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Hematology and Blood Chemistry Values in Free-Living Imperial Cormorants (*Phalacrocorax atriceps*)

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SUMMARY. As part of an on-going, long-term study on the reproductive ecology and health status of imperial cormorants (*Phalacrocorax atriceps*), blood samples were collected to establish baseline values for hematologic parameters (hematocrit, red and white blood cell counts, leukocyte profile, heterophil:lymphocyte ratio, total solids) and serum chemistries (glucose, uric acid, urea, total protein, triglycerides, cholesterol, albumin:globulin ratio, alkaline phosphatase, lactate dehydrogenase, creatine phosphokinase, aspartate aminotransferase, alanine aminotransferase, calcium, phosphorus). One hundred and eighty-four male adults from the Punta León breeding colony in Patagonia Argentina were captured during the chick-rearing period of four breeding seasons, 2004 ($n = 48$), 2005 ($n = 29$), 2010 ($n = 43$), and 2011 ($n = 64$). All birds appeared to be in good body condition and no abnormalities were noted during physical examination. In general, values for the parameters reported in this study were similar to those previously described for other cormorant species. Significant interannual differences were observed in most health parameters analyzed. This study defines baseline health parameters for imperial cormorants and, coupled with previous reports on pathogen exposure, contributes to our knowledge of the overall health status of the species.

RESUMEN. Hematología y bioquímica plasmática de cormoranes imperiales (*Phalacrocorax atriceps*).

Como parte de un estudio a largo plazo sobre la ecología reproductiva y el estado sanitario del cormorán imperial (*Phalacrocorax atriceps*), se recolectaron muestras de sangre para establecer los valores de referencia de parámetros hematológicos (hematocrito, conteo total de eritrocitos y leucocitos, perfil leucocitario, proporción heterófilos:linfocitos, sólidos totales) y bioquímicas plasmáticas (glucosa, ácido úrico, urea, proteínas totales, triglicéridos, colesterol, proporción albúmina:globulina, fosfatasa alcalina, lactato deshidrogenasa, creatina fosfoquinasa, aspartato aminotransferasa, alanina aminotransferasa, calcio, fósforo). Se capturaron 184 machos adultos en la colonia reproductiva de Punta León, Patagonia Argentina, durante el período de crianza de pichones de cuatro temporadas reproductivas, 2004 ($n = 48$), 2005 ($n = 29$), 2010 ($n = 43$) y 2011 ($n = 64$). Aparentemente, todas las aves se encontraron en buena condición corporal y no se observaron anormalidades durante el examen físico. En general, los valores de los parámetros reportados en este estudio resultaron similares a aquellos registrados para otras especies de cormoranes. Se observó una variación inter-anual significativa en la mayoría de los parámetros sanitarios analizados. Este estudio presenta valores basales de diferentes parámetros del estado de salud para cormoranes imperiales, y junto con estudios previos sobre exposición a patógenos, contribuye al conocimiento del estado sanitario de la especie.

Key words: cormorants, hematology, Patagonia Argentina, *Phalacrocorax atriceps*, serum chemistry

Abbreviations: ALP = alkaline phosphatase; ALT = alanine aminotransferase; AST = aspartate aminotransferase; Ca = calcium; CPK = creatine phosphokinase; GLM = generalized linear model; GLMM = generalized linear mixed model; IC = imperial cormorant; ID = male individual identification number; LDH = lactate dehydrogenase; LRT = likelihood ratio test; P = phosphorus; RBC = red blood cell count; WBC = white blood cell count.

Hematology and plasma biochemistry values are useful in assessing individual and population health and fitness for wildlife species (8,13). These parameters may provide a more-sensitive indication of population condition than morphologic data alone (38). Furthermore, they are useful diagnostic tools in clinical practice (8,9,24) and are especially important in birds, which frequently show few overt clinical signs of disease (24). Accurate reference data are also essential for assessing population health; however, its determination can be difficult in wild species due to the inherent variability of natural systems (34,37).

The imperial cormorant (IC, *Phalacrocorax atriceps*) is a colonial seabird widely distributed along the Patagonian coast in Argentina, with some colonies located near urban centers (18,56). Changes due to increased human activity in this area, including fisheries (22,61), tourism (57), and domestic refuse at city dumps, may affect the health of the coastal Patagonian ecosystem and possibly the populations of seabirds nesting there (55). Documenting hematology and serum chemistry values will provide a baseline for comparisons that will allow researchers to monitor the effects of habitat changes through time and will provide a reference for comparison with other wild cormorants.

There are some reports of hematology and biochemistry parameters in wild cormorant species (31,34,49); however, no such baseline data have been established for the IC. Additionally, the reported values to date were obtained from small sample sizes (31,34) or from a combination of sexes and age classes (34,49) with consequent fallible results.

As part of an on-going, long-term study on the reproductive ecology and health status of IC, we collected blood samples from adult males breeding in one of the largest colonies in Patagonia, Argentina during four breeding seasons. Here, we established baseline values for hematologic and serum chemistry parameters based in a sizable sample and standardized data from one class of age and sex.

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Table 1. Hematologic values for adult male imperial cormorants (*Phalacrocorax atriceps*) in Patagonia, Argentina during two breeding seasons (2010 and 2011).

Parameter (units)	Mean	Median	SD	2.5 percentile	97.5 percentile	Range	*n*
Hematocrit (%)[A]	49.93	50.00	2.43	44.00	54.00	44.00–54.00	105
Red blood cells (cells/μl × 10^6)	3.73	3.67	1.15	1.68	5.74	0.40–6.33	96
White blood cells (cells/μl × 10^3)	12.08	12.16	4.95	2.96	22.36	2.38–24.01	92
Differentials							
Heterophils (cells/μl × 10^3)	4.64	4.80	1.09	2.20	6.50	1.20–7.10	106
Lymphocytes (cells/μl × 10^3)[A]	4.16	4.00	1.19	2.30	6.40	1.80–7.20	106
Monocytes (cells/μl × 10^3)[A]	0.32	0.30	0.24	0.00	0.80	0.00–1.00	106
Eosinophils (cells/μl × 10^3)[A]	0.83	0.70	0.72	0.00	2.50	0.00–4.30	106
Basophils (cells/μl × 10^3)[A]	0.004	0.00	0.02	0.00	0.10	0.00–0.10	106
Heterophil:lymphocyte ratio[A]	1.20	1.15	0.65	0.22	2.74	0.11–3.40	106
Total solids (g/dl)[A]	3.82	3.90	0.38	3.00	4.50	3.00–4.90	103

[A]Assumption of normality was not met for these parameters (Kruskal-Wallis test $P < 0.05$).

MATERIALS AND METHODS

Study area. Sampling was conducted at Punta León (43°05′S, 64°30′W), Chubut, Argentina, where ~3200–3400 pairs of IC breed annually (47). Five species share this breeding site with the IC: rock shag (*Phalacrocorax magellanicus*), neotropic cormorant (*Phalacrocorax brasilianus*), kelp gull (*Larus dominicanus*), cayenne tern (*Sterna eurygnatha*), and royal tern (*Sterna maxima*). The spatial distribution of species, the sensitivity to human visitors (58), and the proximity of this colony to urban centers make Punta León very vulnerable to human disturbance (60). This zone has been proposed as a Tourist Nature Reserve (Unidad de Investigación Biológica; Ley 2580/85) and was recently designated as an Important Bird Area for Conservation (59).

Sample collection and storage. Sampling was conducted during the chick-rearing periods (late November) of four breeding seasons (2004, 2005, 2010, and 2011). A total of 184 male adult IC were captured at their nests before foraging trips (48, 29, 43, and 64 during the 2004, 2005, 2010, and 2011 seasons, respectively). Males were identified by their morphometric characteristics (males are larger than females) and vocalizations during nest defense (males "honk" and females "hiss;" see 46). Adults were identified by plumage coloration and development of caruncles (43). Handling procedures included body weight, notation of visual or palpable abnormalities, and blood collection. Twenty-four of the IC in this study were banded as part of a broader study on the behavioral ecology of the IC and were sampled repeatedly during two or more breeding seasons (recaptures).

Blood (<1% of body mass) was drawn by venipuncture of the jugular vein using 10 cc heparinized syringes and 22 or 23 G × 1" needles. All samples were stored in plain glass vacuum tubes (Becton-Dickinson, Rutherford, NJ) and kept cool on ice until processing within 4–6 hr postcollection. After blood collection, body weights were measured using spring scales (Pesola®, Baar, Switzerland) appropriate to the weight of the adult.

Sample processing and analysis. Hematologic analyzes were performed only on samples from the 2010 and 2011 breeding seasons. Once at the field laboratory, thin blood smears were prepared from heparinized blood and fixed with 99% methanol. Microhematocrit tubes were centrifuged in a portable 12-volt centrifuge (1000 × *g*, Mobilespin, Vulcan Technologies, Grandview, MO) for hematocrit determination. Plasma total solids were measured using a hand-held refractometer (Schulco, Toledo, OH) calibrated at the site. White blood cell (WBC) and red blood cell (RBC) counts were performed by the same individual (L. Gallo) using the Leuko-tics® and Ery-tics® systems (Bioanalytic GmbH, Freiburg, Germany), respectively and a Neubauer hemocytometer (Assistant Germany, Karl Hecht AG, Rhöne, Germany). The remaining blood was centrifuged at 1000 × *g* for 20 min and plasma was removed and stored at −80 C until blood chemistry analyses were performed. Blood smears were stained with Tinción 15 (Biopur SRL, Rosario, Argentina) to perform WBC differential counts. Total WBC counts were determined using the methodology described by Dein *et al.* (14).

Plasma chemistries and enzymes were analyzed for samples of all seasons which were processed on a wet automated analyzer (Hitachi Model 902 Automatic Analyzer, Hitachi Science Systems, Ibaraki, Japan) at a commercial veterinary laboratory (Laboratorio de Análisis Clínicos, Chacabuco, Buenos Aires, Argentina). Parameters measured included glucose, uric acid, urea, cholesterol, triglycerides, total protein, albumin:globulin ratio, calcium (Ca), and phosphorus (P) concentrations and alkaline phosphatase (ALP), lactate dehydrogenase (LDH), creatine phosphokinase (CPK), alanine aminotransferase (ALT), aspartate aminotransferase (AST) activities.

Statistical analyses. Before statistical analysis, we excluded outlying values due to bad sample quality, for example as a result of hemolysis (25,32). We pooled data across breeding seasons, and basic statistics including mean, median, range (lowest and highest value), and SD were obtained for each parameter. A Shapiro-Wilk test was performed to evaluate the distribution of each parameter (29). Because most variables did not meet the assumption of normality, the central 95% interval (2.5 and 97.5 percentiles) (44) is also reported.

To explore differences in body weights and blood variables by breeding season we employed a general linear mixed model (GLMM) (36,62) considering the nonindependence of samples of the same individual between seasons (recaptures). Our statistical analyses included body weight, hematologic and biochemical parameters as response variables, breeding season as fixed factor, and the male individual identification number (ID) as the random effect. When models showed signs of nonnormality of residuals, response variables were transformed using natural logarithm (ln) transformations. We evaluated the significance of random effect with a likelihood-ratio test (LRT) (36). In cases where it was significant, we assessed the percentage of variance attributable to the random effect ID (differences between males) and, in those cases in which random effect was not significant (ID effect: $P > 0.05$), we analyzed interannual differences using general linear models (GLM) with fixed effect (12). For statistical analyses we used the NLME package from R software, version 2.12.1 (40). Statistical significance was established at $P < 0.05$.

RESULTS

Physical examination and body weights. All birds ($n = 184$) appeared to be in good condition and no abnormalities or evidence of disease were noted during physical examination. Body weights ranged between 1.9 and 2.7 kg (mean = 2.3, SD = 0.14, $n = 184$) and varied significantly between breeding seasons (F-test = 3.76, $P = 0.02$). Percentage of variance attributable to differences between

Table 2. Biochemical blood values for adult male imperial cormorants (*Phalacrocorax atriceps*) in Patagonia, Argentina, during four breeding seasons (2004, 2005, 2010, and 2011).

Parameter (units)	Mean	Median	SD	2.5 Percentile	97.5 Percentile	Range	*n*
Glucose (mg/dl)[A]	193.78	194.00	27.51	147.00	254.00	110.00–300.00	163
Uric acid (mg/dl)[A]	4.55	7.39	1.15	2.81	7.20	2.43–10.10	155
Urea (mg/dl)[A]	7.77	7.00	3.12	4.00	16.00	1.00–24.00	168
Total protein (g/dl)[A]	3.84	3.80	0.50	3.10	4.80	1.90–5.70	165
Triglyceride (mg/dl)[A]	99.63	96.00	30.23	54.00	182.00	37.00–207.00	175
Cholesterol (mg/dl)	336.61	340.00	64.46	224.00	459.00	155.00–591.00	165
Albumin:globulin ratio[A]	1.10	1.10	0.19	0.80	1.50	0.40–1.70	163
Alkaline phosphatase (IU/L)[A]	2126.98	1993.50	866.39	803.00	3955.00	130.00–4886.00	162
Lactate dehydrogenase (IU/L)[A]	666.25	640.00	214.53	327.00	1177.00	306.00–1259.00	161
Creatine phosphokinase (IU/L)[A]	762.30	707.00	297.64	339.00	1344.00	276.00–1973.00	165
Aspartate aminotransferase (IU/L)[A]	285.28	278.00	57.13	198.00	409.00	108.00–530.00	161
Alanine aminotransferase (IU/L)	88.92	88.50	20.04	26.00	132.50	26.00–151.00	160
Calcium (mg/dl)[A]	9.34	9.30	0.94	7.50	11.00	6.50–14.10	165
Phosphorus (mg/dl)	4.25	4.30	0.72	2.60	5.80	2.40–6.10	159

[A]Assumption of normality was not met for these parameters (Kruskal-Wallis test $P < 0.05$).

males was large (55.48%; ID effect: LRT = 12.96, $P = 0.001$), reflecting large interindividual variation for body weight.

Hematology and plasma biochemistries. Hematology and biochemistry values of adult male imperial cormorants are presented in Tables 1 and 2, respectively. For birds recaptured at least twice ($n = 24$), variation explained by the birds' ID was largest for hematocrit (42.84%; ID effect: LRT = 4.47, $P = 0.02$), monocytes (42.24%; ID effect: LRT = 4.01, $P = 0.02$) and ALP (81.42%; ID effect: LRT = 20.30, $P < 0.001$), reflecting considerable interindividual variation. Additionally, breeding season had a significant effect on most of the hematologic and biochemical parameters evaluated (Table 3).

Table 3. Effect of breeding season on hematologic and biochemical parameters of the imperial cormorant (*Phalacrocorax atriceps*). Statistically significant ($P < 0.05$) differences are marked with an asterisk (*).

	Fixed effect (year)	
Parameter (response variable)[A]	*F*-test[B]	*P*
Hematocrit (%)	1.46	0.25
Red blood cells (cells/μl × 10^6)	31.25	<0.001*
White blood cells (cells/μl × 10^3)	40.43	<0.001*
Heterophils (cells/μl × 10^3)	2.07	0.15
Lymphocytes (cells/μl × 10^3)	39.37	<0.001*
Monocytes (cells/μl × 10^3)	4.64	0.049*
ln Eosinophils (cells/μl × 10^3)	116.00	<0.001*
Heterophil:lymphocyte ratio	5.24	0.02*
Total solids (g/dl)	7.69	0.01*
Glucose (mg/dl)	15.45	<0.001*
ln Uric acid (mg/dl)	17.87	<0.001*
Urea (mg/dl)	2.00	0.12
Total protein (g/dl)	16.94	<0.001*
Triglyceride (mg/dl)	3.67	0.01*
Cholesterol (mg/dl)	2.77	0.04*
Albumin:globulin ratio	40.65	<0.001*
Alkaline phosphatase (IU/L)	2.61	0.09
Lactate dehydrogenase (IU/L)	17.10	<0.001*
Creatine phosphokinase (IU/L)	3.24	0.02*
Aspartate aminotransferase (IU/L)	2.64	0.05
Alanine aminotransferase (IU/L)	2.25	0.08
Calcium (mg/dl)	25.99	<0.001*
Phosphorus (mg/dl)	11.50	<0.001*

[A]Response variables were transformed using natural logarithm (ln) in those models in which the distribution of residuals was nonnormal.

[B]*F*-test is the statistic test from which significant levels (*P* values) for general linear models (GLM) and general linear mixed models (GLMM) are calculated. In GLMM, male individual identification number (ID) was included as a random effect (only significant for hematocrit, monocytes, and alkaline phosphatase).

DISCUSSION

This is the first study of baseline plasma biochemistry and hematology information for IC. All animals sampled in this study were in good physical condition. The body weights of male ICs were similar to those previously reported for this species in other studies conducted at this same colony (46), with significant variability between individuals and breeding seasons.

Overall, most hematology and blood chemistry values reported in this study were similar to those previously described for other wild and captive cormorant species (Table 4) (5,26,31,34,49). Notwithstanding, comparisons with captive birds should be interpreted with caution given that difference in diets, food availability, and physical activity can affect blood parameters (1,11,15, and references therein).

Hematology. Hematocrit values were similar to those reported for free-ranging flightless cormorants (*Phalacrocorax harrisi*) (49) but higher than findings in free-ranging pelagic cormorants (*Phalacrocorax pelagicus*) (34), black-faced cormorants (*Phalacrocorax fuscescens*) (31) and in captive double-crested cormorants (*Phalacrocorax auritus*) (26) and great cormorants (*Phalacrocorax carbo*) (5). These differences are likely not clinically important and may be attributable to sample size, age variations in sampled birds (31), capture method (34), and reproductive stage (5). Also, because individuals in our study were sampled before foraging, which implies about 8 hr of fasting, slightly higher hematocrits may be due to dehydration (7,54).

Total RBC counts were higher than those values published for other captive and free-ranging cormorant species (5,26,31). The variability in RBC counts was not surprising as this parameter has been reported to be highly inconsistent (1.5×10^6 to 6.6×10^6 cells/μl) among different bird species (3) and can also be affected by season, time of day, and environmental temperature (7).

Hematocrit and RBC counts reflect blood viscosity and oxygen carrying capacity. Therefore, variations in these values between cormorant species may be explained by their differential diving

Table 4. Comparison of reported hematologic and biochemical values for captive and free-ranging cormorants. Means and standard deviations are listed first, except for nonnormally distributed parameters where medians are given. Ranges (lowest to highest value) are to the right except for black-faced cormorant, pelagic cormorant, and great cormorant for which they are not available.

	Species[B]								
Parameter[A] (units)	Imperial cormorant (n = 92–175)		Double-crested cormorant (n = 4–17)		Flightless cormorant (n = 21–60)		Black-faced cormorant (n = 5)	Pelagic cormorant (n = 3–5)	Great cormorant (n = 7)
Hct (%)	50.00	44.00–54.00	45.10 ± 4.90	36.00–52.00	49.20 ± 1.30[C]	35.00–58.00	43.40 ± 2.00	30.00 ± 12.00	43.50 ± 2.40
RBC (cells/μl × 10^6)	3.73 ± 1.15	0.40–6.33	2.39 ± 0.42	1.80–3.23	–		2.16 ± 0.08	–	2.90 ± 0.19
WBC (cells/μl × 10^3)	12.08 ± 4.95	2.38–24.01	9.01 ± 3.79	4.20–18.40	8.00 ± 2.80	2.80–14.80	9.1	–	–
H (cells/μl × 10^3)	4.64 ± 1.09	1.20–7.10	5.26 ± 1.72	2.76–8.50	3.66 ± 1.58	0.50–7.10	3.30	6.10 ± 1.30	–
L (cells/μl × 10^3)	4.00	1.80–7.20	2.94 ± 2.98	0.14–8.71	4.00 ± 1.61	0.80–7.60	4.90	2.70 ± 0.50	–
M (cells/μl × 10^3)	0.30	0.00–1.00	0.29 ± 0.24	0.06–0.88	0.22 ± 0.20	0.00–1.20	1.20	0.20 ± 0.40	–
E (cells/μl × 10^3)	0.70	0.00–4.30	0.72 ± 0.83	0.06–2.21	2.08 ± 1.05	0.30–5.20	0.40	0.80 ± 0.20	–
B (cells/μl × 10^3)	0.00	0.00–0.10	0.32 ± 0.27	0.10–0.76	0.05 ± 0.09	0.00–0.30	<0.01	0.20 ± 0.30	–
TS (g/dl)	3.90	3.00–4.90	–	–	–	–	–	4.70 ± 1.60	–
Glu (mg/dl)	194.00	110.00–300.00	259.00 ± 32.00	201.00–306.00	131.53 ± 37.84	64.04–198.20	–	224.00 ± 23.00	238.20 ± 54.60
UA (mg/dl)	7.39	2.43–10.10	5.50 ± 2.00	2.40–9.70	22.04 ± 10.17	6.78–40.68	–	30.00 ± 6.00	–
U (mg/dl)	7.00	1.00–24.00	–	–	–	–	10.81 ± 1.68	–	8.90 ± 2.20
TP (g/dl)	3.80	1.90–5.70	4.1 ± 0.5	3.2–4.9	3.92 ± 0.52	3.10–5.10	3.90 ± 0.90	3.60 ± 0.50	3.12 ± 0.23
Tri (mg/dl)	96.00	37.00–207.00	65.00 ± 39.00	42.00–123.00	–	–	–	228.00 ± 177.00	–
Cho (mg/dl)	336.61 ± 64.46	155.00–591.00	301.00 ± 64.00	206.00–343.00	259.09 ± 34.80	185.61–348.03	–	272.00 ± 72.00	–
ALP (IU/L)	1993.50	130.00–4886.00	2579.00 ± 830.00	743.00–3601.00	–	–	–	1337.00 ± 540.00	–
LDH (IU/L)	640.00	306.00–1259.00	876.00 ± 513.00	347.00–2252.00	–	–	–	1345.00 ± 218.00	–
CPK (IU/L)	707.00	276.00–1973.00	1834.00 ± 1307.00	794.00–5277.00	2076.8 ± 840.60	764.00–3636.00	–	3799.00 ± 3956.00	–
AST (IU/L)	278.00	108.00–530.00	489.00 ± 162.00	275.00–838.00	388.60 ± 131.10	150.00–653.00	–	587.00 ± 339.00	–
ALT (IU/L)	88.92 ± 20.04	26.00–151.00	144.00 ± 24.00	120.00–170.00	–	–	–	214.00 ± 73.00	–
Ca (mg/dl)	9.30	6.50–14.10	10.10 ± 1.00	8.40–11.70	9.20 ± 1.2	6.00–10.80	10.42 ± 0.92	20.20 ± 3.80	–
P (mg/dl)	4.25 ± 0.72	2.40–6.10	3.30 ± 1.10	0.60–4.40	14.55 ± 7.12	2.79–25.39	8.05 ± 0.93	–	–

[A]Hct = hematocrit; RBC = red blood cell count; WBC = white blood cell count; H = heterophil; L = lymphocyte; M = monocyte; E = eosinophil; B = basophil; H:L ratio = heterophil:lymphocyte ratio; TS = total solids; Glu = glucose; UA = uric acid; U = urea; TP = total proteins; Tri = triglycerides; Cho = cholesterol; A:G ratio = albumin:globulin ratio; ALP = alkaline phosphatase; LDH = lactate dehydrogenase; CPK = creatine phosphokinase; AST = aspartate aminotransferase; ALT = alanine aminotransferase; Ca = calcium; P = phosphorus.

[B]Imperial cormorant (*Phalacrocorax atriceps*) = wild adult males during prefledging period (this study); double-crested cormorant (*Phalacrocorax auritus*) = captive, both sexes and ages combined (ISIS) (26); flightless cormorant (*Phalacrocorax harrisi*) = wild birds during breeding period, all ages combined, males or both genders combined (for parameters that did not vary between sexes; i.e., hematologic parameters) (49); black-faced cormorant (*Phalacrocorax fuscescens*) = wild fledglings (31); pelagic cormorant (*Phalacrocorax pelagicus*) = wild adults recently shot during breeding period, both sexes combined (34); great cormorant (*Phalacrocorax carbo*) = captive male adults during nonbreeding period (5).

[C]Standard error instead of standard deviation.

capacities. Imperial cormorant males perform deeper (mean = 42 m) and longer (mean = 164 sec) dives (20,39,51) than most other non–blue-eyed cormorant species including double-crested cormorant, flightless cormorant, and pelagic cormorant (39 and references therein).

Total WBC counts were higher than values reported for other cormorant species, both captive (26,31) and free-ranging (49), although this may reflect species differences and inherent variability in the WBC counting techniques (41). Nevertheless, general causes of leukocytosis include inflammation due to infection, trauma, toxicities, and hemorrhage (7). Because heterophilia is usually present in inflammation processes, and the heterophil counts in this study were similar to those reported for other cormorants (26,49), it is possible that the increased leukocyte counts observed were of noninfectious origin.

The differential WBC counts of ICs were comparable to published ranges for similar species, except for eosinophils in flightless cormorants and monocytes in black-faced cormorants, which showed higher counts (31,49). Heterophils and lymphocytes make up the majority (i.e., about 80% combined) of WBCs in male ICs. These results agree with those reported for other cormorants (26,34,49).

Plasma biochemistries. Imperial cormorants had lower levels of uric acid than those reported for free-ranging flightless cormorants (49) and pelagic cormorants (34) but were similar to those reported for captive double-crested cormorants by the International Species Information System (ISIS) (26). Given that avian species excrete excess nitrogen from protein metabolism in the form of uric acid (28), variations in uric acid might reflect differences in dietary protein content (34,49), and low levels can also indicate short-term food stress (27).

In birds, blood cholesterol and triglyceride concentrations are affected by the qualitative composition of their diet and are regulated by lipid metabolism (16,17). Imperial cormorants had lower levels of triglycerides than those reported for free-ranging pelagic cormorants (34) but similar to those reported for captive double-crested cormorant by ISIS (26). On the contrary, cholesterol levels were slightly higher than values reported for free-ranging flightless cormorants (49) and pelagic cormorants (34) but similar to those reported for captive double-crested cormorants by ISIS (26). Increases in cholesterol have been associated with dehydration during fasting, high levels of dietary fat, or age (4,10,23). Previous studies on nutrient composition of prey suggest that the diet of the IC is mainly rich in protein but has low lipid levels (21). Therefore, the slight increase in cholesterol levels observed in our study is likely related to dehydration from short-term fasting at the time of sampling. Notwithstanding, territorial aggression of males during the breeding season is regulated by circulating testosterone (e.g., 35,42,52). Given that cholesterol is the precursor of sexual steroids (e.g., 33), an increased level could also reflect steroid production by gonads in male ICs during the chick-rearing period when they show the highest nest-defense intensity (48).

Imperial cormorants had wider ranges of LDH, CPK, and AST activities than did other species of cormorants although median values were lower (26,34,49). Previous studies have shown an increase in LDH, CPK, and AST from capture stress (49). The low values found in our study might reflect tolerance to stress during handling (45). Furthermore, CPK and LDH values showed a decrease over sampling periods, which could indicate that these animals may be becoming habituated to researchers (data not shown).

Imperial cormorants had higher values of ALP than those reported by Newman *et al.* (34) for free-ranging pelagic cormorants and lower than those reported for double-crested cormorants (26). Serum ALP is associated with bone activity because higher levels occur with bone fractures, osteomyelitis, and somatic growth (i.e., young birds) and egg laying (10). Our sample included only adult males, and no abnormalities or evidence of disease were noted during physical examination; thus, differences are likely not clinically important and may be attributable to age variations in sampled birds. It is possible that animals in our study were, overall, younger than those sampled by Newman *et al.* (34) and older than those included in ISIS (26).

Imperial cormorants had lower values of ALT than those reported for other cormorant species (26,34). This enzyme has limited clinical value in birds because it can be increased by pathologic changes in almost all tissues (25), thus hindering result interpretation.

Ca and P are essential for the formation and maintenance of the skeleton and together constitute most of the mineral content of the avian body (6). Ca levels in IC were similar to those reported for captive double-crested cormorant (26), free-ranging black-faced cormorant (31), and flightless cormorant (49) and lower than those found in pelagic cormorants (34). Previous studies in free-ranging seabirds report lower Ca levels in adults than in fledglings (2,53). However, such age differences are not seen in all avian species (25,50). Levels of P were similar to those reported for captive double-crested cormorant by ISIS (26) but lower than those described for free-ranging flightless cormorant (49) and black-faced cormorant (31). Phosphorus levels are known to vary widely (19) and are also commonly elevated in young, growing animals (30). Differences in Ca and P levels may be attributable to age variations in sampled birds (31,49), to dietary differences or the capture and restraint methods used (34), or any combination thereof.

In summary, none of the hematologic and biochemical values obtained in this study indicated signs of clinical pathology or illness. These findings are consistent with external examinations and body weights. Therefore, our results provide a useful baseline for continued efforts to monitor the health status of this population of ICs and a further reference for evaluating the health of related species.

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