DOI: 10.7589/2016-06-143

Journal of Wildlife Diseases, 53(3), 2017, pp. 447–458

EFFECTS OF LIVE-TRAPPING AND ISOFLURANE ANESTHESIA ON FREE-RANGING AMERICAN MARTENS (*MARTES AMERICANA*)

Maria C. Spriggs,[1,2,7] Lisa I. Muller,[2] Paul Keenlance,[3] Robert L. Sanders,[4] Jill C. Witt,[5] and Debra L. Miller[2,6]

[1] Mesker Park Zoo & Botanic Garden, 1545 Mesker Park Drive, Evansville, Indiana 47720, USA
[2] Department of Forestry, Wildlife and Fisheries, University of Tennessee, 274 Ellington Plant Sciences Building, Knoxville, Tennessee 37996, USA
[3] Department of Biology, Grand Valley State University, 1 Campus Drive, Allendale, Michigan 49401, USA
[4] Department of Natural Resources, Little River Band of Ottawa Indians, 310 Ninth Street, Manistee, Michigan 49660, USA
[5] Department of Biology, University of Michigan-Flint, 265 Murchie Science Building, 303 E Kearsley Street, Flint, Michigan 48502, USA
[6] Department of Biomedical and Diagnostic Sciences, College of Veterinary Medicine, University of Tennessee, 2407 River Drive, Knoxville, Tennessee 37996, USA
[7] Corresponding author (email: maria.spriggs@buschgardens.com)

ABSTRACT: Seventy-two free-ranging American martens (*Martes americana*) in Michigan, US were immobilized using isoflurane from 2011 to 2015. In total, 129 anesthetic procedures were performed with no mortalities. Hypothermia and hyperthermia were the most common anesthetic complications, and the mean rectal temperatures were significantly higher during summer than in winter. Dental abnormalities were common; the majority of abnormal findings were broken or discolored teeth attributed to previous dental trauma and were not trap-induced. Blood (n=72) was analyzed from 53 martens for venous blood gas, lactate, hematocrit, and/or selected serum biochemistry analytes. Lactate concentration was measured by two different devices (VetScan i-STAT 1 and Lactate Plus) and compared for clinical agreement for 26 samples. Both methods for lactate measurement provided statistically similar results. Using domestic feline reference ranges, the acid-base status and relative arterial oxygen saturation of anesthetized martens in this study were normal as determined by blood pH and pulse oximetry, respectively. Serum biochemistry parameters, multiple environmental parameters, and marten-specific attributes were evaluated for their influence on lactate in American martens using linear regression and an information-theoretic approach with model averaging. Blood urea nitrogen was in all of the top models and was positively related to lactate (β=0.02, 95% confidence interval: 0.00–0.04). Initial body temperature, ambient temperature, and time from trap discovery until immobilization of martens were informative predictors for lactate level. Recommendations for the live-trapping and isoflurane anesthesia of free-ranging martens include using caution during warmer summer months, minimizing disturbance prior to induction, monitoring lactate in addition to vital rates, and being prepared to prevent or treat both hypothermia and hyperthermia during any time of year.

Key words: American marten, anesthesia, blood gas, capture, handling, isoflurane, lactate.

INTRODUCTION

The American marten (*Martes americana*) is a mesocarnivore whose range extends from parts of the US into the boreal forests of Canada (Powell et al. 2003). The species was reintroduced to the Upper Peninsula and northern Lower Peninsula (NLP) of Michigan in the mid-20th century (Earle et al. 2001; Cooley 2004). Current efforts to evaluate the status of these populations require immobilization of martens.

We assessed the efficacy and risk of isoflurane gas anesthesia in live-trapped American martens. Although injectable anesthetics have been used, advantages of isoflurane include rapid induction and recovery, ability to adjust depth of anesthesia, no risk of miscalculations in dosage or erroneous weight estimations, and no requirement for controlled drug access. Lactate values have been used to assess exertion associated with capture of other free-ranging species including wolverine (*Gulo gulo*) because lactate increases under conditions of anaerobic cellular respiration (Fahlman et al. 2008). We report blood gas and lactate values of American martens

under isoflurane anesthesia and expand on a previous report on safe use of isoflurane in American martens (Desmarchelier et al. 2007). Understanding the variables that influence lactate can aid in creating protocols to minimize morbidity or mortality associated with immobilization of American martens.

MATERIALS AND METHODS

We trapped American martens in the Manistee National Forest (43°51′0″N, 85°57′0″W; *n*=43) in the NLP and in both the Ottawa National Forest (46°13′48″N, 88°57′0″W; *n*=5) and the Hiawatha National Forest (46°14′0″N, 84°50′0″W; *n*=24) in the Upper Peninsula using box live traps (Tomahawk Live Trap, Model no. 103 and no. 105, Hazelwurst, Wisconsin, USA) from 2011 to 2015. Traps were checked one or more times daily. We restrained the majority of martens in a fabric cone, and anesthesia was induced via facemask with a portable isoflurane (IsoFlo®, Abbott Laboratories, Abbott Park, Illinois, USA) anesthesia machine as described by Desmarchelier et al. (2007). The machine was carried into the backcountry when necessary by affixing the vaporizer and oxygen cylinder to a backpack (Fig. 1). We chamber-induced some martens by placing the trap inside a large plastic container or by placing an isoflurane-soaked cotton ball at the tip of the facemask but not in direct contact with the nose. Regardless of induction method, we maintained all martens at a level plane of anesthesia with 0.5–5% isoflurane through a facemask, and martens were monitored by trained personnel. We measured body mass and recorded heart rate, respiratory rate, rectal temperature (InitialT), and relative arterial oxygen saturation after induction and periodically throughout the procedure as determined by pulse oximetry (SurgiVet V34041, SurgiVet, Inc., Waukesha, Wisconsin, USA). We recorded induction time (time from onset of isoflurane to recumbency), length of procedure (onset to discontinuation of isoflurane), recovery time to standing, and time to release. Martens were implanted subcutaneously between the shoulder blades with a sterile microchip (AVID Identification Systems, Norco, California, USA). Martens (*n*=55) were fitted with a 21–24 g radiotelemetry collar (Advanced Telemetry Systems, Isanti, Minnesota, USA; Holohil Systems Ltd., Carp, Ontario, Canada) as part of a larger habitat and population study. We measured the time from when an animal was discovered in a trap until immobilization started (TimeSince), number of broken nails, presence of fresh gingival or dental trauma, ambient temperature (AmbientT), season (summer = April–September, winter = October–March), sex, lactation status, and whether the animal was wearing a radiotelemetry collar.

FIGURE 1. An isoflurane vaporizer and E-tank oxygen cylinder are attached to a backpack frame for backcountry immobilization of free-ranging American martens (*Martes americana*) in Michigan, USA, for radiocollar placement and physiological assessment. It is important that the vaporizer filled with isoflurane does not tip over during storage, transport, or use.

To prevent or treat hypothermia, we placed a foam pad under the animal and used towels (as covers), instant hand warmers, warm air from a blow dryer, and minimal amounts of isopropyl alcohol, and moved the anesthesia machine and animal into a heated truck cab as needed. To prevent or treat hyperthermia, we used 70% isopropyl alcohol (First Priority, Elgin, Illinois, USA) on footpads and jugular grooves, and administered subcutaneous fluids and/or fluids per rectum. To prevent or treat dehydration, we administered subcutaneous fluids (0.9% NaCl, 30 mL/kg subcutaneously, Hospira, Lake Forest, Illinois, USA), either warmed or at room temperature as appropriate. We had emergency supplies, including endotracheal tubes and laryngoscope and emergency drugs, available to treat serious complications should they arise. Capture and handling protocols were approved by the Univer-

sity of Tennessee Animal Care and Use Committee (protocol no. 2180).

We sampled animals more than once if recaptured for radiocollar maintenance. Blood was collected from the jugular vein, and the volume did not exceed 1% body mass. We did not repeat sampling in the same animal within 30 d to minimize risk of iatrogenic anemia. Blood was placed into lithium heparin anticoagulant (BD Microtainer® Tubes, Becton Dickinson and Company, Franklin Lakes, New Jersey, USA) and serum separator tubes (Covidien, Mansfield, Massachusetts, USA). We allowed the blood in the serum separator tubes to clot and centrifuged it in the field for 10 min at 1,350 × G and transported serum in a cooler at a controlled temperature between 2.7 C and 5 C.

We analyzed fresh whole or lithium heparinized venous blood in the field within 10 min of collection using a portable blood gas analyzer (VetScan i-STAT 1, Abaxis, Union City, California, USA) with a CG4+ cartridge (analytes: pH, partial pressure of carbon dioxide [pCO_2], bicarbonate [HCO_3], total carbon dioxide [tCO_2], partial pressure of oxygen [pO_2], oxygen saturation [sO_2], lactate) and a portable lactate meter (Lactate Plus, Nova Biomedical, Waltham, Massachusetts, USA). Serum samples were analyzed within 8 h using VetScan (Abaxis) with an equine rotor (creatine kinase [CPK], aspartate aminotransferase [AST]) and a small animal comprehensive rotor (blood urea nitrogen [BUN], creatinine, glucose). We used whole blood to determine hematocrit using microhematocrit capillary tubes (SafeCrit, Westwood, Massachusetts, USA) and the StatSpin® VT centrifuge at 13,700 × G for 120 s (Iris, Westwood, Massachusetts, USA).

Statistical analyses were conducted using JMP® Pro 10.0.02 and SAS 9.4 (SAS Institute Inc., Cary, North Carolina, USA) and Program R (R Development Core Team 2013). We used the Shapiro-Wilk test to test for normalcy of data distribution and *t*-test to compare means. Lactate values were measured simultaneously by i-STAT and the Lactate Plus meter and compared for clinical agreement using the Bland-Altman technique (Bland and Altman 1986). We compared blood parameters and environmental variables with marten-specific attributes to lactate using a priori models from factors hypothesized to affect lactate values. Variables were grouped based on previously reported animal stressors and factors known to influence anaerobic cellular respiration or hypoxia leading to possible increases in lactate. We used mixed linear regression models with repeated measures on individual martens in SAS and Akaike Information Criteria for small sample sizes (AICc) for model selection. We used model averaging for top models where $\Delta AICc \leq 2.0$ (Burnham and Anderson 2002).

RESULTS

We performed 129 anesthetic procedures on 72 individual American martens (40 males, 32 females), including 11 juveniles (<6 mo old), with no mortalities. On average, adult males weighed 1,002±116 g, and females weighed 695±97 g. Ambient temperatures in the field ranged from −12 C to 32 C. Mean induction time for facemask induction was 1.4 min (range: 1–9 min, n=121). Induction time using the isoflurane-soaked cotton-ball method was 1.7 min (range: 1–2, n=3), while chamber method was 7.6 min (range: 3–13, n=5). Average procedure length was 24±8 min (range: 8–51 min, n=121). Average time between discontinuation of isoflurane and recovery to standing was recorded for 112 procedures with a mean of 7±4 min (range: 1–20 min). The time between discontinuation of isoflurane and release was recorded for 116 procedures and averaged 15±6 min (range: 6–48 min). Average rectal temperature, heart rate, respiratory rate, and relative arterial oxygen saturation during summer and winter months are shown in Table 1. Rectal temperature was significantly higher during summer months than winter months at all time points: 0–9 min (P=0.002), 10–19 min (P<0.000), and 20+ min (P=0.012). There was a significant (P=0.03) correlation (r=0.27) between ambient temperature and initial rectal temperature. Heart rate during the first 10 min of anesthesia was significantly higher in summer than in winter (P<0.05). No animals were intubated due to apnea or treated with emergency drugs.

The most common physical exam finding was previously damaged dentition, which was seen in 43% of the 72 individual martens examined. Of the affected martens, 19% had one or more previously fractured but not yet devitalized (discolored) canines, 7% had one or more intact but devitalized canines, 10% had one or more canines that were both fractured and devitalized, and 7% had previously damaged incisors (discolored, fractured,

TABLE 1. Vital signs (mean±SD) of 72 American martens (*Martes americana*) during 121 anesthetic events using isoflurane after being captured in Michigan, USA in summer or winter for radiocollaring and physiological assessment. Significant differences ($P<0.05$) between values for seasons at each time are indicated by differing superscript letters. NR = not reported.

Season	Time after induction (min)	Rectal temperature (C)	Heart rate (beats/min)	Respiratory rate (breaths/min)	Relative arterial oxygen saturation (%)
Summer (n=94)	0–9	39.7±0.6[a]	256±43[a]	47±17	97±3
	10–19	38.7±1.0[a]	234±51	36±13	97±3
	>20	38.0±1.2[a]	234±31	27±11	96±3
Winter (n=35)	0–9	39.1±1.1[b]	236±47[b]	42±16	97±3
	10–19	37.5±1.2[b]	248±23	28±5	97±2
	>20	36.7±0.9[b]	211±40	NR	97±1

and/or excessively worn). Four martens (6%) were noted to have tartar. There was no sex difference for dental damage (P=0.48). In contrast to the previously damaged teeth seen in individual martens, minor recent gingival trauma, presumably due to chewing at the trap, was noted in 9% of 129 procedures. A freshly fractured tooth was seen during two procedures (2%).

Other findings included superficial abrasions on chin, lips, or nose (n=8), collar-associated dermatitis (n=5), hair mats (n=3), puncture or suspected bite wound on leg (n=2), crust on ear pinna (n=2), fleas (n=2), mild epistaxis (n=1), pinpoint ulcer on the anus (n=1), and thorn in skin of leg (n=1). The collar was removed in all cases of dermatitis, and a long-acting ceftiofur crystalline free acid injection (Excede®, 200 mg/mL, Zoetis, Florham Park, New Jersey, USA; 6.6 mg/kg, subcutaneously) was administered to three of the five affected animals. Nail wear or breakage was recorded for 100 animals. One or more nails was recently worn, frayed, or broken in 80 animals, likely due to digging while in the trap.

Martens were treated for hyperthermia (rectal temperature ≥40.5 C) during 18/129 procedures, 16 of which occurred during summer months. All but three of these 18 animals had a final rectal temperature of <40 C by the end of the procedure. Martens were treated for mild hypothermia (<36.6 C) during 12.4% of procedures (n=16), and severe hypothermia (<35.5 C) during 3.9% of procedures (n=5). Of the 21 procedures during which hypothermia occurred, only nine were during winter months and occurred despite the described preventive measures. Subcutaneous fluids were administered during 47% of procedures and became routine practice during summer months for prevention or treatment of hyperthermia, dehydration, hyperlactemia, and/or capture myopathy. Due to difficulty in keeping fluids warm, they were not routinely administered during winter.

We collected 72 blood samples from 53 martens. Eleven martens were sampled two or more times during recapture. Venous blood gas, lactate, and select serum biochemistry values are shown in Table 2. We omitted two values obtained via the i-STAT from analysis because of error messages for temperature of the machine at the time of use. We had only a Lactate Plus value and no i-STAT values for 17 animals. Bland-Altman analysis of 26 events found a mean difference of –0.13±0.49 mmol/L (range: –0.76–1.38 mmol/L) between the Lactate Plus meter and the i-STAT. All but two values were ±2 SD of the mean, and both of the outliers were at the lower end of the range for lactate (Figs. 2, 3). Therefore, we considered both i-STAT and Lactate Plus meter readings as equivalent measurements of lactate. Where both readings were available, we used the mean of the two measurements. If only one method was available, we used that value. Lactate was not normally distributed and was trans-

TABLE 2. Venous blood gas, lactate, and select serum biochemistry values for 53 American martens (*Martes americana*) under isoflurane anesthesia after being captured in Michigan, USA during summer and winter for radiocollaring and physiological assessment. Range values previously reported for polecats and other carnivores (domestic ferret and cat) under isoflurane anesthesia are shown for comparison.[a]

Parameter	*n*	Mean	SD	Range	Polecat (mean±SEM)[b]	Other carnivore ranges
pH	37	7.27	0.07	7.06–7.40	7.193±0.02	7.25–7.40[c]
pCO_2 (mmHg)	37	47.4	13.1	30.4–112.0	55±3	33.0–51.0[c]
HCO_3 (mmol/L)	37	24.4	22.3	14.7–155.0	20.8±0.7	13.0–25.0[c]
tCO_2 (mmol/L)	37	21.9	3.0	16.0–28.0	NR	16–25[c]
pO_2 (mmHg)	36	77.1	29.9	29.9–166	NR	90–110[d]
sO_2 (%)	36	89.7	7.6	69–99	NR	>90[d]
Base excess (mmol/L)	37	−6.4	3.4	(−14)–0	−8.0±0.6	(−5)–(+2)[d]
Lactate (i-STAT) (mmol/L)	37	3.1	2.2	0.7–11.7	NR	0.5–2.7[e]
Lactate (Lactate Plus) (mmol/L)	57	2.7	1.8	0.8–11.8	NR	NR
BUN (mg/dL)	63	35	15	18–79	NR	10–38[e]
Creatinine (mg/dL)	63	0.5	0.2	0.2–1.0	NR	0.2–0.7[e]
Glucose (mg/dL)	63	167	38	87–282	NR	65–145[e]
CPK (U/L)	63	1,930	1,901	191–7,358	NR	50–450[e]
AST (U/L)	63	298	142	98–773	NR	12–43[e]
Hct (%)	72	43	6	30–56	NR	42–52[f]

[a] pCO_2 = partial pressure of carbon dioxide; HCO_3 = bicarbonate; tCO_2 = total carbon dioxide; pO_2 = partial pressure of oxygen; sO_2 = oxygen saturation; BUN = blood urea nitrogen; CPK = creatine kinase; AST = aspartate aminotransferase; Hct = hematocrit; n = total number of samples included. Not all values were obtained for every marten, and some martens were sampled more than once; NR = not reported.

[b] Venous blood gas values reported for polecats (*Mustela eversmanni*) (Gaylord et al. 2011).

[c] Feline venous whole blood (VetScan i-STAT 1 User Manual, Abaxis).

[d] Feline arterial whole blood (VetScan i-STAT 1 User Manual, Abaxis).

[e] Ferret serum (VetScan User Manual, Abaxis).

[f] Hct previously reported for American marten (Nieminen et al. 2007).

formed using a natural log for further analysis.

Blood values were evaluated using a priori models for their influence on lactate included BUN, creatinine, glucose, CPK, AST, and hematocrit. We used Pearson correlation to evaluate relationships between variables. The only highly correlated variables were CPK and AST (r=0.91) and body mass and sex (r=0.85). However, we used these parameters in the analysis because the correlation was not extreme (i.e., r observed was <0.95), and each added additional information (Burnham and Anderson 2002). The top models (ΔAICc <2.0) for predicting lactate with blood variables all included BUN (Table 3). The BUN had a positive relationship with lactate (from model averaging, β=0.02, 95% confidence interval [CI]: 0.00–0.04). Creatinine, glucose, CPK, AST, and HCT improved the model slightly but appeared as uninformative parameters in the top models since the 95% CI of the β values included 0 (Arnold 2010).

The variables InitialT, AmbientT, and TimeSince were the most informative marten-specific and environmental predictors of lactate levels (Table 4). Season was also included in the top model but was not considered since 95% CI for the β included a wide range around 0 (Table 4). The variables InitialT (β=0.06, 95% CI: –0.03–0.16), AmbientT (β=0.02, 95% CI: 0.00–0.04), and TimeSince (β=0.08, 95% CI: –0.06–0.22) were all positively related to lactate.

DISCUSSION

Restraint in the fabric cone for anesthetic induction was effective for almost all procedures (n=121), as martens readily entered the

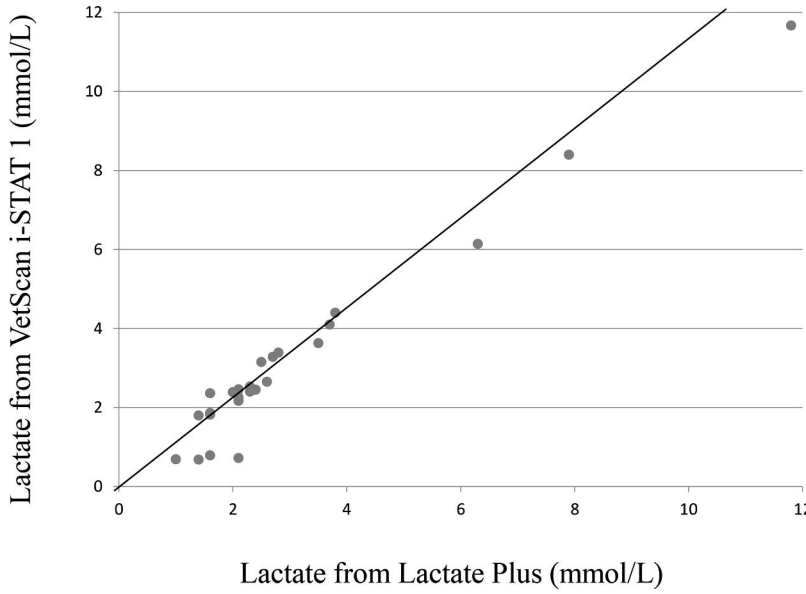

FIGURE 2. Bland-Altman analysis to compare lactate measured by two methods from free-ranging American martens (*Martes americana*) live-trapped and anesthetized using isoflurane under field conditions in Michigan, USA. Lactate was measured using a portable blood gas analyzer (VetScan i-STAT 1) with a CG4+ cartridge (analytes: pH, partial pressure of carbon dioxide, bicarbonate, total carbon dioxide, partial pressure of oxygen, oxygen saturation, lactate) and a portable lactate meter (Lactate Plus). The line of identity, where $y=x$, is indicated by a solid black line.

cone from the trap. Chamber induction was required for five procedures, and four of these procedures were for two animals that had been previously restrained in the fabric cone. It is suspected that they were more cautious of entering the cone than first-time captures.

In some instances, only brief sedation was required for placement of a microchip, and thus the cotton-ball method was attempted for induction in three animals. This method has been used for anesthesia of other species of small mammals (Parker et al. 2008; West et al. 2014). Subjectively, cotton-ball induction in our martens was rapid and deep, but concern for overdose precluded recommendation of this method without further research.

Desmarchelier et al. (2007) reported length of recovery in American martens (defined as the time between discontinuation of isoflurane and behavior considered ready for release) as 6.3±2.8 min. In our study, the mean time between discontinuation of the isoflurane and recovery to standing was 7±4 min, and the mean time to release after discontinuation of isoflurane was 15±6 min. Martens were released when they exhibited normal behavior and equipment was packed; thus release times do not necessarily reflect the earliest possible time that a marten could have been released. Our mean recovery time was shorter than that reported after nonreversible injectable anesthetics in American martens (Desmarchelier et al. 2007). Recovery time (defined as return to normal behavior) of black-footed ferrets (*Mustela nigripes*) anesthetized with isoflurane was 16.3±1.4 min, which was actually longer than when injectable anesthetics were used (Kreeger et al. 1998). One male marten in our study had a prolonged recovery of 48 min during which time he was conscious but recumbent, vocalized only when stimulated, and had a normal rectal temperature at the end of the procedure and normal blood glucose. After being empirically treated with subcutaneous fluids, oxygen, and oral dextrose, the animal was released and recaptured 18 mo later, at which time he had a normal recovery.

While the normal body temperature of the American marten is unknown, the reported

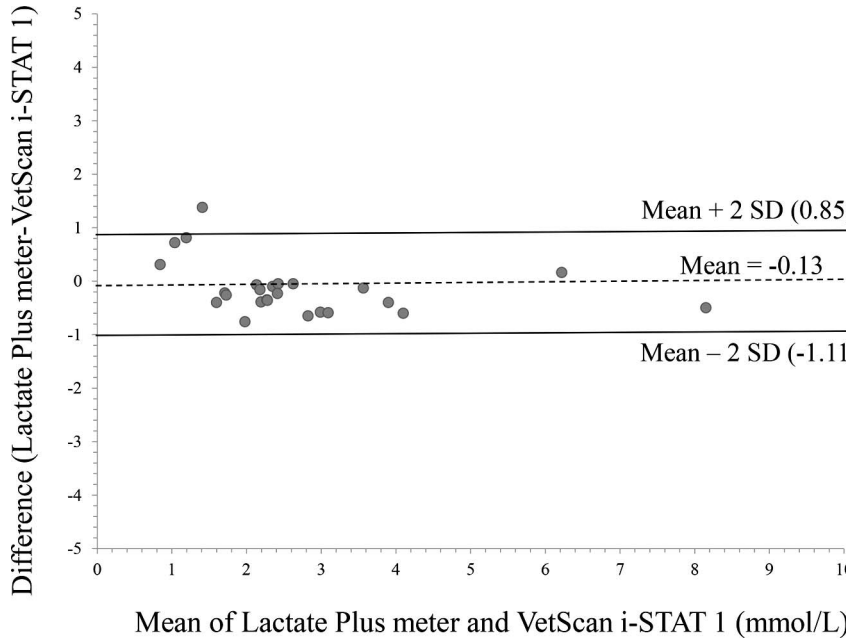

FIGURE 3. Bland-Altman analysis to compare lactate measured by two methods on free-ranging American martens (*Martes americana*) live-trapped and anesthetized using isoflurane under field conditions in Michigan, USA. Lactate was measured using a portable blood gas analyzer (VetScan i-STAT 1) with a CG4+ cartridge (analytes: pH, partial pressure of carbon dioxide, bicarbonate, total carbon dioxide, partial pressure of oxygen, oxygen saturation, lactate) and a portable lactate meter (Lactate Plus). The dashed line represents the mean of the differences between the Lactate Plus meter and VetScan i-STAT. The solid black lines represent the upper and lower 95% confidence limits. Only 2/26 values were outside the confidence limits and occurred when mean lactate <2.

TABLE 3. Models ranked by decreasing Akaiake Information Criterion for small sample sizes (AICc) of blood values affecting lactate[a] collected during 64 anesthetic events of free-ranging American martens (*Martes americana*) anesthetized using isoflurane after capture in Michigan, USA for radiocollaring and physiological assessment.[b]

Lactate model[c]	K	AICc	ΔAICc	Model weight
β_0+BUN+CREAT+Glucose+CPK+AST+*marten*+ε	8	72.9	0.0	0.30
β_0+BUN+CREAT+Glucose+CPK+AST+HCT+*marten*+ε	9	72.9	0.0	0.30
β_0+BUN+*marten*+ε	4	74	1.1	0.17
β_0+BUN+CREAT+*marten*+ε	5	74.2	1.3	0.16
β_0+BUN+CREAT+Glucose+*marten*+ε	6	76.7	3.8	0.04
β_0+CREAT+*marten*+ε	4	79.1	6.2	0.01
β_0+CPK +*marten*+ε	4	80.1	7.2	0.01
β_0+AST+*marten*+ε	4	80.5	7.6	0.01
β_0+Glucose+*marten*+ε	4	81.5	8.6	0.00
β_0+CREAT+Glucose+CPK+AST+*marten*+ε	7	83.1	10.2	0.00
β_0+CREAT+Glucose+CPK+AST+HCT+*marten*+ε	8	84.6	11.7	0.00
β_0+HCT+*marten*+ε	4	94.1	21.2	0.00

[a] Lactate was measured by two different analyzers (VetScan i-STAT 1 and Lactate Plus) and compared for clinical agreement. Mean of both measurements was used when available; otherwise, measurements from either the i-STAT or Lactate Plus were used.

[b] K = number of parameters; ΔAICc = relative difference between AICc of model and AICc of model with lowest AICc.

[c] β_0 = standardized regression coefficient; BUN = blood urea nitrogen; CREAT = creatinine; CPK = creatine kinase; AST = aspartate aminotransferase; *marten* = individual marten; ε = regression error term; HCT = hematocrit.

Table 4. Marten-specific and environmental values affecting lactate[a] in blood collected during 67 anesthetic events of live-trapped American martens (*Martes americana*) using isoflurane in Michigan, USA. Models were ranked by decreasing Akaiake Information Criterion for small sample sizes (AICc).

Lactate model[b]	K[c]	AICc	ΔAICc[c]	Model weight
β_0+InitialT+AmbientT+Season+*marten*+ε	6	88.6	0	0.38
β_0+InitialT+TimeSince+*marten*+ε	5	89.4	0.8	0.26
β_0+InitialT+*marten*+ε	4	89.7	1.1	0.22
β_0+Recovery+*marten*+ε	4	92.8	4.2	0.05
β_0+Body mass+Sex+Lactating+*marten*+ε	6	93.0	4.4	0.04
β_0+InitialT+AmbientT+Season+TimeSince+BrknNails +OralTrma+Recovery+Body mass+Sex+Lactating+*marten*+ε	13	93.9	5.3	0.03
β_0+InitialT+OralTrma+BrknNails+Collar+TimeSince+*marten*+ε	8	93.9	5.3	0.03

[a] Lactate was measured by two different methods (VetScan i-STAT 1 and Lactate Plus) and compared for clinical agreement. Both methods for lactate measurement gave similar results. The mean of both measurements was used when available; otherwise, measurements were used from either the i-STAT or Lactate Plus.

[b] β_0 = standardized regression coefficient; InitialT = rectal body temperature after initial induction with isoflurane; AmbientT = ambient temperature at time of induction; Season = summer (April through September) or winter (October through March); *marten* = individual marten; ε = regression error term; TimeSince = time between trap discovery and induction of anesthesia to within the closest 0.25 h; BrknNails = number toenails broken, worn, or frayed; OralTrma = recent or fresh oral trauma (present or absent); Recovery = time from discontinuation of isoflurane to standing; Collar = presence of a previously placed radiocollar at the time of capture.

[c] K = number of parameters; ΔAICc = relative difference between AICc of model and AICc of model with lowest AICc.

body temperature for the related fisher (*Martes pennanti*) is 38.8±0.8 C (International Species Information System 2002). Desmarchelier et al. (2007) reported American marten rectal temperatures of 34–40.1 C at an average of 12 min postinduction with isoflurane. Temperatures at subsequent time points were not reported. Captive pine martens (*Martes martes*) immobilized with medetomidine-ketamine had rectal temperatures ranging from 38.4 C to 40.1 C (Arnemo et al. 1994). Free-ranging American martens immobilized with a combination of tiletamine-zolazepam and xylazine had rectal temperatures ranging from 37 C to 39.3 C (Belant 2005).

As expected, hyperthermia occurred most often during summer months. Hyperthermia in anesthetized animals results in increased oxygen consumption and increased risk for serious complications, such as capture myopathy (Arnemo and Caulkett 2007). Increased initial temperature was also positively related with lactate values (AIC_c and model averaging; Table 4). Increased oxygen consumption associated with increased body temperatures may explain the significantly higher heart rates seen in the first 10 min of anesthesia during summer months compared to winter months. Anesthesia was discontinued for one marten with an initial temperature of 41.7 C. This animal was recaptured a week later and had an uneventful anesthesia.

While hypothermia occurred during winter months, it also happened during summer months likely due to inhalation of cool oxygen, respiratory evaporative heat loss, reduction in thermoregulation associated with anesthesia, and a large body surface area to volume ratio (Heard 2007). Severe hypothermia can result in prolonged anesthetic recovery (Heard 2007). One hypothermic marten in this study (final rectal temperature 34.8 C) had a longer than average time to standing and time to release of 18 and 27 min, respectively. Frequent monitoring of body temperature and proactive efforts to prevent ongoing heat loss are indicated for martens under isoflurane anesthesia.

Desmarchelier et al. (2007) reported heart rate (216 ± 17.2 beats per min; n=8), respiratory rate (31 ± 11.5 breaths per min; n=18), and relative arterial oxygen saturation via pulse oximetry (>95%; n not reported) of

American martens after an average of 10 min of isoflurane anesthesia. These are similar to reported vital rates in this study after 10–19 min of isoflurane anesthesia during both summer and winter (Table 1). Although pulse oximetry has not been validated for American marten, relative arterial oxygenation appears to be adequate in isoflurane anesthetized martens.

In contrast to old dental damage seen in 43.1% of martens examined, recent oral trauma including gingival abrasions (9.3%) or freshly fractured teeth (1.6%) occurred in a minority of procedures presumably due to biting at the trap. Similar to other small mustelids, martens use their canine teeth to kill prey by severing the spinal cord between vertebrae (Dayan et al. 1992). Dental damage was common in a review of large carnivore skull specimens, and individuals apparently functioned well despite damaged dentition (Van Valkenburgh 1988). In a study of 155 North American river otters (*Lontra canadensis*) captured with leghold traps, all otters had injuries ranging from superficial abrasions, missing claws, or digit injuries (Tocidlowski et al. 2000). Seventy-four percent of river otters obtained from a supplier for a reintroduction program had fractured teeth (Serfass et al. 1993). In contrast, trap-associated injury was uncommon in this study.

There were five instances of skin infection of the neck associated with transmitter collar wear. All five animals were subsequently retrapped and were found to have fully recovered. Given the length of a marten's neck and muscle relaxation achieved with anesthesia, it is important to consider how the collar will fit after recovery from anesthesia, a return to normal neck posture, as well as after potential weight gain. Similar complications and the use of collar break-away mechanisms have been reported previously in martens, and research into optimal collar material, size, and fit is warranted (Thompson et al. 2012).

Reference ranges for blood gas and serum biochemistry values are not available for the American marten. Using the domestic cat (*Felis catus*), another obligate carnivore, reference range provided by the manufacturer of the i-STAT analyzer (i-STAT User Manual, Abaxis), pH, pCO_2, HCO_3, and tCO_2, were within normal range under isoflurane anesthesia in this study (Table 2). Compared to venous blood gas values of polecats (*Mustela eversmanni*) anesthetized with isoflurane, the mean blood pH, HCO_3, and base excess were higher and pCO_2 lower in this report of martens under isoflurane, indicating better acid-base status and ventilation (Gaynor et al. 2011). The i-STAT manufacturer has not developed reference values for venous pO_2, venous sO_2, or base excess. We would expect lower levels for these values seen in marten venous samples as compared to arterial samples from domestic cats. The mean creatinine values for martens in this report were within the reference range for domestic ferrets (*Mustela putorius furo*) and domestic cats provided by the VetScan analyzer manufacturer. The mean BUN (35 mg/dL) was highly variable but above the upper end of the reference range for cats (10–30 mg/dL) and within the reference range for ferrets (10–38 mg/dL). The BUN values for 32% of martens were above the ferret reference range. Elevated BUN with normal creatinine may have been due to recent ingestion of raw meat bait or dehydration (Tripathi et al. 2011). Only one marten was azotemic with both an elevated BUN and creatinine, consistent with dehydration or renal dysfunction (Tripathi et al. 2011). Mean blood glucose was higher in martens than the ferret reference range and was likely attributable to transient excitement, catecholamine release, and/or stress (Evans 2011). Additionally, mean values for CPK and AST enzymes were also above ferret and cat reference ranges and could be a result of enzyme release from skeletal muscle associated with exertion (Hall and Bender 2011).

Lactate values from the Lactate Plus meter were previously reported to be higher than those from the i-STAT for the same sample, but correlation between each method and the reference method (Vitros LAC slide assay) was good (Karon et al. 2007). Comparison of the i-STAT and Lactate Plus meter using a Bland-Altman analysis showed that both methods provided similar clinical values. We

found the field measurement of lactate with body temperature and physical exam to be an objective measurement of the overall condition. The Lactate Plus meter and test strips were inexpensive, required only a drop of blood, and field personnel were able to easily perform the test. We found it to be a suitable alternative to the much more expensive i-STAT blood gas analyzer and cartridges.

In this study, average lactate values for live-trapped martens were just above the reference range for domestic cats with 12% of martens having lactate levels >5.0 mmol/L. Lactate production largely occurs under conditions of anaerobic cellular respiration or hypoxia (Allen and Holm 2008). Causes of tissue hypoxia include exercise, low arterial oxygen, anemia, and shock (Allen and Holm 2008). Lactate is cleared by the liver and kidneys, and its half-life depends on the cause of hyperlactatemia (Pang and Boysen 2007; Allen and Holm 2008).

The most parsimonious best fit model of lactate values using blood parameters was BUN (Table 3). Blood urea nitrogen is the by-product of protein metabolism and is excreted primarily by the kidneys (Tripathi et al. 2011). Exertion could lead to both an elevated lactate and increased BUN if an animal becomes dehydrated. Values for CPK and AST were not considered as important predictors of lactate, possibly because these enzymes take longer to rise and be metabolized than does lactate. Thus, the lack of relationship of lactate with CPK and AST may be due to metabolism of lactate during the in-trap time prior to blood sampling (Hall and Bender 2011). Lactate had a positive relationship with initial body temperature, ambient temperature, and time since discovery in trap until immobilization (Table 4 and model averaging); therefore, increased lactate likely reflected an increase in exertion particularly during higher temperatures. There was a weak positive but significant correlation between initial body temperature and ambient temperature. Time in trap was important but may have been a better predictor of lactate if we had known the actual time the marten entered the trap. A delay between finding the animal and inducing anesthesia may have resulted in the animal exerting itself, causing an increased lactate level. Alternatively, some martens created a nest in the trap and presumably rested until disturbed. This behavior and metabolism may have reduced lactate concentrations over time.

We considered martens with elevated lactate levels to be at higher risk for complications, and they were given subcutaneous fluids and monitored more closely during anesthesia. No animal in this study experienced mortality or overt signs of capture myopathy regardless of lactate level. We confirmed survival of martens with markedly elevated lactate levels within a week after release using telemetry.

Management of American martens in the NLP may require additional translocations making knowledge of anesthesia and physical findings useful to biologists and veterinarians. We recommend isoflurane anesthetic for field immobilization of martens as an effective and safe anesthesia at a wide range of ambient temperatures. When using isoflurane, one should be prepared to prevent and treat hypothermia and hyperthermia at any ambient temperature. Measurement of lactate could provide useful information to guide the treatment of individual animals and the refinement of capture and handling protocols. Future research should assess hydration status and measure physiologic state of trapped martens, and using trap timers would be useful in assessing time spent in a trap which can contribute to stress and changed blood values. We also recommend administering subcutaneous fluids during summer, limiting the number of traps to that which can be checked early in the day, having equipment ready before disturbing the animal, and minimizing noise and visual stimulation prior to immobilization.

ACKNOWLEDGMENTS

The authors thank the field crew, particularly C. Schumacher, T. Hillman, and M. Nichols. We thank D. Mulcahy, R. Wilkes, and R. Gerhold for

their contributions to this manuscript. We gratefully acknowledge the Evansville Zoological Society, Ulysses S. Seal Conservation Grant through the Minnesota Zoo, and PPG Sustainability & Conservation Fund through the Pittsburgh Zoo & PPG Aquarium for financial support. We acknowledge the Little River Band of Ottawa Indians Natural Resource Department, Sault Tribe of Chippewa Indians Natural Resource Department, Keweenaw Bay Indian Community Natural Resource Department, and the Detroit Zoo for their contributions to the fieldwork.

LITERATURE CITED

Allen SE, Holm JL. 2008. Lactate: physiology and clinical utility. *J Vet Emerg Crit Care* 18:123–132.

Arnemo JM, Caulkett N. 2007. Stress. In: *Zoo animal & wildlife immobilization and anesthesia,* West G, Heard D, Caulkett N, editors. Blackwell Publishing, Ames, Iowa, pp. 103–109.

Arnemo JM, Moe RO, Soli NE. 1994. Immobilization of captive pine martens (*Martes martes*) with medetomidine-ketamine and reversal with atipamezole. *J Zoo Wildl Med* 25:548–554.

Arnold TW. 2010. Uninformative parameters and model selection using Akaike's Information Criterion. *J Wildl Manage* 74:1175–1178.

Belant JL. 2005. Tiletamine–zolazepam–xylazine immobilization of American marten (*Martes americana*). *J Wildl Dis* 41:659–663.

Bland JM, Altman DG. 1986. Statistical methods for assessing agreement between two methods of clinical measurement. *Lancet* 327:307–310.

Burnham KP, Anderson DR. 2002. *Model selection and multimodel inference: A practical information-theoretic approach*, 2nd Ed. Springer, New York, New York, 488 pp.

Cooley TM, Schmitt SM, Friedrich PD, Bostick DP. 2004. American marten survey—2001. *State of Michigan Department of Natural Resources Wildlife Report No. 3471.* Michigan Department of Natural Resources, Lansing, Michigan, 3 pp.

Dayan T, Tchernov E, Simberloff D, Yom-Tov Y. 1992. Tooth size: Function and coevolution in carnivore guilds. In: *Structure, function and evolution of teeth*. Smith P, Tchernov E, editors. Freund Publishing House, Tel Aviv, Israel, pp. 215–222.

Desmarchelier M, Cheveau M, Imbeau L, Lair S. 2007. Field use of isoflurane as an inhalant anesthetic in the American marten (*Martes americana*). *J Wildl Dis* 43: 719–725.

Earle RD, Mastenbrook LH, Reis TF. 2001. Distribution and abundance of the American marten in northern Michigan. *State of Michigan Department of Natural Resources Wildlife Report No. 3321.* Michigan Department of Natural Resources, Lansing, Michigan, 26 pp.

Evans EW. 2011. Proteins, lipids, and carbohydrates. In: *Duncan & Prasse's veterinary laboratory medicine clinical pathology,* 5th Ed., Latimer KS, editor. Wiley-Blackwell, Ames, Iowa, pp. 173–210.

Fahlman Å, Arnemo JM, Persson J, Segerström P, Nyman G. 2008. Capture and medetomidine-ketamine anesthesia of free-ranging wolverines (*Gulo gulo*). *J Wildl Dis* 44:133–142.

Gaynor JS, Wimsatt J, Mallinckrodt C, Biggins D. 1997. A comparison of sevoflurane and isoflurane for short-term anesthesia in polecats (*Mustela eversmanni*). *J Zoo Wildl Med* 28:274–279.

Hall RL, Bender HS. 2011. Muscle. In: *Duncan & Prasse's veterinary laboratory medicine clinical pathology,* 5th Ed., Latimer KS, editor. Wiley-Blackwell, Ames, Iowa, pp. 283–287.

Heard D. 2007. Monitoring. In: *Zoo animal and wildlife immobilization and anesthesia*, West G, Heard D, Caulkett N, editors. Blackwell Publishing, Ames, Iowa, pp. 83–87.

International Species Information System. 2002. *Physiological data reference values, ferret body temperature*. International Species Information System, Bloomington, Minnesota. Compact disc available through www.isis.org.

Karon BS, Scott R, Burritt MF, Santrach PJ. 2007. Comparison of lactate values between point-of-care and central laboratory analyzers. *Am J Clin Pathol* 128:168–171.

Kreeger TJ, Vargas A, Plumb GE, Thorne ET. 1998. Ketamine-medetomidine or isoflurane immobilization of black-footed ferrets. *J Wildl Manage* 62:654–662.

Nieminen P, Rouvinen-Watt K, Saarela S, Mustonen A-M. 2007. Fasting in the American marten (*Martes americana*): A physiological model of the adaptations of a lean-bodied animal. *J Comp Physiol B* 177:787–795.

Pang DS, Boysen S. 2007. Lactate in veterinary critical care: Pathophysiology and management. *J Am Anim Hosp Assoc* 43:270–279.

Parker WT, Muller LI, Gerhardt RR, O'Rourke DP, Ramsay EC. 2008. Field use of isoflurane for safe squirrel and woodrat anesthesia. *J Wildl Manage* 72: 1262–1266

Powell RA, Buskirk SW, Zielinski WJ. 2003. Fisher and American marten. In: *Wild mammals of North America: Biology, management, and conservation*, Feldhammer GA, Thompson BC, Chapman JA, editors. Johns Hopkins University Press, Baltimore, Maryland, pp. 635–649.

R Development Core Team. 2013. *R: A language and environmental for statistical computing in R foundation for statistical computing.* www.r-project.org. Accessed May 2015.

Serfass TL, Peper RL, Whary MT, Brooks RP. 1993. River otter (*Lutra canadensis*) reintroduction in Pennsylvania: Prerelease care and clinical evaluation. *J Zoo Wildl Med* 24:28–40.

Thompson CM, Green RA, Sauder J, Purcell KL, Sweitzer RA, Arnemo JM. 2012. The use of radiotelemetry in research on *Martes* species: Tech-

niques and technologies. In: *Biology and conservation of American martens, sables, and fishers: A new synthesis*, Aubry KB, Zielinski WJ, Raphael MG, Proulx G, Buskirk SW, editors. Cornell University Press, Ithaca, New York, pp. 308–312.

Tocidlowski ME, Spelman LH, Sumner PW, Stoskopf MK. 2000. Hematology and serum biochemistry parameters of North American river otters (*Lontra canadensis*). *J Zoo Wildl Med* 31:484–490.

Tripathi NK, Gregory CR, Latimer KS. 2011. Urinary system. In: *Duncan & Prasse's veterinary laboratory medicine clinical pathology*, 5th Ed., Latimer KS, editor. Wiley-Blackwell, Ames, Iowa, pp. 274–278.

Van Valkenburgh B. 1988. Incidence of tooth breakage among large, predatory mammals. *Am Nat* 131:291–302.

West G, Carter T, Shaw J. 2014. Edentates (Xenarthra). In: *Zoo animal and wildlife immobilization and anesthesia*, 2nd Ed., West G, Heard D, Caulkett N, editors. Blackwell Publishing, Ames, Iowa, pp. 533–538.

Submitted for publication 17 June 2016.
Accepted 10 December 2016.