

PHYSIOLOGICAL VARIATIONS OF SERUM ELECTROLYTES (Na, K, Ca, P, Mg, Cu) IN FARM-HOUSED *Caiman latirostris* AND *Caiman yacare* (Crocodylia: Alligatoridae)

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ABSTRACT: The purpose of the study was to establish the reference interval for serum electrolytes from captive northeastern Argentine caimans, as well as to detect physiological variations related to species, sex, age, feeding and year season. Samples from 223 healthy subadults, both sexes specimens of *Caiman latirostris* (n=109) and *Caiman yacare* (n=114), were obtained. Serum values of sodium (149.1 ± 5.8 meq/l) and potassium (4.62 ± 0.7 meq/l) were determined by flame photometry. Calcium (9.07 ± 0.8 mg/dl), inorganic phosphorus (4.28 ± 0.8 mg/dl), and magnesium (2.62 ± 0.31 mg/dl) were measured by spectrophotometry. Copper concentration (98.9 ± 16.2 μ g/dl) was determined by atomic absorption spectrophotometry. Magnesium levels were significantly higher in *C. latirostris* than in *C. yacare*, and also they were higher in males than in females ($p < 0.05$). Potassium was lower in *C. latirostris*. Growth produced significant decrease of calcium and inorganic phosphorus. All serum electrolytes registered lowest values in winter. Handling and feeding systems applied on farmed reptiles caused calcium, inorganic phosphorus, sodium, magnesium, and copper values higher than those registered in zoo specimens. Obtained data are applicable for caimans nutritional control, as well as for diagnosis and illness prevention.

Key words: *Caiman latirostris*, *Caiman yacare*, serum electrolytes, physiological variations

VARIACIONES FISIOLÓGICAS DE ELECTROLITOS SÉRICOS (Na, K, Ca, P, Mg, Cu) EN *Caiman latirostris* Y *Caiman yacare* (Crocodylia: Alligatoridae) ALOJADOS EN CRIADEROS

RESUMEN: El propósito del estudio fue establecer el intervalo de referencia para el ionograma sérico de caimanes autóctonos del noreste argentino mantenidos en cautiverio, así como detectar variaciones fisiológicas atribuibles a especie, sexo, edad, alimentación y estación del año. Se emplearon 223 ejemplares sanos, sub-adultos de ambos sexos, de las especies *Caiman latirostris* (n=109) y *Caiman yacare* (n=114). Mediante fotometría de llama se determinaron las concentraciones séricas de sodio ($149,1 \pm 5,8$ meq/l) y potasio ($4,62 \pm 0,7$ meq/l). Por espectrofotometría se obtuvieron los valores de calcio ($9,07 \pm 0,8$ mg/dl), fósforo inorgánico ($4,28 \pm 0,8$ mg/dl) y magnesio ($2,62 \pm 0,31$ mg/dl). El cobre ($98,9 \pm 16,2$ μ g/dl) fue determinado por espectrofotometría de absorción atómica. La concentración de magnesio fue significativamente más alta en *C. latirostris* que en *C. yacare* y mayor en machos que en hembras ($p < 0,05$). El potasio fue más bajo en *C. latirostris*. El crecimiento produjo disminuciones significativas de calcio y fósforo inorgánico. Todos los electrolitos registraron sus niveles más bajos en invierno. Los sistemas de manejo y alimentación aplicados en un criadero produjeron valores de calcio, fósforo inorgánico, sodio, magnesio y cobre superiores a los registrados en un zoológico. Los datos obtenidos son aplicables al control nutricional, así como al diagnóstico y prevención de las enfermedades de estos reptiles.

Palabras claves: *Caiman latirostris*, *Caiman yacare*, electrolitos séricos, variaciones fisiológicas

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INTRODUCTION

Argentine caiman population was diminishing until the decade of 1980, when severe controls against stealthy hunt were imposed. Nowadays, these measures and the introduction of rearing programs as *ranching* system, allowed an increase of its populational density (1).

The *ranching* system consists on the gathering of eggs from wild nests, and an ulterior captive rearing, with restitution of juvenile caimans to the same environment, in a similar number to the proportion of specimens that would have survived under natural conditions, in safeguard of the ecological balance (2). Caimans constitute an important income source for the regional economies, through the export of leather and meat. This production generates currencies nearby to 8 million dollars per year in Argentina (3).

Caiman latirostris and *Caiman yacare* are autochthonous species that inhabit fresh water of our country. The age determination of these reptiles is impossible if date of birth is ignored (4). Therefore, caimans are divided in four important biological categories, according to their total longitude: I (0.23-0.40 m: younger than one year), II (0.41-1.30 m: subadults), III (1.31-1.70 m: adult reproducers, males and females), and IV (> 1.70 m: mature males) (1). Adults caimans are unselective carnivores that can consume either live animals or carrion; in hatcheries they are maintained on balanced food with mineral supplement (2). Environmental temperature is important in feeding behavior and the digestive process. Most crocodilians cease feeding when the temperature goes below 25 °C. The optimal feeding temperature is considered between 25 and 35 °C (5).

Knowledge of serum electrolyte concentrations and their physiological variations are incomplete and controversial in autochthonous caimans. Electrolytes are responsible of the internal environment osmolarity, and their imbalances originate serious illnesses (6). All crocodilians have salt-excretory lingual glands, even those that are fresh water inhabitants, such *C. latirostris* as *C. yacare*. *Alligatoridae* members' lingual glands are much simpler in structure and haven't much ability to concentrate or secrete NaCl compared with other families (7). Periodic water submergence is necessary for all crocodilians in order to counteract water loss. Water intakes consists of incidental drinking and drinking when feeding; water is also absorbed through the skin. Juvenile crocodilians may lose up to 20% of liveweight per day through dehydration. Daily movement between land and water is necessary to maintain their correct osmoregulation (5).

Loss of sodium in fresh water crocodiles is fairly low, and the main sodium route loss is through the skin. Evaporative water loss oc-

curs from the respiratory tract, the mouth, and the skin. Kidneys are the primary route for the excretion of waste nitrogenous but not for NaCl excretion; kidneys are not able to regulate water and salt reabsorption (5). Alligators and caimans are less tolerant to saltwater but they can survive for a considerable period of time in a saltwater environment. This is probably due to their efficient regulation of water loss and sodium absorption. Hatching and juvenile crocodiles have a greater rate of water loss than adult crocodiles, and osmoregulation via behavior and movements between a dry and wet environment is especially important (5).

When Nile crocodiles reared in fresh water are exposed acutely to saltwater, suffer marked dehydration, lethargy, stop eating and loss mass. Cloacal urine osmolality did not increase with increasing salinity. There was a trend of decreasing cloacal urine sodium and chloride and increasing cloacal urine potassium with increased salinity, indicating that urine was not an important route for sodium and chloride excretion (8). *Crocodylus niloticus* held in hypertonic media showed a body weight loss and raised plasma and urinary sodium, potassium, chloride, and osmotic concentration. Within few hours after returning to fresh water, rapid imbibition had greatly restored body weight and produced significant plasma dilution. The compensatory drinking behavior displayed by crocodiles may thus involve the renin-angiotensin system (9).

Mineral nutrition deficiencies are frequent in farmed *C. latirostris* and *C. yacare* (2, 4, 10). Data on caiman serum electrolyte concentrations could be useful to optimize the diagnosis of their sanitary, metabolic and nutritional dysfunctions (11). It could also cooperate with the search of real nutritional requirement of these reptiles in captivity.

The purpose of this try was to determine serum electrolyte reference values from subadults *C. latirostris* and *C. yacare* specimens, as well as to obtain physiological variations related to species, sex, liveweight, dimensions, feeding and year season. The study is part of a larger project that intends to improve their rearing system.

MATERIALS AND METHODS

Experimental subjects

During 2 years, 223 subadults caimans (1), 50% of each sex, clinically healthy, from *C. latirostris* (n=109) and *C. yacare* (n=114) species, were studied. Some of them (n=29) were maintained at the Corrientes City's Zoo, in small ponds, without roof, with running water, which was constantly renovated; they were fed on chicken viscera and fish. Remaining animals (n=194) were housed at the «El Cachapé» farm (Chaco, northeastern Argentina), in roofed tanks with underground water,

which was renewed daily and stayed at 26.9 ± 3.5 °C (heated by gas), fed with meat flour supplemented with vitamins and minerals; sporadically they received bovine viscera. To evaluate growth, reptiles were divided in 3 development stages, considering liveweight and corporal length.

Taking of samples

Hematic and morphometric studies were performed 4 times a year, in each season, during morning hours (8-9 a.m), after a 12-hours fast. Liveweight was obtained using a hanging roman portable scales and corporal dimensions were evaluated with a metallic tape measure. Blood sampling was carried out through the postocipital venous sinus puncture, while reptile was manually held and its jaw remained tied. Blood was centrifuged to obtain serum, which was maintained cooled at 5 °C until its analysis, which was made before 3 hours of extraction.

Laboratory techniques

Sodium and potassium were evaluated using Biopur reagents, in a Metrolab 305-D flame photometer (586 and 768 nm respectively). Serum copper was determined by atomic absorption spectrophotometry, in apparatus GBC932 Plus (324.7 nm). Remainder electrolytes were measured in a Labora Mannheim 4010 UV-visible spectrophotometer, using Wiener Lab reagents, through regular laboratory methods: calcium (cresolphthalein complexone, 570 nm), inorganic phosphorus (phosphomolybdate, 620 nm), and magnesium (xylylidyl blue, 510 nm). All biochemical determinations were checked by an intra-laboratory quality control, using comparative electrolyte patterns (Standatrol).

Statistical analysis

Distributive normality was verified by Wilk-Shapiro test (WS). Parametric statistics included measures from central tendency (arithmetic mean, \bar{x}) and dispersion (standard deviation, SD). Fiduciary probability was evaluated by confidence intervals (CI±95%). Homogeneity of the variance was verified by Bartlett test. Analysis of variance

(ANOVA) was made by one way linear model. Mean comparisons were carried out by Tukey test. Coefficient of linear association was evaluated by correlation (Pearson test). Calculations were made with the aid of a statistical software (*Statistix* 1996). For all inferences a 5% significance was specified, below which the equality null hypothesis was rejected.

RESULTS

Descriptive statistics obtained in all studied caimans are detailed in Table 1. Values approximately normal distributed (WS), allowed the use of parametric statistics. Data dispersion (SD) did not exceed the limits recommended by parametric statistics. Confidence intervals (CI) were adjusted around arithmetic means (\bar{x}), but individual ranges were wide.

Electrolyte values obtained on each studied species are shown in Table 2. Potassium serum levels were significantly higher in *C. yacare*, and magnesium serum concentrations were significantly higher in *C. latirostris* ($p < 0.05$).

Physiological variations related to sex and age (established groups according to the increase of liveweight and total length) are shown in Table 3. Serum magnesium values were significantly higher in males than in females. Calcium and inorganic phosphorus decreased significantly according to age increase (liveweight and length increases).

Pearson test revealed high linear association degree ($p < 0.05$) between liveweight and variables as total longitude ($r = 0.90$), muzzle tail longitude ($r = 0.83$), head longitude ($r = 0.79$), head wide ($r = 0.86$), and thoracic perimeter ($r = 0.88$). When considering growth stages, liveweight correlated to serum concentrations of calcium ($r = 0.88$), inorganic phosphorus ($r = 0.92$) and sodium ($r = 0.84$). Total length increment also correlated to the decrease of such electrolytes.

Variations of serum electrolytes during year seasons (temperature) and place of origin (feeding and handling systems) are shown in Table 4. Sodium, potassium, calcium, inorganic phosphorus, magnesium and copper concentrations were

Table 1. Values obtained in total studied population ($n = 223$).

Tabla 1. Valores obtenidos en la población total estudiada ($n = 223$).

Parameter	$\bar{x} \pm SD$	WS	CI±95%	range
Na (meq/l)	149.1 ± 5.8	0.941	148.2 149.9	134 159
K (meq/l)	4.62 ± 0.7	0.948	4.53 4.72	3.3 5.9
Ca (mg/dl)	9.07 ± 0.8	0.952	8.95 9.19	7.80 11.22
P (mg/dl)	4.28 ± 0.8	0.949	4.13 4.42	2.42 6.90
Mg (mg/dl)	2.62 ± 0.31	0.993	2.57 2.66	1.90 3.36
Cu (µg/dl)	98.9 ± 16.2	0.956	91.5 106.3	31 168

x: arithmetic mean, SD: standard deviation, WS: Wilk-Shapiro distributive normality test (critical value: 0.947, $\alpha = 0.05$), CI±95%: confidence interval.

Table 2. Values obtained on each species.
Tabla 2. Valores obtenidos en cada especie.

Parameter	<i>C. latirostris</i> (n = 109)		<i>C. yacare</i> (n = 114)	
	$\bar{x} \pm SD$	CI±95%	$\bar{x} \pm SD$	CI±95%
Na (meq/l)	149.4 ± 5.6	148.3 150.6	148.7 ± 6.1	147.5 149.9
K (meq/l)	4.75 ± 0.6 ^a	4.61 4.88	4.91 ± 0.7 ^b	4.36 4.64
Ca (mg/dl)	9.15 ± 0.9	8.98 9.33	8.98 ± 0.8	8.81 9.17
P (mg/dl)	4.22 ± 0.8	4.01 4.43	4.32 ± 0.9	4.12 4.53
Mg (mg/dl)	2.71 ± 0.31 ^a	2.66 2.77	2.52 ± 0.28 ^b	2.45 2.58
Cu (µg/dl)	95.1 ± 14.2	84.2 103.9	103.5 ± 15.9	93.1 115.7

x: arithmetic mean, SD: standard deviation, CI±95%: confidence interval. In each line, different letters indicate significant differences (Tukey test, p < 0.05).

Table 3. Variations according to sex, liveweight, and total length in both species (x).
Tabla 3. Variaciones según sexo, peso vivo y longitud total en ambas especies (x)

Parameter	sex		liveweight (kg)			total length (cm)		
	male	female	< 3.5	3.6-5.0	> 5	< 99	100-110	> 110
Na (meq/l)	148.7	149.9	150.0	149.3	148.2	150.1	149.5	146.7
K (meq/l)	4.54	4.68	4.64	4.56	4.67	4.66	4.59	4.62
Ca (mg/dl)	9.09	8.92	9.10 ^a	9.08 ^a	9.01 ^b	9.12 ^a	9.06 ^b	9.02 ^b
P (mg/dl)	4.29	4.26	4.42 ^a	4.37 ^a	4.09 ^b	4.50 ^a	4.38 ^a	4.01 ^b
Mg (mg/dl)	2.71 ^a	2.52 ^b	2.64	2.59	2.65	2.63	2.60	2.61
Cu (µg/dl)	102.0	97.8	98.2	99.3	98.5	99.8	101.5	95.6

x: arithmetic mean. In each line, different letters indicate significant differences (Tukey test, p < 0.05).

Table 4. Variations according to year season and feeding system in both species (x).
Tabla 4. Variaciones según estación del año y sistema de alimentación en ambas especies (x)

Parameter	year season				feeding	
	spring	summer	autumn	winter	farm	zoo
Na (meq/l)	149.7 ^a	151.4 ^a	146.8 ^b	146.2 ^b	149.8 ^a	145.4 ^b
K (meq/l)	5.02 ^a	4.85 ^a	4.38 ^b	4.04 ^b	4.59	4.65
Ca (mg/dl)	9.37 ^a	9.43 ^a	8.76 ^b	8.62 ^b	9.09	8.98
P (mg/dl)	4.59 ^a	4.70 ^a	4.22 ^b	3.61 ^c	4.31	4.25
Mg (mg/dl)	2.62 ^a	2.68 ^a	2.70 ^a	2.55 ^b	2.77 ^a	2.41 ^b
Cu (µg/dl)	110.2 ^a	125.1 ^a	103.4 ^a	66.5 ^b	113.7 ^a	88.5 ^b

x: arithmetic mean. In each line, different letters indicate significant differences (Tukey test, p < 0.05).

significantly lower in winter than in summer. No significant differences were registered between summer and spring (warm and mild temperatures respectively). Sodium, magnesium and copper serum levels were significantly higher in reptiles reared by *ranching* system than in those housed at the zoo.

DISCUSSION

In crocodiles, the lack of fast causes an increase of certain plasma components (12). Eventual variations due to postprandial effect and circadian rhythm (6) were excluded from the experimental design, because samples were taken during fast, in uniform morning hours. Wide individual ranges verified in this study should be attributed to reptilian physiological particularities, because its blood parameters

fluctuate considerably due to feeding system, habitat, climate and sex (11). Blood values from other aquatic animals, such as amphibians, also register greater oscillations due to their scarce regulation mechanisms, and to a higher tolerance to hemodilution and hemoconcentration (6).

Sodium serum concentration mean value obtained in the present study for *C. latirostris* and *C. yacare*, was similar to the natremia reported for fresh water specimens as *C. niloticus*: 154±1 meq/l (13) and *Alligator mississippiensis*: 148±5 meq/l (14) and 141 meq/l (15), as well as for saltwater specimens as *C. porosus*: 143-161 meq/l (12) and *Crocodylus acutus*: 149 meq/l (15). Since sodium is the main responsible of the internal environment osmotic pressure (6), this fact ratifies that crocodiles are able to regulate efficiently their osmolarity, so much in fresh water

as in saltwater external environments.

Potassium serum levels obtained here matched to those published for fresh water specimens as *C. niloticus*: 3.8 ± 0.5 meq/l (13) and *A. mississippiensis*: 4.1 ± 0.7 meq/l (14) and 3.8 meq/l (15). On the other hand, kalemias from saltwater specimens would be higher: 7.9 meq/l in *C. acutus* (15) and 3.8 - 7.2 meq/l in *C. porosus* (12).

Calcemia from caimans studied here was slightly lower than that reported in fresh water specimens as *C. niloticus*: 11.88 mg/dl (13) and *A. mississippiensis*: 11.3 ± 0.5 mg/dl (14) and 10.4 mg/dl (10, 16). However, it was considerably higher than those obtained in saltwater specimens as *C. acutus*: 13.6 mg/dl (13) and *C. porosus*: 9.64 - 13.8 mg/dl (12). Serum calcium would be also high on land reptiles as *Iguana iguana*: 9.0 - 25.1 mg/dl, *Boa constrictor*: 10 - 22 mg/dl, and *Testudo radiata*: 10.8 - 14.4 mg/dl (15).

The inorganic phosphorus value registered here in *C. latirostris* and *C. yacare* was lower than those obtained in *A. mississippiensis*: 8.2 ± 2.2 mg/dl (14), *C. niloticus*: 9.3 mg/dl (16) and *C. porosus*: 3.7 - 8.9 mg/dl (12). Perhaps these low values should be related to nutritional reasons, keeping in mind that in most vertebrates, the quantity of serum phosphates is directly proportional to dietary phosphorus intake (6).

On the contrary, serum magnesium concentrations obtained in this study were approximately similar to those published for *C. niloticus*: 2.18 mg/dl (13), *C. porosus*: 1.94 - 3.40 mg/dl (12), *A. mississippiensis*: 3.64 mg/dl (10) and *C. acutus*: 4.65 mg/dl (13). Data on copper serum concentrations from reptiles, were not found.

Data on *C. latirostris* obtained here matched to those reported on 12 juvenile animals from the same species (11) in sodium serum concentration (150.8 ± 2.9 meq/l), but considerably differ from levels of potassium (6.12 ± 1.03 meq/l), calcium (14.3 ± 4.5 mg/dl) and inorganic phosphorus (9.58 ± 1.99 mg/dl). Electrolyte serum values published for *C. latirostris* (n = 65) in other investigation (4) are similar to those found in the current study for potassium (4.6 ± 0.16 meq/l), calcium (8.9 ± 0.6 mg/dl) and inorganic phosphorus (5.64 ± 0.46 mg/dl), although the sodium level was lower (144 ± 1 meq/l). Data about calcium and inorganic phosphorus obtained in another assay (17) on 50 *C. latirostris* juvenile and adults specimens, are not very different (10.12 ± 1.52 and 5.4 ± 2.0 mg/dl respectively) to those registered here.

Some electrolyte values obtained here on *C. yacare* are similar to those published on the same species (11) by other investigators (sodium: 148.2 ± 3.1 meq/l, potassium: 4.58 ± 1.22 meq/l and inorganic phosphorus: 5.16 ± 0.93 mg/dl). On the contrary, ion levels reported on 33 *C. yacare*

juvenile specimens (4) were surprisingly lower (sodium: 129.3 ± 2.9 meq/l, potassium: 3.51 ± 0.23 meq/l and calcium: 5.86 ± 0.84 mg/dl). In this try, potassium values were significantly higher in *C. yacare* than in *C. latirostris*; results obtained in another study were in the other way around (4). Higher values of inorganic phosphorus in *C. latirostris* than in *C. yacare* (4, 11), were not corroborated. Reference data on magnesium and copper serum concentrations were not found from autochthonous caimans.

Differences between sexes were scarce in current study. *Crocodylus palustris* subadult specimens did not register significant differences related to sex either, although calcium and potassium serum values were higher in juvenile males than in juvenile females (18). During late vitellogenesis of adult female *A. mississippiensis*, elevated plasma vitellogenin and estradiol-17beta were not correlated with plasma phosphorus but were correlated with plasma calcium concentration (19).

The increase of age in studied caimans (increases of liveweight and total length) correlated to calcium and inorganic phosphorus serum concentration decrease. This is a common fact in growing vertebrates and it is related to endocrine mechanisms that cause deposition of calcium phosphate (hydroxyapatite) in bones (6).

Phosphorus and calcium are also necessary for development of crocodile dermal bones (osteoderms). Calcium concentration in osteoderms from *Crocodylus johnstoni*, was inversely related to the solubility of phosphate (but not to those of sodium nor potassium), and varied according to age (size), but not sex nor reproductive status (20).

Maybe the lowest winter values registered in all studied electrolytes should be related to the ceasing of mineral intake caused by the interruption of feeding during cold station (5). *A. mississippiensis* frequently suffers malnutrition due to corporal reserves depletion happened during the winter; the feeding is resumed in spring (14). Temperature acute decreases increased blood pH, but had no significant effect on plasma sodium, chloride and bicarbonate concentration, and only had minor effects on plasma potassium concentration from *A. mississippiensis* (21).

Calcium, inorganic phosphorus, sodium, magnesium and copper serum concentrations were lower (significantly for last three electrolytes) in caimans fed on diet supplied at the zoo, than in those fed on diet given at the hatchery. This fact perhaps reflects mineral deficiencies in the diet composition, considering that -in mammals- mineral serum levels, especially inorganic phosphorus, magnesium and copper, are nutritional indicators that vary according to diet mineral amount (6). Copper hepatic and renal

concentrations from *C. niloticus* are similar to those reported in other vertebrates and acquire relevance for the evaluation of water degree contamination (22).

Preys captured by crocodiles in wild life provide appropriate minerals and trace-elements levels, but the food consumed during captivity not always have appropriate electrolytical proportions (10). Nowadays the mean problem of northeastern Argentine caiman *ranching* system is the feeding improvement; dietary calcium and phosphorus deficiencies cause metabolic and endocrine dysfunctions in crocodiles (4). Imbalance of dietary calcium / phosphorus rate generates severe osseous anomalies in these reptiles (10). Juvenile caimans can suffer nutritional secondary hyperparathyroidism due to the lack dietary calcium (2).

Electrolytes carry out indispensable homeostatic functions, such as bones and teeth formation, polarization of membranes, integration of enzymatic systems, energy storage, acid-base balance, breathing, and clotting (6). Divalent metal ions such as calcium and magnesium, but not copper, are necessary to activate the serum complement cascade in *A. mississippiensis* (23). The yolk of *A. mississippiensis* egg is the primary source of calcium, phosphorus and magnesium for the embryo development; these elements are depleted rapidly from the yolk once the embryo enters the growing phase (24).

An episode of mineral deficiency was registered when a food change was made on *C. yacare*. The diet of replacement of a bovine meat supplemented with minerals and vitamins, into another diet made by meat and blood flour (without supplement), caused marked decrease of calcium and inorganic phosphorus plasma concentrations, although it also generated plasma albumin increase (4). Calcium fall reached surprisingly low levels (initial: 11.78 mg/dl, final: 2.48 mg/dl). Nevertheless, this information should contain some error, because such hypocalcemia is incompatible with life; calcemias below 7 mg/dl cause tetany on muscular cells, specially when plasma albumin are high (6).

Biochemical values can be useful to evaluate caiman physiological state and to detect its illnesses on an early stage (11), but should have been correctly determined in the laboratory and they should be compared with adequate reference intervals (6). Several authors early quoted do not describe the techniques used in their works. Other investigators used dry chemistry methods, such as reactive strips. According to each case, heparinized plasma or serum were used. In certain studies, reptiles were not divided according to sex, age, or year season. Blood extraction technique could be another cause of values variation, but they are not always specified in consulted

references (15).

Standardization of clinical pathology techniques is important to appreciate the laboratory tests as effective tools on caiman breeding, mainly because ideal conditions for intensive crocodile rearing have not yet been established. Consequently, mortality is often directly linked to handling and nutritional failures (25).

In conclusion, serum electrolyte reference values from subadults *C. latirostris* and *C. yacare* captive specimens, as well as significant physiological variations related to species, sex, age (liveweight, dimensions), environmental temperature (year season), and feeding system, are established. Obtained data can be applied to caiman rearing improve in Argentine northeastern.

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REFERENCES

1. Waller T, Micucci PA. Relevamiento de la distribución, hábitat y abundancia de los crocodilos de la Provincia de Corrientes, Argentina. Memorias de la 1ra. Reunión Regional del Grupo de Especialistas en Cocodrilos, Santa Marta (Colombia). 1993; p. 341-385.
2. Prado WS, Gómez O, Balla P. Crianza en cautiverio y reintroducción de individuos (*C. latirostris* y *C. yacare*). Boletín Técnico de la Fundación Vida Silvestre Argentina. 2001; 55: 26-38.
3. Larriera A. Programa de ranching experimental en Argentina. Anales del III Congreso Internacional sobre Manejo de Fauna Silvestre, Santa Cruz (Bolivia). 1997; p. 166.
4. Ferreyra H, Uhart M. Evaluación y evolución del estado sanitario de *Caiman latirostris* y *Caiman yacare* en el Refugio El Cachapé. Boletín Técnico de la Fundación Vida Silvestre Argentina. 2001; 55 (Anexo III): 1-15.
5. Lane TJ. Crocodilians. In: Reptile Medicine and Surgery (Mader DR Ed.), Ed Saunders, Philadelphia (USA). 1996; p. 78-98.
6. Coppo JA. Fisiología Comparada del Medio Interno, Ed Dunken, Buenos Aires (Argentina). 2001; p. 212-216.
7. Taplin L, Grigg GC. Salt glands in the tongue of the estuarine crocodile *Crocodylus porosus*. Science. 1981; 212: 1045-1048.
8. Leslie AJ, Spotila JR. Osmoregulation of the Nile crocodile, *Crocodylus niloticus*, in Lake St. Lucia, Kwazulu/Natal, South Africa. Comp Biochem Physiol A Mol Integr Physiol. 2000; 126: 351-365.
9. Balment RJ, Loveridge JP. Endocrines and osmoregulatory mechanisms in the Nile crocodile, *Crocodylus niloticus*. Gen Comp Endocrinol. 1989; 73: 361-367.

10. Frye FL. Reptile Clinician's Handbook, Ed Krieger, Florida (USA). 1994; p. 214.
11. Uhart M, Prado W, Beldoménico P, Rossetti C, Ferreyra Armas MC, Martínez A, Bardón JC, Avilés G, Karesh W. Estudios sanitarios comparativos de yacarés (*Caiman latirostris* y *Caiman yacare*) silvestres y cautivos. Boletín Técnico de la Fundación Vida Silvestre Argentina. 2001; 55: 39-50.
12. Millan JM, Janmaat A, Richardson KC, Chambers LK, Formiatti KR. Reference ranges for biochemical and haematological values in farmed saltwater crocodile (*Crocodylus porosus*) yearlings. Aust Vet J. 1997; 75: 814-817.
13. Watson PA. Effects of blasting on Nile crocodiles, *Crocodylus niloticus*. Proceedings of the 10th Working Meeting of the Crocodile Specialist Group IUCN, Gainsville, Florida (USA). 1990: p. 240-252.
14. Schoeb TR, Heaton-Jones TG, Clemmons RM, Carbonneau DA, Woodward AR, Shelton D, Poppenga RH. Clinical and necropsy findings associated with increased mortality among American alligators of Lake Griffin, Florida. J Wildl Dis. 2002; 38: 320-337.
15. Stein G. Hematologic and blood chemistry values in reptiles. In: Reptile Medicine and Surgery (Mader DR Ed.), Ed Saunders, Philadelphia (USA). 1996; p. 479-483.
16. Foggin CM. Diseases and disease control on crocodile farms in Zimbabwe. In: Wildlife Management: Crocodiles and Alligators (Webb GJ, Manolis SC, Whitehead PJ, Ed.), Ed Surrey Beatty, Chipping Norton (USA). 1987; p. 351-362.
17. Troiano JC, Althaus R. Hallazgos hematológicos en *Caiman latirostris* (Crocodylia: Alligatoridae) en condiciones de cautiverio. Memorias del IV Workshop sobre Conservación y Manejo del Yacaré Overo, Santo Tomé (Santa Fe, Argentina). 1994; p. 12-24.
18. Stacy BA, Whitaker N. Hematology and blood biochemistry of captive mugger crocodiles (*Crocodylus palustris*). J Zoo Wildl Med. 2000; 31: 339-347.
19. Guillette LJ, Woodward AR, Crain DA, Masson GR, Palmer BD, Cox MC, You-Xiang Q, Orlando EF. The reproductive cycle of the female American alligator (*Alligator mississippiensis*). Gen Comp Endocrinol. 1997; 108: 87-101.
20. Jeffree RA, Markich SJ, Tucker AD. Patterns of metal accumulation in osteoderms of the Australian freshwater crocodile, *Crocodylus johnstoni*. Sci Total Environ. 2005; 336: 71-80.
21. Douse MA, Mitchell GS. Time course of temperature effects on arterial acid-base status in *Alligator mississippiensis*. Respir Physiol. 1991; 83: 87-102.
22. Almlí B, Mwase M, Sivertsen T, Musonda MM, Flaoyen A. Hepatic and renal concentrations of 10 trace elements in crocodiles (*Crocodylus niloticus*) in the Kafue and Luangwa rivers in Zambia. Sci Total Environ. 2005; 20: 75-82.
23. Merchant ME, Verret B, Elsey RM. Role of divalent metal ions in serum complement activity of the American alligator (*Alligator mississippiensis*). Comp Biochem Physiol B Biochem Mol Biol. 2005; 141: 289-293.
24. Packard MJ, Packard GC. Mobilization of calcium, phosphorus, and magnesium by embryonic alligators (*Alligator mississippiensis*). Am J Physiol. 1989; 257: 1541-1547.
25. Huchzermeyer FW. Diseases of farmed crocodiles and ostriches. Rev Sci Tech. 2002; 21: 265-276.