North American Crane Working Group

PROCEEDINGS OF THE NORTH AMERICAN CRANE WORKSHOP

2016

SERUM CHEMISTRY, BLOOD GAS, AND PHYSIOLOGICAL MEASURES OF SANDHILL CRANES SEDATED WITH ALPHA-CHLORALOSE

Richard R. Sim
Barry K. Hartup

Sim, R. R., and B. K. Hartup. 2016. Serum chemistry, blood gas, and physiological measures of sandhill cranes sedated with alpha-chloralose. Proceedings of the North American Crane Workshop 13:107-110.

SERUM CHEMISTRY, BLOOD GAS, AND PHYSIOLOGICAL MEASURES OF SANDHILL CRANES SEDATED WITH ALPHA-CHLORALOSE

RICHARD R. SIM, School of Veterinary Medicine, University of Wisconsin, 2015 Linden Drive, Madison, WI 53706 USA
BARRY K. HARTUP, International Crane Foundation, E-11376 Shady Lane Road, Baraboo, WI 53913, USA

Capture techniques that lessen handling stress may also lessen pathologic influences on physiologic measures, improving the validity of these measures for use in individual health assessment of free-ranging wildlife. Since 1990, the International Crane Foundation (ICF) has successfully used chemical immobilization with alpha-chloralose (AC; $C_6H_{11}Cl_3O_6$), a chloral derivative of glucose, to facilitate captures of sandhill cranes (*Grus canadensis tabida*) for ecological studies (Hayes et al. 2003). Although this chemical has been used orally for the immobilization of many species, the physiologic effects of AC are not well understood in cranes. The primary purpose of this study was to measure serum chemistry, venous blood gas, and physiological values in free-ranging sandhill cranes successfully immobilized using this technique.

Sandhill cranes were captured near Briggsville, Wisconsin (43°36′N, 89°36′W), between 1996 and 1999 as previously described (Langenberg et al. 1998, Hayes et al. 2003). Once at a nearby holding pen, each crane was banded, weighed, measured and blood samples were taken from the left or right medial metatarsal or right jugular veins. Starting in 1997, the cloacal temperature and minute heart and respiratory rates of each crane were determined prior to blood collection. Sampling occurred approximately 1 hour post-capture. Samples were analyzed for the concentrations of selected serum enzymes and electrolytes (glucose, aspartate aminotransferase [AST], alanine aminotransferase [ALT], alkaline phosphatase [AP], creatine kinase [CK], lactic dehydrogenase [LDH], cholesterol, total protein [TP], phosphorous [P], calcium [Ca], sodium [Na], potassium [K], chloride [Cl], bicarbonate [HCO_3], uric acid, and anion gap) by Marshfield Laboratories (Marshfield, WI). Blood samples were also analyzed for venous blood gas levels by using an i-STAT Portable Clinical Analyzer (i-STAT PCA; Sensor Devices Incorporated, Waukesha, WI). The EG7+ cartridges used with the i-STAT PCA provided tests for hydrogen ion concentration (pH), carbon dioxide tension (pCO_2), oxygen tension (pO_2), Na, K, ionized calcium (iCa), and hematocrit (PCV) measurement; in addition, the i-STAT provided HCO_3, total carbon dioxide (tCO_2), base excess (BE), oxygen saturation (sO_2), and hemoglobin (Hb) as calculated values based on the measurements. The i-STAT PCA has been validated for use in chickens for all analytes except K and BE (Steinmetz et al. 2007).

The mean and standard deviation (SD) for each serum chemistry, blood gas, and physiologic value were calculated for hatch-year juvenile and adult sandhill cranes. Age was determined through field markings (Lewis 1979). Sex of each bird was determined through relative size (Nesbitt et al. 1992), behavior (Archibald 1976), or through genetic analysis of blood samples (Griffiths et al. 1998). Individual cranes that were clinically abnormal at the time of sampling or had values that differed from the mean by more than 3 SD were omitted from the analyses. Reference ranges for all parameters represent the mean ± 2 SD. Results have been pooled by sex due to sampling of 8 unknown sex juveniles and a preliminary analysis that showed no sex differences among adults.

Samples were obtained from 23 juveniles (6 males, 9 females, 8 unknown; mean age = 135 days, range 118-175 days; estimated hatch date of May 1), and 47 adults (21 males, 26 females). Serum chemistry findings are based on data from 19 juveniles and 37 adults (Table 1). Venous blood gas analysis using the i-STAT PCA, as well as summary physiological measures, reflect data from 14 juveniles and 35 adults (Table 2).

Our results for electrolytes, cholesterol, TP, P, and Ca are similar to those of Ellis et al. (1996), and reference ranges for these values and glucose and uric acid are similar to those found by Olsen et al. (2001). Alkaline phosphatase levels were increased in juveniles compared to adults, and likely due to ongoing skeletal development described similarly by Olsen et al. (2001). Reference ranges for AST were similar in both age groups of this study, but higher than those reported by Olsen et al. (2001). Abnormal AST activities have been linked to vitamin E, selenium or methionine deficiencies, liver damage, pesticide and carbon tetrachloride intoxication and muscle damage (Ritchie et al. 1994). CK results produced very wide reference ranges for

Table 1. Laboratory serum chemistry results from adult and hatch-year greater sandhill cranes sedated with alpha-chloralose, near Briggsville, Wisconsin, 1996-1999.

	Adults					Hatch-year				
	Mean (*n* = 37)	SE	Min.	Max.	Reference range	Mean (*n* = 19)	SE	Min.	Max.	Reference range
Glucose (mg/dL)	237	5	172	312	174.6-299.5	257	6	210	308	206.5-307.8
AST (U/L)	269	11	158	466	139.7-398.1	294	15	182	423	160.2-426.8
ALT (U/L)	35	2.2	11	60	7.9-61.1	33	4.1	5	65	0-68.3
AP (U/L)	147	18	13	605	0-371.4	383	49	190	963	0-813
CK (U/L)	1440	203	84	5425	0-3914.7	1611	297	162	4689	0-4203.8
LDH (U/L)	370	26	202	853	55.1-684.6	456	44	171	931	76.43-835.6
Cholesterol (mg/dL)	142	4	93	195	93.0-190.9	142	7	65	199	79.4-205.2
TP (g/dL)	3.3	0.1	2.5	4.4	2.5-4.1	3.2	0.1	2.7	3.8	2.6-3.8
P (mg/dL)	1.9	0.1	1.0	4.7	0.3-3.6	4.9	0.3	3.3	7.1	2.6-7.3
Ca (mg/dL)	9.3	0.1	8.7	10	8.4-10	9.3	0.2	7.2	11	7.5-11
Na (mmol/L)	142	1	136	151	136-147	141	1	138	151	135.2-147.3
K (mmol/L)	3.9	0.1	2.1	6.8	2.2-5.6	4.3	0.2	2.4	7.1	2.4-6.3
Cl (mmol/L)	104	0.4	100	111	98.7-109	103	0.5	97	107	98.7-108
HCO_3 (mmol/L)	29	0.5	24	35	23-35.4	27	0.7	23	32	21-33.8
Uric Acid (mg/dL)	4.4	0.2	2.4	7.4	1.7-7.2	4.7	0.3	2.7	6.5	2.2-7.1
Anion Gap (mmol/L)	12	0.5	6	18	5.6-18.8	15	0.6	11	19	9.2-20.3

Table 2. Physiological and i-STAT PCA venous blood gas results from adult and hatch-year greater sandhill cranes sedated with alpha-chloralose, near Briggsville, Wisconsin, 1997-1999.

	Adults					Hatch-year				
	Mean (*n* = 35)	SE	Min.	Max.	Reference range	Mean (*n* = 14)	SE	Min.	Max.	Reference range
Cloacal temp (°C)	40.1	0.1	38.7	41.8	38.7-41.6	39.7[a]	0.2	38.7	41.3	38.2-41.3
Heart rate	142[b]	6	84	200	76-208	164[a]	18	80	280	34-294
Respiration rate	30	2	12	70	3-58	29[a]	2.6	18	52	9.6-48
pH	7.43[c]	0.02	7.23	7.52	7.29-7.56	7.40	0.03	7.24	7.48	7.26-7.54
pCO_2 mmHg)	39.6[c]	1.5	30.6	66.0	22.7-56.4	45.5	3.2	31.6	69.3	21.9-69.1
pO_2 (mmHg)	61	1.7	38	78	41-81	58.6	2.2	42	71	41.7-75.4
Na (mmol/L)	140	0.3	135	146	136-144	140	0.6	136	143	136-144
K (mmol/L)	4.2	0.1	3.4	5.1	3.3-5.1	4.3	0.1	3.8	4.8	3.6-4.9
iCa (mmol/L)	1.15	0.02	0.98	1.28	1.00-1.30	1.24	0.03	1.13	1.34	1.11-1.38
PCV (%)	38	0.6	32	48	31-46	34.8	1.0	29	42	27.4-42.2
Hb (g/dL)	13	0.2	11	16	11-16	12	0.3	10	14	9.5-14
BE (mmol/L)	1[c]	0.4	−2	6	−3-5	2.6	0.8	−1	11	−6.7-8.9
HCO_3 (mmol/L)	26[c]	0.4	21	30	21-30	27.3	0.9	23	36	20.4-34.2
tCO_2 (mmol/L)	27[c]	0.4	22	31	22-31	28.8	1.0	24	38	21.5-36.1
sO_2 (%)	90[c]	1.3	66	96	75-100	88.1	2.1	67	95	72.6-100

[a] *n* = 13; physiological measures not recorded for 1 hatch-year crane.
[b] *n* = 34; heart rate was not recorded for 1 adult crane.
[c] *n* = 32; smaller *n* for these parameters was due to i-STAT PCA system failures of single or multiple values for some samples.

both age groups and were much higher than results reported from captive populations. In healthy turkeys, CK has been reported to be very sensitive to stress and exercise (Lumeij 1987). Muscle damage, neuropathies, vitamin E or selenium deficiencies, and lead toxicity can be possible causes of CK increases. LDH is another enzyme that can be elevated with hemolysis, hepatic necrosis, and muscle damage, but its non-specificity limits its diagnostic value in birds. The hatch-year cranes' LDH values in this study were greater than adult birds and greater than all age groups reported by Olsen et al. (2001). We believe the variously elevated values of AST, CK, and LDH were attributable to muscular exertion experienced during the induction phase or initial sedative effects and 1-hour lag in blood sampling from alpha-chloralose capture. The mild elevations that we observed were subclinical and a byproduct of the capture technique. These changes are distinct from values determined using captive cranes that experience minimal exertion prior to blood sampling following physical capture and restraint. As shown elsewhere, exertional myopathy is a significant risk factor with the use of AC, yet morbidity and mortality rates with this method are comparable to or less than most other contemporary alternative capture techniques (Hartup et al. 2014).

Heart and respiration rates for sedated greater sandhill cranes are previously unpublished. The heart rate reference range for hatch-year birds has a higher upper limit than for adults. The reference range established in this study for cloacal temperature is broader than that reported by Ellis et al. (1996), but this would be expected given possible exertion and stress experienced by free-ranging birds with varied levels of sedation (Hayes et al. 2003).

With the exception of Na, K, PCV, and Hb, the venous blood gas parameters measured using the i-STAT PCA are previously unpublished for this species. Langenberg et al. (1998) reported preliminary results from a small number of cranes, but provided additional information on these parameters up to 8 hours following immobilization. Olsen's (2001) reports for PCV and Hb in captive greater sandhill cranes are consistent with those of this study, but Ellis et al. (1996) reported higher means for Na. Potassium and BE were found to be unreliably measured by the i-STAT PCA when compared to laboratory assays (Steinmetz et al. 2007). All the venous blood gas parameters are comparable between adult and hatch-year cranes with a high degree of overlap, except for iCa that was higher in the younger birds. Bone remodeling associated with normal juvenile growth may have been responsible for this difference.

Cranes in this study were clinically asymptomatic and the omission of individuals with any outlier values presumably selected for a healthy reference pool, but underlying disease states, like parasitic infection, were still probable. Other effects from age, sex, reproductive status, nutrition, disease, and environment can affect each bird's health status as reflected through its serum chemistry and physiology. Our findings extend the normative physiological reference data available for free-ranging sandhill cranes.

ACKNOWLEDGMENTS

We thank N. Businga and J. Langenberg for collection of serum samples and ICF Department of Field Ecology staff for safe capture of cranes. This study was supported by a grant from the Companion Animal Fund, School of Veterinary Medicine, University of Wisconsin-Madison.

LITERATURE CITED

Archibald, G. W. 1976. The unison call of cranes as a useful taxonomic tool. Ph.D. Dissertation, Cornell University, Ithaca, New York, USA.

Ellis. D. H., G. F. Gee, and C.M. Mirande. 1996. Cranes: Their biology, husbandry, and conservation. U.S. Department of Interior, National Biological Service, Washington, D.C., and International Crane Foundation, Baraboo, Wisconsin, USA.

Griffiths, R., M. C. Double, K. Orr, and R. J. G. Dawson. 1998. A DNA test to sex most birds. Molecular Ecology 7:1071-1075.

Hartup, B. K., L. Schneider, J. M. Engels, M. A. Hayes, and J. A. Barzen. 2014. Capture of sandhill cranes (*Grus canadensis tabida)* using alpha-chloralose: a 10-year follow-up. Journal of Wildlife Diseases 50:143-145.

Hayes, M. A., B. K. Hartup, J. M. Pittman, J. A. Barzen. 2003. Capture of sandhill cranes using alpha-chloralose. Journal of Wildlife Diseases 39:859–868.

Langenberg J. A., N. K. Businga, H. E. Nevill. 1998. Capture of wild sandhill cranes with alpha-chloralose: techniques and physiologic effects. Pages 50-53 *in* Proceedings of the AAZV and AAWV Joint Conference, American Association of Zoo Veterinarians, 17–22 October, Omaha, Nebraska, USA.

Lewis, J. C. 1979. Field identification of juvenile sandhill cranes. Journal of Wildlife Management 43:211-214.

Lumeij T. J. 1987. A contribution to clinical investigative methods for birds, with special reference to the racing pigeon (*Columba livia domestica*). Thesis, Utrecht University, Netherlands.

Nesbitt, S. A., C. T. Moore, and K. S. Williams. 1992. Gender prediction from body measurements of two subspecies of sandhill cranes. Proceedings of the North American Crane Workshop 6:38-42.

Olsen, G. H., M. M. Hendricks, and L. E. Dressler. 2001. Hematological and serum chemistry norms for sandhill and whooping cranes. Proceedings of the North American

Crane Workshop 8:178-184.

Ritchie, B. W., G. J. Harrison, and L. R. Harrison. 1994. Avian medicine: principles and application. Wingers Publishing, Lake Worth, Florida, USA.

Steinmetz, H. W., R. Vogt, S. Kastner, B. Riond, and J. M. Hatt. 2007. Evaluation of the i-STAT portable clinical analyzer in chickens (*Gallus gallus*). Journal of Veterinary Diagnostic Investigation 19:382-388.

PROCEEDINGS OF THE NORTH AMERICAN CRANE WORKSHOP 13:107-110

Key words: alpha-chloralose, blood gas, capture, clinical pathology, cranes, *Grus canadensis tabida*, physiology, serum chemistry.