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Effect of Capture Method on Hematological and Serum Biochemical Values of Red Deer (*Cervus elaphus*) in Turkey

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Abstract: Hematological and serum biochemical values were measured in blood samples collected from ten adult red deer (*Cervus elaphus*) which were captured physically or by the use of a combination of xylazine/ketamine. The investigated hematological values were white blood cell count, red blood cell count, hemoglobin, packed cell volume, mean corpuscular volume and platelets count. The serum values of glucose, total bilirubin, Blood Urea Nitrogen (BUN), creatinine, uric acid, cholesterol, Very Low Density Lipoprotein (VLDL), High Density Lipoprotein (HDL), triglycerides, Aspartate aminotransferase (AST), Alanine aminotransferase (ALT), Alkaline Phosphatase (ALP), Gamma-Glutamyl Transferase (GGT), Creatine Kinase (CK), Lactate Dehydrogenase (LDH), amylase, sodium, potassium, chloride, calcium, phosphorus, magnesium, Total Protein (TP), albumin and globulin were also measured. Heart and respiratory rates decreased in chemically captured animals, while rectal temperature did not fall in any group. The physically captured animals revealed significantly higher packed cell volume, mean corpuscular volume, glucose, triglycerides, aspartate aminotransferase, alanine aminotransferase, creatine kinase and lactate dehydrogenase than the chemically captured deer.

Key words: *Cervus elaphus*, chemical capture, biochemical values, hematological values, physical capture, serum, red deer

INTRODUCTION

Accurate assessment of the health and nutritional status of wild or semi-captive animals is important for their wellbeing. Normal hematological and serum biochemical values for several deer species are not available and the values in healthy or clinically ill deer are compared with baseline values from domestic small ruminants such as sheep and goats (Sahoo and Arora, 2002; Singh *et al*., 1988).

The capture and immobilization of wild deer is likely to be one of the most stressful events in their life as is clearly indicated by capture-induced changes in the hematological and biochemical blood parameters (Gupta *et al*., 2007; Marco and Lavin, 1999). Appropriate chemical immobilization can be employed to safely capture any species minimizing stress and the risk of injuries associated with other methods (Janicki *et al*., 2006). Numerous reports have described the use of xylazine (and ketamine) or tiletamine/zolazepam for anesthetizing cervids (Fernandez-Moran *et al*., 2000; Galka *et al*., 1999; Janovsky *et al*., 2000; Miller and Adams, 2003; Miller *et al*., 2004; Murray *et al*., 2000; Tsuruga *et al*., 1999). Xylazine has a wide safety margin but has been associated with prolonged induction, unreliable immobilization and at high dosages apnea and bradycardia when used alone (Murray *et al*., 2000; Wallingford *et al*., 1996). Ketamine can be used as a sole anesthetic agent in numerous species but volume restrictions often preclude its use for darting procedures. It can also induce excessive muscle rigidity and violent recoveries as side effects (Arnemo *et al*., 2005; DelGuidice *et al*., 1989; Ryeng *et al*., 2002; Tsuruga *et al*., 1999). Despite the limitation of these drugs as anesthetic agents alone, their use in combination can provide satisfactory anesthesia (Caulkett and Haigh, 2004; Janovsky *et al*., 2000).

To the best of the knowledge, there are no comparative studies related to hematological and serum biochemical values of red deer which were physically or chemically captured in Turkey. The present research was performed to provide data on these parameters in red deer kept in Bursa, Turkey.

MATERIALS AND METHODS

Animals: The study was conducted with animals kept in a deer conservation station (Yesiltarla Deer Conservation

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Station) of the Ministry of Forestry in a foothill area near the village of Kirazliyayla, Bursa, Turkey (40°11'N/29°04'E, 750 m elevation). There were a total of 56 red deer in the station and of these animals 10 adult red deer (5 male and 5 female) were selected and placed separately in a 10 ha area for this study. The animals were between one and 5 years old and the body weights were between 130-270 (mean 194±45) kg. The animals were apparently healthy as evidenced by normal appetite and alertness to external environment. The animals grazed naturally and additional pelleted food was also supplied water was available *ad libitum*. For the study, food was withheld from the animals 12 h prior to capture.

In the first phase of the study, the animals were captured by physical means according to standard techniques (Arora, 1988). In the second phase after a period of one mo, the same animals were captured by chemical means using the procedure described below. In both groups, physiological data (heart rate, respiratory rate and rectal temperature) were collected after immobilization. Rectal temperature was checked once after the first contact using a rectal digital thermometer (Kruuse, Marslev, Denmark). Heart rate was measured by cardiac auscultation and respiratory rate by direct observation. After the clinical examinations, blood samples were collected as described below.

Anesthetic drugs and dosages: Xylazine (Rompun®, Bayer, Leverkusen, Germany supplied in a sterile vial as a lyophilized powder containing 500 mg xylazine) were diluted in 5 mL ketamine (Ketamidor®, 100 mg mL^{-1}, Richter Pharma AG, Wels, Austria). The final mixture contained 100 mg xylazine and 100 mg ketamine per mL. The animals were darted with dartguns (3 mL dart with 1.5×38 mm needle) (Daninject, Boerkop, Denmark) at the upper hind quarter muscles from a distance of 20-25 m. Each animal received 1.08±0.18 mg kg^{-1} xylazine/ketamine combination.

Blood collection and analysis: Blood was collected with an 18-ga jugular venipuncture using Vacutainer blood collection system (BD Vacutainer™, Bioland, Eskisehir, Turkey). After collection into plain tubes for serum and EDTA-tubes for hematology, blood samples for serum collection were centrifuged within 2 h and sera were harvested. Samples for hematology were refrigerated and analyses were completed within 6 h. Hematological examination (White Blood Cell count [WBC], Red Blood Cell count [RBC], Hemoglobin concentration [Hgb], Mean Corpuscular Volume [MCV], Platelet count [PLT]) was performed by means of an automated analyzer (Cell-DYN 3500, Abbott Park, IL, USA). Packed Cell Volume (PCV) was measured with standard methods by using a hematocrit centrifuge (NF 815, Nuve, Ankara, Turkey) at 4000 rpm for 5 min to adjust values obtained with the analyzer.

Biochemical analyses were performed by using a chemistry analyzer (Aeroset, Abbott, UK). The investigated parameters were glucose, total bilirubin, Blood Urea Nitrogen (BUN), creatinine, uric acid, cholesterol, Very Low Density Lipoprotein (VLDL), High Density Lipoprotein (HDL), triglycerides, Aspartate aminotransferase (AST), Alanine aminotransferase (ALT), Alkaline Phosphatase (ALP), Gamma-glutamyl Transferase (GGT), Creatine Kinase (CK), Lactate Dehydrogenase (LDH), amylase, sodium, potassium, chloride, calcium, phosphorus, magnesium, Total Protein (TP), albumin and globulin.

Statistical analysis: All data are expressed as mean±Standard Deviation (SD). The physiological, hematological and biochemical values from both capture groups were compared by using Student's unpaired t-test and Mann-Whitney U-test when appropriate. Results were analysed using SPSS 13.0 computer software (SPSS Inc., Chicago, IL, USA). A $p<0.05$ was considered significant.

RESULTS AND DISCUSSION

It was possible to perform clinical examinations and blood sampling without problems in both groups. Xylazine/ketamine combination resulted in rapid sedation with complete laterally recumbent immobilization after a single dose for all animals captured by chemical means. No anesthetic-related problems were encountered.

Table 1 and 2 shown the physiological, hematological and biochemical values obtained from deer captured by physical and chemical means. A decrease in the

Table 1: Physiological and hematological values in red deer captured by physical and chemical means

Factors	Physical capture (n = 10)	Chemical capture (n = 10)
Heart rate (beat min^{-1})	28.22±1.56*	22.22±1.20
Respiratory rate (breath min^{-1})	13.44±1.33*	8.11±1.36
Rectal temperature (°C)	37.63±0.26	37.05±0.20
White Blood Cell (WBC) count (10^3 μL^{-1})	4500±560	4300±720
Red Blood Cell (RBC) count (10^6 μL^{-1})	9.80±1.46	9.04±1.69
Hemoglobin (Hgb) concentration (g dL^{-1})	12.60±1.10	12.40±2.2
Packed Cell Volume (PCV) (%)	49.40±3.40*	35.20±4.60
Mean Corpuscular Volume (MCV) (fL)	58.10±3.0*	39.90±3.40
Platelets (PLT) (10^3 μL^{-1})	870.80±196.30	749.50±180.10

Means in the same line with different superscripts are significantly different (*$p<0.05$)

Table 2: Biochemical values in red deer captured by physical and chemical means

Factors	Physical capture (n = 10)	Chemical capture (n = 10)
Glucose (mg dL^{-1})	134.25±23.9*	117.22±28.35
Total bilirubin (mg dL^{-1})	0.12±0.05	0.06±0.04
Blood Urea Nitrogen (BUN) (mg dL^{-1})	8.47±1.21	7.07±2.14
Creatinine (mg dL^{-1})	1.62±0.15	1.58±0.27
Uric acid (mg dL^{-1})	0.32±0.18	0.21±0.03
Cholesterol (mg dL^{-1})	55.44±14.93	50.25±19.63
Very Low Density protein (VLDL) (mg dL^{-1})	3.75±1.70	2.0±1.0
High Density Lipoprotein (HDL) (mg dL^{-1})	34.44±9.27	31.75±10.30
Triglycerides (mg dL^{-1})	18.50±7.41*	7.88±6.77
Aspartate aminotransferase (AST) (IU L^{-1})	173.50±29.39*	59.60±12.00
Alanine amimotransferase (ALT) (IU L^{-1})	64.75±15.21*	25.70±7.36
Alkaline Phosphatase (ALP) (IU L^{-1})	144.50±25.26	104.33±20.24
Gamma-Glutamyl Transferase (GGT) (IU L^{-1})	20.77±5.86	19.25±4.57
Creatine Kinase (CK) (IU L^{-1})	2285.75±845.73**	359.66±243.20
Lactate Dehydrogenase (LDH) (IU L^{-1})	511.75±93.89*	404.33±120.35
Amylase (U L^{-1})	31.88±29.27	17.75±2.75
Sodium (mmol L^{-1})	151.50±11.70	146.44±7.33
Potassium (mmol L^{-1})	5.32±0.61	4.90±0.40
Chloride (mmol L^{-1})	109.0±6.16	107.22±4.84
Calcium (mg dL^{-1})	8.61±1.10	8.62±1.12
Phosphorus (mg dL^{-1})	6.87±2.04	6.33±1.20
Magnesium (mg dL^{-1})	2.02±0.20	1.97±0.09
Total protein (g dL^{-1})	6.77±0.79	6.77±0.99
Albumin (g dL^{-1})	2.77±0.53	2.58±0.40
Globulin (g dL^{-1})	4.20±0.74	4.02±0.53

Means in the same line with different superscripts are significantly different (*$p<0.05$, **$p<0.01$)

heart and respiratory rates was seen in the chemically captured animals ($p<0.05$), while rectal temperature was similar in both groups. In the hemogram, WBC, RBC, Hgb and PLT values did not differ between red deer captured by physical and chemical means but PCV and MCV values were higher in the group of animals captured by physical means ($p<0.05$) (Table 1).

Regarding the biochemical constituents, the plasma activity of AST, ALT, LDH ($p<0.05$), CK ($p<0.01$) and the concentrations of glucose and triglycerides ($p<0.05$) were significantly higher in the group of animals captured by physical means. In this group, the mean plasma activity of AST and ALT were at least doubled and CK was approximately six times higher when compared with the other group. No significant differences were observed between the two methods of capture regarding the total bilirubin, BUN, creatinine, uric acid, ALP, GGT, amylase, sodium, potassium, chloride, calcium, phosphorus, magnesium and total protein levels. Physical capture group tended to have higher values for all other serum parameters mentioned above, except for calcium and total protein (Table 2).

Both physical and chemical methods are used to capture deer, the chosen method depending on the individual circumstances and desired objective (Jones, 1984; Marco and Lavin, 1999). In this study, physical and chemical immobilization of red deer were utilized in a state of semi-captivity in a distinct area. No difficulty was encountered during the assesment of physical parameters and blood collection in both capture methods. Heart rate and respiratory rate decreased in the chemically immobilized animals as reported previously for xylazine/ketamine (DelGuidice *et al*., 1989) and medetomidine/ketamine (Tsuruga *et al*., 1999) anesthesia, although contradictory results have also been reported (Galka *et al*., 1999).

Xylazine/ketamine combination is commonly used for the chemical immobilization of deer (Galka *et al*., 1999; Gupta *et al*., 2007; Haigh and Hudson, 1993; Nimitsuntiwong *et al*., 2000). The dose in the study was similar to doses used previously (DelGuidice *et al*., 1989; Galka *et al*., 1999; Janovsky *et al*., 2000) and provided a smooth and uneventful induction of immobilization with no side effects or mortality.

Change in several blood parameters is a common finding after both physical and chemical capture in deer (Cross *et al*., 1988; Montane *et al*., 2003). Studies show that stress induced by physical capture of animals results in more significant changes than those induced by chemical immobilization (Cross *et al*., 1988; Marco and Lavin, 1999). In the present study PCV and MCV were higher in the physical capture group, whereas WBC, RBC, Hgb and PLT values did not differ between the groups. Changes in PCV has been attributed to the contractions of spleen after catecholamine release during physical capture (Cross *et al*., 1988).

When compared with the chemically induced capture, physical capture resulted in higher levels of glucose, triglycerides, AST, ALT, CK and LDH most remarkable changes occurring in the AST, ALT and CK levels. The increase in glucose level can be attributed to the hyperglycaemic effect of catecholamines and glucocorticoids released during the stress involved in the physical capture group (Spraker, 1993).

Significant increases were observed at the enzymes indicator of muscle damage in the physically captured animals. This is probably the result of increased physical exertion during capture (Kock *et al*., 1987). CK and AST have been reported to reflect muscle damage more sensitively than LDH and ALT (Duncan and Prasse, 1986). In this study plasma AST, ALT, CK and LDH activities were greater in the physically captured animals but the most remarkable increase was found in the CK activity. When compared with other studies where

capture of animals was achieved physically, the CK values recorded in this study were higher (Marco and Lavin, 1999) but lower (Chapple *et al*., 1991) than those reported by other researchs.

Cholesterol concentration is normally strictly regulated in animals varying only slightly due to diet and the time of year (Bartley, 1989). Some researchers have found higher concentrations of cholesterol in animals captured by physical means and attributed this to the effect of catecholamines and corticosteroids (Marco *et al*., 1997; Marco and Lavin, 1999). In this study, cholesterol concentration was similar in both groups and was slightly lower than the values reported by other researchers (Marco and Lavin, 1999; Peinado *et al*., 1999). While cholesterol levels were similar in both groups, higher triglyceride levels were observed in the physically captured animals.

The results obtained in this research are in aggreement with those reported elsewhere about total bilirubin (Audigé, 1992). BUN and creatinine (Audigé, 1992; Marco and Lavin, 1999; Powell and DelGiudice, 2005), sodium (Knox *et al*., 1988; Marco and Lavin, 1999; Peinado *et al*., 1999), calcium (Audigé, 1992; Nimitsuntiwong *et al*., 2000; Peinado *et al*., 1999), magnesium (Peinado *et al*., 1999) and total protein (Kay, 1987; Peinado *et al*., 1999). However, lower concentrations of total bilirubin (Marco and Lavin, 1999), uric acid (Peinado *et al*., 1999), chloride (Arnemo *et al*., 1994; Marco and Lavin, 1999), phosphorus (Audigé, 1992; Nimitsuntiwong *et al*., 2000; Peinado *et al*., 1999) and albumin (Peinado *et al*., 1999) and higher levels of sodium (Wilson and Pauli, 1983) and potassium (Arnemo *et al*., 1994; Wilson and Pauli, 1983) were seen in the present study when compared with other reports.

CONCLUSION

This is the first study carried out in Turkey that establishes reference values and comparatively investigates the effect of physical and chemical capture in red deer. Most hematological values were similar in both capture methods, whereas higher PCV and MCV levels were observed after physical capture. Increases were observed in several serum enzyme levels of physically captured animals and these animals tended to have higher values for all serum parameters than the chemical capture group except for calcium and total protein. The results suggest that stressful effect of capture and handling must be considered when evaluating blood parameters.

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