# Blood profile in green iguanas after short-term anaesthesia with propofol

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ABSTRACT: Blood haematology and plasma chemistry profiles were examined in a group of ten 17 monthsold green iguanas two minutes before propofol administration (10 mg/kg of body weight) and two minutes after recovery from anaesthesia. The induction time was very short (35.50  $\pm$  22.54 s), the recovery time was 21.50  $\pm$ 7.44 minutes. At five and fifteen minutes of anaesthesia there was a considerable drop in the heart rate. The most marked drop, however, was noted during the tenth minute. Following the administration of propofol green iguanas showed marked changes in the respiratory frequency that were not accompanied by any changes in the levels of  $SpO_{2}$ . RBC (1.06 ± 0.08 vs. 1.21 ± 0.06  $10^{12}$ /l) were decreased with a high significance (P < 0.01) two minutes after recovery from the propofol anaesthesia. At the same time there was a significant (P < 0.05) decrease of haemoglobin concentration (76.70  $\pm$  13.39 vs. 83.73  $\pm$  9.24 g/l) and PCV (0.34  $\pm$  0.04 vs. 0.36  $\pm$  0.04 l/l) as well as a significant (P < 0.05) increase of WBC (7.95 ± 3.38 vs. 5.20 ± 2.52  $10^9$ /l), heterophils (3.14 ± 1.48 vs. 1.43 ± 0.43  $10^9$ /l) and basophils  $(0.60 \pm 0.53 \text{ vs. } 0.23 \pm 0.17 \cdot 10^9 \text{/l})$ . The following parameters increased two minutes after anaesthesia with a high significance (P < 0.01): plasma concentration of total protein ( $55.12 \pm 5.94$  vs.  $49.02 \pm 3.54$  g/l), uric acid  $(231.07 \pm 77.69 \text{ vs. } 157.58 \pm 60.58 \, \mu\text{mol/l})$ , AST  $(1.23 \pm 0.52 \, \text{vs. } 0.67 \pm 0.34 \, \mu\text{kat/l})$ , TAG  $(3.37 \pm 1.11 \, \text{vs. } 1.48 \pm 1.41 \, \text{vs. } 1.48 \pm 1.41 \, \text{vs. } 1.48 \pm 1.41 \, \text{vs. } 1.48 \, \text{v$ 0.78 mmol/l), phosphorus (2.29  $\pm$  0.38 vs. 1.85  $\pm$  0.35 mmol/l). The increase in plasma calcium levels (3.51  $\pm$  0.11 vs.  $3.21 \pm 0.23$  mmol/l) was significant at the level of P < 0.05. All the measured values were within the reference range of healthy green iguanas.

Keywords: lizards; reptilian haematology; plasma chemistry; immobilization

Many injectable anaesthetics and their combinations may be used to induce immobilisation and sedation of reptiles (Lumb and Jones, 1984; Bennett, 1996; Heard, 2001; Knotek, 2004; Read, 2004; Knotek et al., 2005). Total intravenous anaesthesia (TIVA) has been infrequently used (Bouts and Gasthuys, 2002; Lock and Bennett, 2003). Contrary to this, there have already been reports of positive experiences with propofol (2,6-di-isopropylphenol) for some time (Lawton, 1992; Divers, 1996; Bennett et al., 1998). The requirement for intravenous administration is a major disadvantage for the use of propofol in reptiles. Young and small

reptiles can be particularly difficult to catheterize (Bennett, 1996). The main advantage of this drug in reptiles is its short duration of effect. If administered intravenously, propofol will start its activity in reptiles within one minute (McArthur et al., 2002). It produces short-term anaesthesia for about 20 min (Lloyd, 2003). Propofol is not painful for reptiles and the perivascular injection does not cause irritation or inflammation (Divers, 1996). High dosages of propofol can be associated with complications. Side effects of propofol include bradycardia and hypotension. Propofol anaesthesia is associated with prolonged apnoea in lizards.

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It is recommended that reptiles be intubated and given assisted ventilation after the administration of propofol to prevent hypoxemia and hypercapnia (Bennett et al., 1998). It is also recommended that propofol be used very carefully in reptiles suffering from chronic heart diseases and/or respiratory diseases. When evaluating anaesthetics in reptiles it is good to check blood parameters which characterize the inner environment and its state in the species being studied. There is sufficient data on haematological and biochemical profiles both in healthy green iguanas and clinical patients (Divers et al., 1996; Bruder, 1998; Harr et al., 2001; Knotek et al., 2002; Pejrilova, et al., 2004; Knotkova et al., 2005). The aim of this study was to evaluate selected blood parameters during short-term anaesthesia with propofol in the green iguana (Iguana iguana rhinolopha).

## MATERIAL AND METHODS

A group of ten 17 months-old green iguanas (*Iguana iguana rhinolopha*) from a captive breeding program were included in this trial (Table 1).

The lizards were housed in an experimental room and maintained in terraria with a specific regime of light, temperature and air humidity (Knotkova et. al., 2005). The reptiles had been fasted for 24 hours before the experiment. Propofol (Propofol 1% Fresenius, Fresenius Kabi GmbH, Germany) was administered into the ventral coccygeal vein (vena coccygea ventralis) as a bolus of 10 mg/kg with the use of Luer  $0.7 \times 30$  mm (Terumo, Belgium) hypodermic needles and 2 ml syringes (Braun, Germany). The iguanas were maintained in sternal recumbency on a paediatric electrical heating pad (heating level 2). Righting reflex was assessed at each time period and recorded as detected or not detected (Bennet et al., 1998). Induction time was defined as the interval from administration of propofol to loss of the righting reflex. Recovery time was defined as the interval from loss of the righting reflex to its return. A cloacal pulse oximeter probe (Vet/OX, SurgiVet, USA) was placed in the cloaca and positioned at the level of the terminal colon to enable the monitoring of functional haemoglobin oxygen saturation (SpO<sub>2</sub>) and heart rates during the first minute and then at five-minute intervals throughout the anaesthetic period.

Table 1. Selected clinical parameters in the green iguana during propofol anaesthesia

Values	Iguanas											
	1	2	3	4	5	6	7	8	9	10	$\overline{x}$	SD
Sex (m/f)	f	f	f	m	m	m	f	m	f	f	_	_
Weight (kg)	0.55	0.35	0.27	0.37	0.31	0.33	0.30	0.35	0.58	0.40	0.38	0.10
SCL (cm)	26.0	22.0	19.5	23.5	20.5	21.0	21.0	21.5	25.5	23.0	22.35	2.14
Induction time (s)	50	50	80	10	20	30	45	15	10	45	35.50	22.54
Recovery time (min)	20	20	25	38	23	17	10	15	22	25	21.50	7.44
Heart rate (at min 1)	82	81	81	87	98	103	98	98	89	75	89.20 <sup>a</sup>	9.52
Heart rate (at min 5)	77	83	78	81	98	100	96	96	80	72	86.10 <sup>b</sup>	10.28
Heart rate (at min 10)	80	82	80	82	92	95	97	96	80	70	85.40°	9.01
Heart rate (at min 15)	63	80	83	83	96	100	98	90	78	71	84.20 <sup>b</sup>	12.00
Heart rate (at min 20)	80	82	82	89	91	100	97	96	78	71	86.60	9.45
SpO <sub>2</sub> (at min 1)	70	99	92	90	82	83	85	82	90	91	86.40	7.88
SpO <sub>2</sub> (at min 5)	89	99	91	86	82	77	88	83	80	85	86.00	6.24
SpO <sub>2</sub> (at min 10)	90	89	89	89	78	83	86	90	85	89	86.80	3.88
SpO <sub>2</sub> (at min 15)	90	90	86	99	79	85	84	88	81	89	87.10	5.59
SpO <sub>2</sub> (at min 20)	89	99	89	90	79	90	89	89	79	90	88.30	5.76

a-bP < 0.05; a-cP < 0.01

Blood haematology and plasma chemistry profiles were compared in iguanas two minutes before propofol administration and two minutes after recovery from anaesthesia (Table 1). Blood (1 ml) was collected from the ventral coccygeal vein using the Omnican 0.30 × 12 mm insulin kit (Braun, Germany). Whole blood was placed into heparinized tubes (Leciva inj., Prague), centrifuged immediately, and plasma was removed and frozen (-20°C). The plasma was analyzed using an automated analyzer (CobasMira, Roche) for total protein (TP), glucose, uric acid, alkaline phosphatase (ALP), alanine aminotransferase (ALT), aspartate aminotransferase (AST), creatine phosphokinase (CK), cholesterol (Chol), triglycerides (TAG) and phosphorus (P). Plasma calcium concentrations were analyzed with an Atomspec analyzer (Hilger 1550). Haematocrit (PCV) was measured by microhaematocrit tubes. Haemoglobin (Hb) was determined by the cyanmethaemoglobin method, and total red and white blood cell counts were found out according to the Natt and Herrick method (Knotek et al., 2002; Pejrilova et al., 2004). Blood smears were air-dried and stained using May-Grunwald and Giemsa-Romanowski stains. Two hundred leukocytes were counted for each smear and classified as heterophils, eosinophils, basophils, lymphocytes, azurophils and monocytes (Pejrilova et al., 2004). To compare the heart rate and SpO2 of iguanas during different periods of the propofol anaesthesia the paired t-test was applied (Microsoft Excel software, Microsoft XP). Differences between the haemogram and plasma chemical values of iguanas before and after anaesthesia were compared using the paired *t*-test (Microsoft Excel software, Microsoft XP). The Grubbs' test was applied to

Table 2. Selected blood parameters in the green iguana before and after propofol anaesthesia

	Green iguanas ( $n = 10$ )								
Values	2 min bei	fore propof	ol administration	2	_				
	min.	max.	$\bar{x} \pm SD$	min.	max.	$\bar{x} \pm SD$	_		
Haemoglobin (g/l)	74.10	101.80	83.73 ± 9.24	55.60	93.00	76.70 ± 13.39	P < 0.05		
PCV (l/l)	0.32	0.41	$0.36 \pm 0.04$	0.28	0.39	$0.34 \pm 0.04$	P < 0.05		
RBC (10 <sup>12</sup> /l)	1.13	1.54	$1.21 \pm 0.06$	0.91	1.16	$1.06 \pm 0.08$	P < 0.01		
WBC(10 <sup>9</sup> /l)	2.00	10.50	$5.20 \pm 2.52$	2.50	10.50	$7.95 \pm 3.38$	P < 0.05		
Heterophils (10 <sup>9</sup> /l))	0.92	5.70	$1.43 \pm 0.43$	0.60	5.57	$3.14 \pm 1.48$	P < 0.05		
Eosinophils (10 <sup>9</sup> /l))	0	0.10	$0.03 \pm 0.04$	0	0.36	$0.01 \pm 0.02$			
Basophils (10 <sup>9</sup> /l)	0	0.51	$0.23 \pm 0.17$	0	1.47	$0.60 \pm 0.53$	P < 0.05		
Azurophils (10 <sup>9</sup> /l))	0	0.45	$0.31 \pm 0.17$	0	1.08	$0.45 \pm 0.34$			
Monocytes (10 <sup>9</sup> /l)	0	0.15	$0.06 \pm 0.05$	0	0.36	$0.07 \pm 0.14$			
Lymphocytes (10 <sup>9</sup> /l)	0.56	5.67	$2.79 \pm 1.60$	0.60	5.52	3.73 ± 1.61			
Total protein (g/l)	42.80	54.30	$49.02 \pm 3.54$	46.90	64.30	55.12 ± 5.94	P < 0.01		
Glucose (mmol/l)	8.09	13.43	$10.76 \pm 1.83$	8.07	14.30	11.17 ± 1.64			
Uric acid (µmol/l)	70.07	290.90	157.58 ± 60.58	129.00	384.50	231.07 ± 77.69	P < 0.01		
ALP (μkat/l)	0.42	1.10	$0.70 \pm 0.24$	0.16	0.87	$0.62 \pm 0.22$			
ALT (μkat/l)	0.11	0.35	$0.23 \pm 0.09$	0.16	1.32	$0.50 \pm 0.41$			
AST (µkat/l)	0.36	1.29	$0.67 \pm 0.34$	0.72	2.34	$1.23 \pm 0.52$	<i>P</i> < 0.01		
Creatine phosphokinase (µkat/l))	1.49	104.90	$32.45 \pm 37.95$	17.47	287.30	112.93 ± 95.22			
Cholesterol (mmol/l)	3.61	6.31	$5.05 \pm 0.93$	3.69	7.87	$5.00 \pm 1.33$			
TAG (mmol/l)	0.87	3.03	$1.48 \pm 0.78$	1.36	5.43	3.37 ± 1.11	<i>P</i> < 0.01		
Ca (mmol/l)	2.80	3.54	$3.21 \pm 0.23$	3.36	3.69	$3.51 \pm 0.11$	P < 0.05		
P (mmol/l)	1.46	2.50	$1.85 \pm 0.35$	1.80	2.85	$2.29 \pm 0.38$	<i>P</i> < 0.01		

exclude extremely low and high values (Matouskova et al., 1992).

## **RESULTS**

Table 1 presents the onset and duration of individual stages of anaesthesia. The induction time was very short. It was shorter than one minute for nine out of the ten iguanas (mean  $35.50 \pm 22.54$  s). The onset of anaesthesia in two iguanas was so quick that they were lacking reflexes within 10 seconds. All green iguanas were calm during the period of recovery. No signs of excitement or myoclonic activity were seen in iguanas in our study. Recovery in all iguanas was uneventful and complete within 22 minutes. Heart rates dropped considerably at five, ten and fifteen minutes of anaesthesia. The most marked drop was noted during the tenth minute. There was no significant difference in the heart rate of fully recovered iguanas twenty minutes after propofol administration, compared to the heart rate during the first minute. Despite marked respiratory rate changes in all green iguanas following the administration of propofol, there were no significant changes in SpO<sub>2</sub> values.

Comparing selected parameters in blood collected before and after the anaesthesia, we found significant differences in the concentration of haemoglobin, PCV, RBC and WBC (Table 2). Counts of heterophils and basophils were also significantly increased. The blood plasma was characterised by significantly increased concentrations of total protein, uric acid, TAG, calcium and phosphorus and increased activities of AST.

## **DISCUSSION**

Propofol used as an anaesthetic agent in lizards has a rapid onset of action. Following intravenous or intraosseous administration in green iguanas we may expect the onset of anaesthesia within several minutes (Bennett at al., 1998). Divers (1996) pointed out that induction times for propofol anaesthesia in reptiles were generally rapid, typically being less than a minute. Our results are in agreement with the above-mentioned data because the time period from the intravenous administration to the loss of the righting reflex was not longer than one minute. Propofol at a dose of 10 mg/kg of body weight resulted in marked changes in the

respiratory depth and frequency in the group of the studied green iguanas. It is known that periods of apnoea lasting up to five minutes are not exceptional. Although individual differences occur, it is recommended that propofol should be used in reptiles only when inhalation systems are available for the prospective resuscitation. Reptiles readily metabolize propofol without the risk of accumulation when used repeatedly and recovery is quick and calm (Lawton, 1992; Lock a Bennett, 2003).

Traditionally when dealing with patients undergoing anaesthesia, veterinary medicine pays attention to respiratory and heart rates, EKG parameters, tissue oxygen saturation and the CO<sub>2</sub> concentration in the expired gasses (ETCO<sub>2</sub>), blood pressure and body temperature (Bettschart-Wolfensberger et al., 2005). The anatomy of reptiles makes continuous monitoring of these parameters rather difficult, and therefore relatively few detailed studies have been performed to date (Apelt, 1993; Bennett et al., 1998; Mosley et al., 2003a,b). Heart rates and blood SpO<sub>2</sub> parameters, in particular, are sufficient for monitoring clinical practice (Diethelm et al., 1998). Our results document the fact that in five to fifteen minutes following the intravenous administration of propofol there are marked changes of heart rate in green iguanas. The oxygen concentration in peripheral blood (SpO<sub>2</sub>), however, does not vary much even when all green iguanas show changes in the respiratory rate and some of them undergo four- to five-minute periods of apnoea.

The short-term anaesthesia of ten green iguanas using propofol was safe. All haematological and biochemical parameters remained within the physiological range of healthy individuals (Harr et al., 2001; Knotek et al., 2002; Pejrilova et al., 2004; Knotkova et al., 2005). The lower concentration of haemoglobin, PCV and RBC could be explained by haemodynamic changes and re-distribution of blood cellular elements in the vascular bed. It is known that propofol induces moderate systemic hypotension, arterial vasodilatation and venodilatation (Branson and Gross, 1994). Significantly higher counts of leukocytes, heterophils and basophils, in particular, could be explained as a reaction to the stress due to triple manual handling and restraint as well as puncture of the ventral coccygeal vein. Propofol is also known to activate the systemic inflammatory response (Kumar et al., 2005).

The marked rise in the concentration of total protein, uric acid, calcium and phosphorus following anaesthesia may be explained as a consequence of the short-term restriction of blood flow through some organs. However, it was documented that liver blood flow was preserved with propofol and that renal and portal venous blood flows remained essentially unaltered with propofol anaesthesia in mammals (Wouters et al., 1995). The more than twofold rise in the concentration of TAG in the blood plasma may be a result of the induction with propofol. Hypertriglyceridaemia has recently been documented in intensive care patients receiving propofol in human hospitals (Devlin et al., 2005).

Reaction to the triple blood sampling from the ventral coccygeal vein causing some local muscle trauma may be responsible for the increased AST activity. Increased activities of creatine phosphokinase should be expected in such cases. Indeed, the mean level of CK during the second sampling was considerably higher than prior to the propofol anaesthesia. The difference, however, was not statistically significant due to the large standard error value. It is possible that this trend could be confirmed when studying a more numerous collection of reptiles. Blood collection through a permanent catheter (Heard, 2001) would be necessary to exclude repeated traumas to the tail muscles as a cause of increased activities of AST and CK. For technical reasons, these methods were not used in this study.

Further studies measuring haematological and biochemical parameters and their changes in the green iguana following propofol anaesthesia of longer duration are in progress.

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