral infections. Most hawks captured in this study appeared healthy on physical examination, showing no signs of clinical disease. Seriously ill birds might not be able to migrate and therefore would not be included in this study. Birds in this study with leukocytosis or leukopenia may have been ill, but the source of possible infection was not determined. We warn, however, that with lure traps a hungrier subset of the migrating population might have been sampled. This group may include birds who have no evidence of disease yet are subclinically ill and perhaps hungrier than healthy birds. But because no evidence of clinically significant disease states could be detected in the subjects, the white blood cell counts and other hematologic parameters for all birds from this study should be analyzed as a spectrum of hematologic findings for migrating, clinically healthy sharp-shinned hawks.

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