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Hematology of Sloth Bears (Melursus ursinus ursinus) from Two Locations in India

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ABSTRACT: Standard hematology parameters were determined for 122 sloth bears (Melursus ursinus ursinus) at the Sur Sarovar Bird Sanctuary, Uttar Pradesh, India (27°0'N; 77°45′E), and the Bannerghatta Biological Park, Karnataka, India (12°48'N; 77°34'E) from March 2003 to July 2006. These two native sloth bear habitats have different climatic conditions and provided an opportunity to examine the effect of climate on the physiologic hematology values of these bears. We primarily analyzed the influence of age, sex, season, and body weight on the different hematology parameters. Several values were significantly different in sloth bear cubs (≤1 yr) when compared to adult and sub-adult bears (>1 yr). The cubs had a lower erythrocyte count, hemoglobin concentration, packed cell volume (PCV), and mean cell hemoglobin (MCV) values when compared to adult and subadult bears. The cubs also had higher leukocyte counts, due to higher circulating neutrophils, as compared to adult and subadult bears. Within subadult and adult bears, we also identified a sexual dimorphic difference in leukocyte count in adult and subadult bears, wherein female bears had higher counts than males. This difference was the result of a significantly higher number of circulating neutrophils in female bears. Platelet counts were also higher in females as compared to males. On comparing different seasons, leukocyte counts were higher in winter as compared to the summer and monsoon seasons. When compared based on location, erythrocyte counts were higher in subadult and adult bears at Bannerghatta, which was at a higher altitude than Sur Sarovar. Within subadult and adult bears, we did not find any significant influence of age or body weight on the different hematologic parameters. In this study we have obtained mean hematologic values for sloth bears in their native habitat to serve as a reference for this species. This report will be useful to develop and evaluate health profiles of sloth bears under various ecological conditions.

Key words: Blood, hematology, India, Melursus ursinus ursinus, sloth bear.

Hematology, essential for assessing health in both captive and wild animal populations, has been studied for several species in the bear family (Ursidae). Most of these studies have been done for American black bears (Ursus americanus; Svihla et al., 1955; Youatt and Erickson, 1958; Hellgren et al., 1993), American brown bears (grizzly; Ursus arctos horribilis; Cattet et al., 2003a; 2003b) and European brown bears (Ursus arctos arctos; Seal et al., 1967; Kusak et al., 2005). Factors influencing the hematology parameters in some of these species have also been studied based on nutritional condition (Gau and Case, 1999), seasonal patterns of metabolism (Erickson and Youatt, 1961; DelGiudice et al., 1991; Hissa et al., 1994; Kusak et al., 2005), and the effect of immobilizing drugs (Bush et al., 1980; Cattet et al., 2003b; Kusak et al., 2005). However, there have been no defined studies evaluating hematology values for the sloth bear (*Melursus ursinus*).

Sloth bears inhabit the tropical and subtropical regions of the Indian subcontinent and are distributed from the foothills of the Himalayas to the southern end of the Western Ghats mountain range in India, as well as in the island of Sri Lanka (Prater, 1965). In these regions, two subspecies are present, *Melursus ursinus ursinus* distributed across the Indian peninsula, and the shorthaired and relatively smaller *Melursus ursinus inornatus* seen only in Sri Lanka (Pocock 1933). Around 5–7 million yr ago, the ancestor of sloth bears was the first to diverge from

the sun (Helarctos malayanus), American black, Asiatic black (Ursus thibetanus), brown, and polar (Ursus maritimus) bears in the ursine phylogenetic tree (Zhang and Ryder, 1993, 1994; Talbot and Shields, 1996; Yu et al., 2004). Ursavus, the ancestral taxon, was strictly carnivorous (Martin, 1989), and phylogenetic analyses suggest that the extant sloth bears arose by rapid radiation events leading to a divergence that transformed a generalized carnivore to an ecomorph (Waits et al., 1999). Despite long periods of evolutionary separation, sloth bears have retained their carnivore morphology but have developed unique physiologic adaptations to live in tropical/subtropic climes, with substantial reliance on frugivory (feeding on fruits) and myrmecophagy (feeding on ants and termites) (Pocock, 1933; Laurie and Seidensticker, 1977; Gokula et al., 1995; Bargali et al., 2004). Earlier studies on bear ecology have accurately predicted this development to be due to competitive pressure and temporal patterning of resource availability in the habitat (Jaffeson, 1975; Laurie and Seidensticker, 1977). As a result, these medium-sized bears, in contrast to other ursids, have developed several characteristics common to other myrmecophagous mammals. These characteristics include a more nocturnal activity pattern (Sunquist, 1982; Yoganand et al., 2005), extended parental behavior (Laurie and Seidensticker, 1977), and smaller home ranges (Sunquist, 1982; Joshi et al., 1995). Due to such adaptations, sloth bears have metabolic differences compared to other species of bears. They have been shown to have an overall lower metabolic rate compared to brown and polar bears, and torpor (winter sleep) is unheard of in sloth bears (McNab, 1992).

One study during the second half of the twentieth century estimated approximately 7,600 sloth bears occupying the Indian Peninsula (Jaffeson, 1975). Although more continuous throughout this range at that time, habitat loss and fragmentation due

to expanding human habitation and agriculture have destabilized this population that now occurs only in isolated pockets (Garshelis et al., 1999). Further deterioration of these habitat pockets is evidenced by an increase in the number of humanbear conflicts in the encroached buffer regions (Rajpurohit and Krausman, 2000; Bargali et al., 2005). A recent report detecting the unusual presence of sloth bear hair in tiger scat (Biswas and Sankar, 2002) suggests an imbalance in the food chain due to scarcity of prey-base for tigers, underscoring the pressure on the ecosystem as a whole in these areas (Akhtar et al., 2004, 2006). Beyond these issues, the use of bear gall bladder in traditional medicine and illegal trade of cubs for training and exhibition as "dancing bears" have greatly accelerated their population decline. Sloth bears were classified as vulnerable in the International Union for the Conservation of Nature and Natural Resources (IUCN) Red List of Threatened Species in 1990 (IUCN, 1990) and are protected under Schedule I of The Indian Wildlife (Protection) Act (IWPA), 1972 (IWPA, 1972). Although a recent survey has not been conducted, studies on some of these disturbed and fragmented ranges showed inadequate resources to support self-sustaining sloth bear populations (Akhtar et al., 2004; Yoganand et al., 2005).

Despite the need for conservation and rescue efforts for sloth bears, limited information is available regarding sloth bear biology, including physiologic reference data. The dissimilarities in sloth bear physiology, when compared to other ursids evident from their morphologic appearance (Pocock, 1933), feeding behavior (Gokula et al., 1995; Joshi et al., 1997), and physiologic state (McNab, 1992), warrants studies for better understanding and improved veterinary care and management of sloth bear populations. The only published material showing some sloth bear hematology values was in relation to the effect of dissociative anesthetics on bears; this study included three sloth bears at the National Zoological Park, Smithsonian Institution, Washington, D.C. (Bush et al., 1980). Therefore, we carried out this study to: 1) determine hematology reference values for sloth bears at two locations within their native habitat; 2) determine if there were any differences due to age, sex, season, body weight, and location; and 3) compare values of these parameters to existing data on other ursids.

Wild, orphaned, and rescued and rehabilitated sloth bears, under free ranging or semicaptive conditions, were utilized for this study: 84 bears at the Center for the Conservation and Rehabilitation of Bears, Agra Bear Rescue Facility, Sur Sarovar Bird Sanctuary, Keetham, Agra, Uttar Pradesh, India $(27^{\circ}0'\text{N}; 77^{\circ}45'\text{E})$ and 38 bears at the Bannerghatta Bear Rescue Center, Bannerghatta Biological Park, Bannerghatta, Bangalore, Karnataka, India (12°48'N; 77°34'E). Both these locations are within the recorded habitat range for this species. At both facilities, bears were maintained on an enriched native diet of local seasonal fruits and grains. Age of the bears was obtained from animal records. Ages ranged from 6 mo to 20 yr, and for the purposes of this study, bears were classified as cubs (≤1 yr) or adults and subadults (>1 yr). Cubs were analyzed separately and were not included in the general comparisons elaborated below. Subadult (>1 yr to ≤ 3 yr) and adult (>3 yr) bears were not significantly different in body weight and were therefore evaluated together for all analyses except for those based on age. Rescued bears used for this study were introduced into the facility at least 8 mo before sampling. All bears sampled appeared physically healthy, with normal behavioral responses, and were clinically healthy during examination at the time of sampling (as determined by body temperature, hydration, heart/respiration rate, and a detailed external physical examination). Furthermore, all bears utilized in this study were also pretested for exposure to diseases enzootic in these regions and for intestinal parasites; they were mostly parasite-free intestinally and were found negative for hepatitis B, rabies, tuberculosis, and leptospirosis.

Blood samples were collected throughout the year from March 2003 to July 2006, thus covering the three major seasons in India; summer (1 April through 31 May), monsoon (1 June through 31 October), and winter (1 November through 31 March). Each bear was sampled only once for this study. Bears were immobilized using a ketamine-xylazine combination (Page, 1986); ketamine hydrochloride (5 mg/kg body weight; Ketamil[®], Troy Laboratories Pty Ltd., Smithfield, NSW, Australia) and xylazine hydrochloride (Xylazil®, 2 mg/kg body weight; Troy Laboratories Pty Ltd.). A premedication of atropine sulfate (Atrosite®, 0.025 mg/kg body weight; Troy Laboratories Pty Ltd.) was administered approximately 30 min prior to immobilization. These drugs were administered using a blowgun on unsuspecting bears, thus causing minimal excitation during the procedure. Blood was collected from the jugular vein within 10 min after immobilization using a 20-gauge sterile hypodermic needle in Vacutainers (Becton Dickinson, Franklin Lakes, New Jersey, USA), with and without ethylene diamine tetraacetic acid for hematology and serology, respectively. Samples were immediately stored on cool packs at 4-8 C and transported from both field locations to the laboratory at the Agra Bear Rescue Facility (Keetham, Agra). Standard hematology parameters such as erythrocyte count, leukocyte count, platelet count, hemoglobin, packed cell volume (PCV), erythrocyte sedimentation rate (ESR), mean cell volume (MCV), mean cell hemoglobin (MCH), and mean cell hemoglobin concentration (MCHC) were analyzed for each sample within 24 hr of collection using a hematology analyzer (Medsource Ozone Biomedicals Pvt. Ltd., Faridabad, India). Blood smears

were made and stained using Wright-Giemsa stain. Differential counts evaluating the percentage of each cell type in the smear were done under oil immersion using a light microscope. Absolute differential leukocyte counts were determined by multiplying relative percentages with the total leukocyte count.

Data from subadult and adult bears were categorized and analyzed based on: 1) sex (male versus female); 2) location (Sur Sarovar versus Bannerghatta); and 3) season (summer, monsoon and winter). Sloth bear cubs were analyzed separately and comparisons were made to the grouped subadult and adult bears. The sex ratios at Sur Sarovar and Bannerghatta were taken into consideration while comparing the two locations. Statistical analyses were performed using JMP 6.0.2 software (SAS Institute, Cary, North Carolina, USA). The distribution for each variable was evaluated before and after categorization. Each distribution was tested for normality using the normal probability plot and the Shapiro-Wilk statistic. Outliers were only a rare occurrence and were not removed from any of the analyses. Basic statistics including mean, median, standard deviation, and 25% and 75% quartiles were determined for each variable. For normally distributed variables, a 95% confidence interval for means was calculated. Homogeneity of variance for each variable was tested using the Levene's test for equality of variances. For variables satisfying the assumptions of normality and homogeneity of variance, comparisons were made either using a 2sample t-test (two groups) or by using a one-way analysis of variance (ANOVA; >two groups). If significant P values occurred in the ANOVA, specific between-group differences were evaluated using the post hoc Tukey-Kramer test. For data violating the assumptions of normality and homogeneity of variance, a Kruskal-Wallis one-way ANOVA was performed. Potential effects of age or body weight on the different parameters were also investigated; simple bivariate plots

were constructed and the fit for a regression model was tested for each parameter. For all the above statistics, values of P < 0.05 were considered significant.

The majority of bears sampled were adults (n=100). Adult and sub-adult (n=10) bears were evaluated together for all comparisons except for age. Bear cubs (n=12) were analyzed separately. The overall sex ratio of adult and sub-adult bears from both Sur Sarovar (n=75) and Bannerghatta (n=35) locations was 1.2 (60 males; 50 females). The sex ratios within each location were 1.5 (45 males; 30 females) at Sur Sarovar and 0.75 (15 males; 20 females) at Bannerghatta. For the sloth bear cubs sampled in this study, the sex ratio was 1:1 (six males; six females). Samplings were randomly distributed during the 3 yr of this study. Analysis of seasonal effects was performed on grouped data for summer (n=48), monsoon (n=50), and winter (n=12). There was a significant difference between the average male and female body weights (males: 93.2±22.0 kg; females: $83.2\pm22.1 \text{ kg}$; P=0.02). However, the difference between male and female body weight did not reach statistical significance when comparisons were made within the Sur Sarovar and Bannerghatta locations (P=0.29 and P=0.06, respectively).

Results of hematology values determined in the 110 adult and sub-adult bears are shown in Table 1. These are presented as averages for the entire population and are also separated based on sex. Some parameters measured in this study for sloth bears were comparable to average values recorded in studies on nonhibernating black (Svihla et al., 1955; Youatt and Erickson, 1958; Hellgren et al., 1993), brown (Kusak et al., 2005) and grizzly (Cattet et al., 2003a) bears. We did not observe a lower erythrocyte count in female bears as recorded for European brown bears (Kusak et al., 2005). The MCV and MCH values were higher in male bears compared to females (P < 0.01for both values); however, these values

Table 1. Hematology values for sloth bears, *Melursus ursinus*, combined and categorized based on sex.

		Bears combined ^b		Adult/sub-adult males ^c		Adult/sub-adult females ^d	
Parameters ^a (units)	n	Mean (median)±SD	95% CI or [25– 75% quartiles]	n	Mean (median)±SD	n	Mean (median)±SD
Erythrocytes (10 ¹² /l)	92	$5.7(5.7)\pm1.1$	5.5–5.9	49	$5.5 (5.6) \pm 1.1$	44	$5.9(5.9)\pm1.0$
Leukocytes (10 ⁹ /l)	107	$12.9\ (12.0)\pm4.0$	12.1 - 13.6	59	$12.1^{f,g} (11.3) \pm 3.7$	49	$13.8^{f,g} (13.2) \pm 4.2$
Neutrophils (10 ⁹ /l)	107	$8.6 (7.6) \pm 3.6$	8.0-9.3	59	$8.0^{f,g} (6.8) \pm 3.7$	49	$9.5^{f,g}(8.9)\pm3.3$
Lymphocytes (10 ⁹ /l)	107	$2.9(2.8)\pm1.3$	2.6 - 3.1	59	$2.9(2.9)\pm1.0$	49	$2.9(2.5)\pm1.6$
Monocytes (10 ⁹ /l)	107	$0.3 (0.2) \pm 0.2$	0.3 - 0.4	59	$0.3 (0.2) \pm 0.2$	49	$0.3 (0.2) \pm 0.3$
Eosinophils (10 ⁹ /l)	107	$0.9 (0.7) \pm 0.9$	0.8 - 1.1	59	$0.9 (0.6) \pm 0.8$	49	$1.0 (0.8) \pm 0.9$
Basophils $(10^9/l)^e$	119	$0.006 (0.0) \pm 0.03$	[0.0-0.0]	59	$0.005 (0.0) \pm 0.03$	49	
Platelets (10 ¹¹ /l)	58	$4.3(4.3)\pm1.3$	4.0 - 4.6	33	$4.0^{\rm f,g,h} (4.2) \pm 1.2$	25	$4.7^{\rm f,g,h}$ $(4.6)\pm1.4$
Hemoglobin (gm %)	106	$14.5 (14.5) \pm 1.8$	14.1 - 14.8	59	$14.4 (14.4) \pm 2.0$	48	$14.5 (14.6) \pm 1.9$
PCV (%)	106	41.9 (43.9)±11.3	39.7-44.1	59	$41.5 (43.5) \pm 12.5$	48	$42.0 (44.2) \pm 10.2$
ESR (mm, 60 min) ^e	84	$30.5 (18.0) \pm 31.8$	8 [8.0–41.8]	44	$27.0 (17.0) \pm 26.9$	41	$33.6 (20.0) \pm 36.4$
MCV (fl)	91	$79.4\ (79.4)\pm12.8$	3 76.7–82.1	49	$82.6^{f,g} (82.5) \pm 13.3$	43	$75.0^{f,g} (76.3) \pm 11.8$
MCH (pg)	91	$25.6 (26.5) \pm 3.1$	24.9 - 26.2	49	$26.2^{f,g} (27.1) \pm 3.0$	43	$24.7^{f,g} (25.5) \pm 3.3$
MCHC (gm %)	91	$32.1 (33.3) \pm 3.7$	31.3-32.9	49	$31.9 (33.3) \pm 2.6$	43	$32.3 \ (33.3) \pm 4.6$

^a PCV = packed cell volume; ESR = erythrocyte sedimentation rate; MCV = mean cell volume; MCH = mean cell hemoglobin; MCHC = mean cell hemoglobin concentration.

were only significantly different between males and females at the Sur Sarovar, but not the Bannerghatta, location. Leukocyte counts were found significantly higher in female bears compared to male bears (P=0.02). The total neutrophil count was also higher in females compared to males (P=0.03). However, when compared within the Sur Sarovar and Bannerghatta locations, these were true only for bears at Sur Sarovar; although we saw a trend, the difference did not reach statistical significance for bears at Bannerghatta. None of the other differential leukocyte counts were different between male and female bears. Although unexplained, the higher neutrophil counts in female bears appeared to be similar to that reported for humans (Bain and England, 1975). A reciprocal sexually dimorphic difference in neutrophil count has been reported in brown bears, wherein males had a higher

neutrophil count compared to females (Kusak et al., 2005). Female sloth bears also had a higher platelet count compared to male bears in this study (P=0.03). Similar differences in platelet counts between the sexes have been reported for humans (Bain, 1985). A recent study exploring platelet function in mice has shown that there are also sexually dimorphic functional differences, where platelets derived from females were more responsive to stimuli than those derived from males (Leng et al., 2004). The involvement of estradiol in triggering proplatelet formation (Nagata et al., 2003) and platelet potentiation (Moro et al., 2005) have also been reported.

Several of the hematology values in sloth bears cubs were significantly different from the adult and sub-adult bears (Table 2). The cubs had a lower erythrocyte count compared to adult and subadult bears

^b Adults/sub-adults combined (mean body weight±SD): 88.7±22.5 kg.

^c Adult/sub-adult males (mean body weight±SD): 93.3±21.9 kg.

^d Adult/sub-adult females (mean body weight±SD): 83.2±22.1 kg.

^e Data for this parameter were not normally distributed.

^f Significantly different from the opposite sex (P < 0.05).

g Parameter significantly different from the opposite sex within the Sur Sarovar location (P<0.05).

^h Parameter significantly different from the opposite sex within the Bannerghatta location (P<0.05).

Table 2. Hematology values for sloth bear cubs.

	Cubs ≤12 months ^b					
Parameters ^a (units)	n	Mean (median)±SD	95% CI or [25–75% quartiles]			
Erythrocytes (10 ¹² /l)	12	$4.8^{\rm d} (4.7) \pm 0.8$	4.3–5.4			
Leukocytes (10 ⁹ /l)	12	$15.2^{d} (15.5) \pm 3.2$	13.1–17.2			
Neutrophils (10 ⁹ /l)	12	$12.1^{d} (14.5) \pm 3.1$	10.1–14.0			
Lymphocytes (10 ⁹ /l)	12	$2.3(2.1)\pm1.2$	1.6–3.1			
Monocytes (10 ⁹ /l)	12	$0.2 (0.2) \pm 0.1$	0.2-0.3			
Eosinophils (10 ⁹ /l)	12	$0.6^{\rm d} \ (0.6) \pm 0.4$	0.3-0.8			
Basophils $(10^9/l)^c$	12	$0.01 (0.0) \pm 0.03$	[0.0-0.0]			
Platelets (10 ¹¹ /l)	9	$5.5(5.8)\pm1.8$	4.0-6.9			
Hemoglobin (gm %)	12	$11.3^{d} (10.7) \pm 2.7$	9.5-13.0			
PCV (%)	12	$33.6^{d} (33.5) \pm 6.5$	29.5–37.7			
ESR (mm, 60 min) ^c	12	$43.0 (22.5) \pm 43.1$	[11.3–90.8]			
MCV (fl) ^c	12	$70.1^{d} (73.2) \pm 7.9$	[66.8–75.1]			
MCH (pg)	12	$22.8^{d} (23.4) \pm 2.7$	21.2-24.6			
MCHC (gm %) ^c	12	$32.3 (33.1) \pm 6.1$	[31.7–33.6]			

^a PCV = packed cell volume; ESR = erythrocyte sedimentation rate; MCV = mean cell volume; MCH = mean cell hemoglobin; MCHC = mean cell hemoglobin concentration.

(P < 0.01). This was also reflected in the measures of hemoglobin concentration, PCV, MCV, and MCH, all of which were significantly decreased in the cubs (P < 0.01for all). Lower values for these erythrocyte parameters in cubs could be indicative of plasma expansion associated with rapid development and body growth in neonates, thereby exceeding the rate of red blood cell production. Similar observations have been reported for other neonatal mammals in several other studies (Bryden and Lim, 1969; Boily et al. 2006). The lower hemoglobin, PCV, MCV, and MCH, compared to subadult and adult bears, indicated that the cubs may evidence microcytic anemia during the rapid growth phase; this is similar to a report on black bear cubs (Matula et al., 1980). Previous studies on brown bears have also noted that young bears had a lower erythrocyte, hematocrit, and hemoglobin value when compared to older bears (Pearson and Halloran, 1972). Cubs also had higher neutrophil and lower eosinophil numbers (P<0.01 and P=0.02, respectively) and overall a higher leukocyte count (P=0.04) as compared to adult and subadult

bears. This indicates a potential physiologic leukocytosis due to higher activity in cubs compared to adult bears (Swenson, 1984).

The two locations where samples were collected from sloth bears had differences in the seasonal variations of temperature and humidity. In Bannerghatta, which is at a higher altitude (920 m), the temperature and humidity values averaged 27 C (20-32 C) and $\sim 60\%$ respectively in summer, 24 C (20–29 C) and ~77% in monsoon, and 22 C (15–28 C) and \sim 62% in winter. In Sur Sarovar, which is at a lower altitude (168 m), these values were 29 C (22-39 C) and ~14% in summer, 28 C (20-39 C) and \sim 60% in monsoon, and 14 C (6– 27 C) and \sim 59% in winter (Source: The Weather Underground, Inc., Ann Arbor, Michigan, USA, www.wurderground.com; values recorded in 2005-2006). Comparing bears between these two regions, we found some significant differences in hematologic values (Table 3). Erythrocyte counts and MCHC were higher in bears at Bannerghatta compared to bears at Sur Sarovar (P < 0.01 for both values). However, PCV, MCV, and MCH values were all

^b Cubs (mean body weight±SD): 23.5±6.7 kg.

^c Data for this parameter were not normally distributed.

^d Significantly different from adult bears (P<0.05).

Location^b Season Sur Sarovar Bannerghatta Monsoon Summer Winter $Mean \pm SD$ Parameters^a (units) Mean±SD Mean±SD $Mean \pm SD$ Mean±SD $6.2^{f} \pm 1.2$ Erythrocytes (10¹²/l) 58 $5.4^{e} \pm 0.9$ 34 44 5.7 ± 0.7 38 5.9 ± 1.4 10 5.2 ± 0.8 $16.2^{h} \pm 4.7$ $14.5^{\rm f} \pm 4.8$ Leukocytes (10⁹/l) 73 $12.1^{\rm e} \pm 3.4$ 48 $12.5^{g}\pm3.3$ $12.4^{g}\pm4.2$ 34 48 11 $12.2^{h} \pm 5.3$ Neutrophils (10⁹/l) 73 $7.8^{e} \pm 3.0$ 34 $10.5^{\rm f} \pm 4.1$ 48 $8.0^{g}\pm2.5$ 48 $8.4^{g}\pm3.6$ 11 Lymphocytes (10⁹/l) 73 2.9 + 1.134 3.0 ± 1.6 48 3.0 ± 1.2 48 2.8 ± 1.3 11 2.8 ± 1.4 $0.4^{\rm f} \pm 0.3$ Monocytes (10⁹/l) 73 $0.3^{e} \pm 0.2$ 34 48 0.4 ± 0.3 48 0.3 ± 0.2 11 0.4 ± 0.3 Eosinophils (10⁹/l) $0.4^{\rm f} \pm 0.5$ 73 $1.2^{\rm e} \pm 0.9$ 34 48 1.0 ± 1.0 48 1.0 ± 0.7 0.6 ± 0.8 11 $0.02^{f} \pm 0.06$ Basophils $(10^9/l)^d$ 73 $0.0^{e} \pm 0.0$ 34 48 0.01 ± 0.1 48 0.0 ± 0.0 11 0.0 ± 0.0 Platelets $(10^{11}/l)^i$ $6.3^{h} \pm 1.8$ 58 4.3 ± 1.3 0 26 $4.5^{g}\pm1.0$ 28 $3.8^{g}\pm1.2$ 4 Hemoglobin (gm %) $47\ 14.3^{g,h} \pm 1.7$ $15.0^{h} \pm 1.7$ 73 14.6 ± 1.7 33 14.2 ± 2.0 11 13.3g±2.0 $33.6^{\text{f}} \pm 15.1$ $47.0^{h} \pm 6.6$ $34.4^{g} \pm 12.9$ PCV (%) 73 $44.7^{e} \pm 6.3$ 33 47 $38.5^{g}\pm12.7$ 48 $18.8^{\mathrm{f}} \pm 25.5$ ESR (mm, 60 min)^d $35.7^{e} \pm 33.2$ 58 26 38 37.8 ± 34.7 38 21.4 ± 26.7 8 39.1 ± 32.6 $69.0^{\text{f}} \pm 11.1$ MCV (fl) 58 $85.3^{e} \pm 9.6$ 33 43 77.1 ± 10.9 38 83.0 ± 14.9 10 75.7 ± 9.2 $23.5^{f} \pm 4.1$ $26.7^{\mathrm{e}} \pm 1.4$ MCH (pg) 58 33 43 25.4 ± 2.9 38 25.9 ± 3.4 10 25.1 ± 2.7 $33.8^{f} \pm 0.9$ $31.1^{e} \pm 4.3$ 33 MCHC (gm %) 58 43 32.3 ± 4.8 38 31.5 ± 2.6 10 33.1 ± 1.8

Table 3. Effect of geographical location and season on hematology in sloth bears.

significantly lower in bears at Bannerghatta compared to bears at Sur Sarovar location (P < 0.01 for all). The erythrocyte count, hemoglobin, and associated parameters were those that have been previously reported to change in concurrence with changes in altitude, relative atmospheric O₂ concentration, environmental temperature, and other climatic factors (Sealander, 1964; Swenson, 1984). Although the bears at Bannerghatta (at a higher altitude) had a higher mean erythrocyte count and MCHC compared to the bears located at Sur Sarovar, the associated lower PCV, MCV, and hemoglobin values in these bears suggest that their erythrocytes were relatively microcytic when compared to the bears at Sur Sarovar. The ESR as determined for 60 min was significantly delayed in bears at Sur Sarovar as compared to bears at Bannerghatta (P=0.01). Leukocyte counts were higher in bears at Bannerghatta compared to the bears at Sur Sarovar (P < 0.01). The Bannerghatta bears also had a higher number of circulating neutrophils (P < 0.01) and monocytes (P=0.02) compared to the bears at Sur Sarovar, whereas bears located at Sur Sarovar had an increased number of eosinophils (P < 0.01) compared to bears at Bannerghatta. This higher eosinophil count potentially suggests the prevalence of specific allergens or certain subtle parasitisms in the Sur Sarovar region. The difference in sex ratio partially explained the higher leukocyte and neutrophil counts in bears at Bannerghatta; however, comparisons of the same sexes between the two locations were not significantly different.

Analysis of data collected from the different seasons showed no significant differences in cell counts between bears

^a PCV = packed cell volume; ESR = erythrocyte sedimentation rate; MCV = mean cell volume; MCH = mean cell hemoglobin; MCHC = mean cell hemoglobin concentration.

^b Age and body weights of adult/sub-adult bears compared were not significantly different between the two locations (P < 0.05); Male:Female ratio for the two locations were 1.5 for Sur Sarovar and 0.75 for Bannerghatta.

 $^{^{}c}$ Age and body weights of a dult/sub-adult bears compared were not significantly different between the three seasons (P<0.05).

^d Data for this parameter were not normally distributed.

 $^{^{\}mathrm{e,f}}$ Significantly different between Sur Sarovar and Bannerghatta locations (P<0.05).

gh Tukey-Kramer comparisons for each parameter between the three seasons (indicated only for parameters showing significant differences; P<0.05).

¹ Comparisons for this parameter are not robust due to insufficient statistical power.

sampled during either the monsoon or summer seasons. However, bears sampled in winter showed a significantly increased leukocyte count compared to bears sampled during the monsoon (P < 0.01) and summer (P < 0.01) seasons. This increase was again associated with a higher neutrophil count in bears sampled during both the monsoon (P < 0.01) and summer (P < 0.01) seasons. Although increases in leukocyte counts, especially neutrophils, can be associated with inflammatory reactions to potential pathogens or parasites (Swenson, 1984), the higher values seen during winter in this study were not accompanied by an incidence of clinical disease in any of the animals. Because segmented- and band-neutrophils were not differentiated in this dataset, this finding is hard to interpret. Hemoglobin concentrations were significantly higher during summer when compared to winter months (P < 0.01), but both these values were not significantly different from the monsoon season. Although the increase seen in erythrocyte counts in summer was not significantly different from the other seasons, PCV was significantly elevated during summer as compared to monsoon (P < 0.01) and winter (P < 0.01). No significant influences of age or body weight on the different hematologic parameters were detected within the adult and sub-adult bear populations.

In this study, we have generated hematology reference values for the sloth bear within its native habitat. We have also examined the effect of age, sex, season, and two geographical locations on these parameters. The significant differences seen in sloth bear cubs in comparison to adult bears, as well as the sexually dimorphic differences, emphasize the clinical importance of using age- and sex-associated reference values. The large sample size for each parameter measured in this study minimized the influence of unavoidable variables such as capture and handling stress (Cattet et al., 2003b; Kusak et al., 2005). Finally, altitude, climatic conditions,

habitat quality, and the nutritional status of animals (Hellgren et al., 1993; Gau and Case, 1999) most likely contributed to some of the differences recorded between sloth bears located at the Bannerghatta and Sur Sarovar locations.

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