TRANSPORT STRESS IN ROE DEER (*CAPREOLUS CAPREOLUS*): EFFECT OF A SHORT-ACTING ANTIPSYCHOTIC

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Abstract *Animal Welfare 2002, 11*: 405-417

*The aim of this study was to evaluate the effect of a short-acting antipsychotic (acepromazine) on the stress response to transport in roe deer (*Capreolus capreolus*). Twenty-one roe deer were submitted to a nine-hour road journey in order to reintroduce and restock this species into Catalonia (north-eastern Spain). The animals were divided into two groups: animals in the treatment group received an intramuscular injection of acepromazine (0.13 mg kg^{-1} in 0.5 ml; n = 9) while animals in the control group received the same volume of saline (n = 12). Clinical (heart rate and body temperature, measured during transport using remote devices), haematological, and biochemical indicators of stress were used to evaluate the effect of the antipsychotic. Heart rate increased during transportation, but no differences were found between groups. Body temperature decreased during transportation in both groups, but this reduction was faster in acepromazine-treated animals. Comparison of blood parameters before and after transport revealed significantly lower red blood cell counts and haemoglobin concentrations after transport in treated animals compared with control animals; a reduction in lymphocyte count, eosinophil count and serum creatinine levels over transport in treated animals; a decrease in serum potassium levels over transport in the control group; an increase in serum creatine kinase (CK), aspartate aminotransferase (AST), and alanine aminotransferase (ALT) activities over transport in control animals; an increase in serum urea and chloride concentrations over transport in both groups; and finally, a decrease in serum glucose concentrations in both groups. These results demonstrate the suitability of acepromazine in transport operations in order to reduce the stress response and prevent its adverse effects in roe deer.*

Keywords: *animal welfare, antipsychotic, road transport, roe deer, stress*

Introduction

The regular translocation of wildlife has become a common management practice. Transportation of animals is often essential for wildlife managers, zoo keepers and farmers, and in order to minimise welfare problems it is necessary that the best transport conditions are used.

There are a number of reviews in the scientific literature concerning the road transport of cattle (Tarrant 1990; Trunkfield & Broom 1990; Warriss 1990; Knowles 1995; Knowles 1999), sheep (Knowles 1998) and pigs (Warriss 1987, 1998a,b; Tarrant 1989). However,

only limited information exists on the effects of transport in wild and semi-wild animals, which may respond to handling in different ways to the more docile, domesticated species (Brelurut 1991).

Roe deer are valuable wild ungulates because of their ecological function as undergrowth clearers, and because of the economic importance of the game activity that they generate. The species is undergoing expansion, and is increasingly submitted to reintroduction and restocking operations (García-Ferré *et al* 1995) that require transportation. Several studies have been carried out into the behavioural and physiological responses of farmed red deer (*Cervus elaphus*) to road transport (Waas *et al* 1997; Grigor *et al* 1998; Waas *et al* 1999), but no studies have been carried out on roe deer. It is widely believed that short-acting tranquillisers are ineffective when the animals concerned are highly stressed at the moment of administration, particularly in free-ranging animals, but this needs to be further explored (Diverio *et al* 1996).

Capture myopathy, also called exertional rhabdomyolysis, is one of the main adverse consequences of stress in wild animals. Capture myopathy is a syndrome that occurs in wild (free-ranging and captive) mammals and birds, and is associated with the stress of capture, restraint and transportation. In ungulates, the syndrome is characterised clinically by depression, muscular stiffness, lack of coordination, paralysis, metabolic acidosis and death. Pathologically, capture myopathy resembles the myodegenerative disorders of domestic cattle, sheep, horses and swine (Chalmers & Barret 1982), and it is mainly characterised by muscular and renal lesions (Spraker 1993).

Acepromazine is a member of the phenothiazine group of short-acting antipsychotic agents. Although the exact mechanisms of action are not fully understood, the phenothiazines block post-synaptic dopamine receptors in the central nervous system, and may also inhibit the release of and increase the turnover rate of dopamine. They are thought to depress portions of the reticular activating system, which is important for the control of body temperature, basal metabolic rate, emesis, vasomotor tone, hormonal balance and alertness. In addition to their dopamine-blocking actions, phenothiazines have varying degrees of anticholinergic, antihistaminic, antispasmodic, and α-adrenergic-blocking effects. Their onset of action is fairly slow, averaging up to 15 min following intravenous administration (Plumb 1995). Cowan *et al* (1962) studied the use of acepromazine for tranquillising deer of the genus *Odocoileus*. The recommended dose for deer species is 0.05–0.1 mg kg^{-1} (Arnemo *et al* 1993). Our main reason for choosing acepromazine for this investigation was the widespread use of this drug in veterinary practice. Another possible choice would be one of the butyrophenones, but these present an increased risk of extrapyramidal side-effects (Poling *et al* 1990). Hofmeyr (1981) described extrapyramidal syndrome in several wild ungulate species treated with haloperidol.

Methods

Animals

Twenty-one roe deer were used in this study: 10 males (six adults, four fawns [less than one year old]), and 11 females (six adults, five fawns), captured by means of drive-nets in the National Game Reserve of Chizé in mid-western France. The mean liveweight of the subjects was 19.73 ± 1.3 kg (range 12.3–26.7 kg; n = 21).

Treatment groups

Nine randomly selected animals, four males (one adult, three fawns) and five females (three adults, two fawns), received intramuscularly a 0.13 mg kg^{-1} (± 0.013 SEM) dose of

acepromazine (Calmo Neosán®, SmithKline Beecham, Madrid, Spain) in a volume of 0.5 ml. Twelve control animals, six males (five adults, one fawn) and six females (two adults, four fawns), received intramuscularly 0.5 ml of saline (Fisiológico Braun®, Braun Medical S.A. Rubí, Barcelona, Spain).

Procedure

Animals were captured by means of drive-nets and were immediately placed in individual transport boxes designed for roe deer. They were then transported for 5 min in a van to the control centre of the game reserve. Once there, blood samples were withdrawn and the animals marked with ear tags, weighed and aged and then reintroduced into their transport boxes. In addition, the right thoracic and left precordial areas of certain animals were clipped in order to install the heart-rate recording equipment, and the body-temperature recording device was introduced into the rectum. The transport boxes were placed in the van, where the animals remained until the onset of road transport ('pre-transport phase', 25 min). Transportation lasted 9 h and was mainly on the motorway. A second blood sample was taken upon arrival and the recording equipment removed.

This procedure was repeated twice, the overall study being carried out in two parts. The first part took place in January 1999 with 10 animals being transported, and the second part took place in March 2001 using 11 animals.

Equipment

Ten roe deer (five per group) were fitted with a telemetric heart-rate recording device (Polar Vantage NV™, Polar Electro Oy, Finland). The device comprised a transmitter and a receiver. The transmitter was attached to a girth belt supplied by the manufacturer for use by athletes. The two electrodes forming the transmitter were covered with electrode coupling gel to maintain a good electrical contact. The heart-rate signal was telemetrically transmitted within a range of 1 m to the wrist monitor, which was placed around the neck of the animal. Heart rate was measured at 60 s intervals throughout the journey. Using the manufacturer's software program, the data were transferred later for further analysis.

Thirteen roe deer (seven in the treatment group, six in the control group) were fitted with a telemetric body-temperature recording device (Mätman Datalogger®, Chipsobits Eltex AB, Sweden). Rectal temperature was measured at 90 s intervals, and the total recording period lasted for 5.5 h along the journey. Using the manufacturer's software program, the data were transferred later for further analysis. Ambient temperature during capture and transport was 6–12°C.

Blood collection and analyses

Two blood samples were taken, one before and one after transportation. Blood samples were obtained using disposable syringes and 0.8 × 25 mm needles. Blood collected from the jugular vein was placed in a tube with EDTA K_3 as an anticoagulant, and used for haematological analyses. The remainder of the blood was placed in a serum collection tube, allowed to clot at room temperature, and used for biochemical analyses. Serum was kept at –18°C until all analyses were completed. All serum samples were analysed at the same time after a three-month period. Samples from the control and treated groups were treated equally. Haematological examinations of red blood cell (RBC) count, haemoglobin concentration, mean corpuscular volume (MCV), mean corpuscular haemoglobin concentration (MCHC), mean corpuscular haemoglobin (MCH) and white blood cell (WBC) count were performed by means of a semi-automatic analyser (Sysmex F-800, Toa Medical Electronics, Japan).

Packed cell volume (PCV) was measured using the standard microhaematocrit method with a haematocrit centrifuge (Micro-Haematocrit Centrifuge, Hawksley, Lancing, UK) at 11,000 rpm for 5 min to adjust the values obtained with the analyser. Differential leukocyte count was performed using blood smears stained with commercial Diff-Quick® stain (Química Aplicada SA, Amposta, Spain). Biochemical analyses were performed using an automated analyser (COBAS MIRA, Roche, Nutley, NJ, USA) except for sodium and potassium concentrations, which were measured using flame photometry (Corning 410C, Corning Medical, Medfield, USA), and serum cortisol, which was determined using an ELISA commercial kit (DRG Cortisol EIA-1887, DRG Diagnostics, Germany).

Statistical analyses

Repeated measures ANOVAs were carried out using the PROC MIXED procedure of the SAS® statistics software package (SAS Institute Inc, Cary, NC, USA). The main factor was treatment (acepromazine or saline) and the repeated factor was time. The animals' sex, age and transport group (first or second transport procedure), and their interactions, were included in the statistical model. A type 1 autoregression (AR[1]) structure for the covariance matrix of the repeated measures was used. Least squares means (LS MEANS) were used because the distribution of animals between groups was unbalanced. In all cases, the accepted significance level was $P < 0.05$.

Results

Heart rate did not differ significantly between groups. It decreased significantly ($P < 0.05$) during the pre-transport period (Figure 1) and increased during the transport period (Figure 2) in both groups. Body temperature (Figure 3) decreased over time in both groups, but it reached baseline levels 2 h earlier ($P < 0.05$) in those animals that received acepromazine than in the control animals.

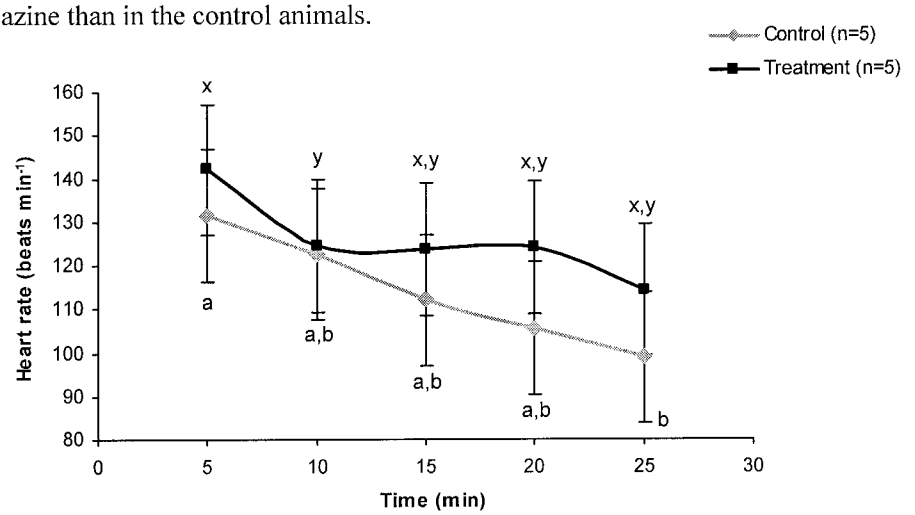

Figure 1 **Heart rate (mean ± SEM) during pre-transport period for control and treated roe deer. [a,b]Values with different superscripts are significantly different from each other in the control group ($P < 0.005$). [x,y]Values with different superscripts are significantly different from each other in the treatment group ($P < 0.005$).**

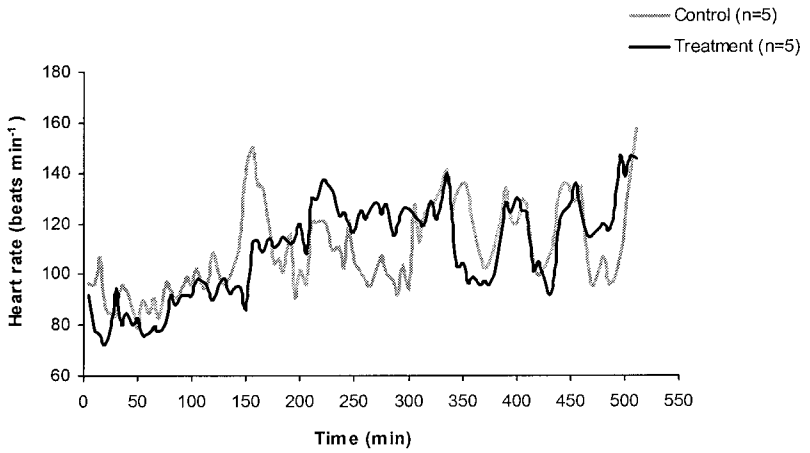

Figure 2 **Mean heart rate over transport for control and treated roe deer.**

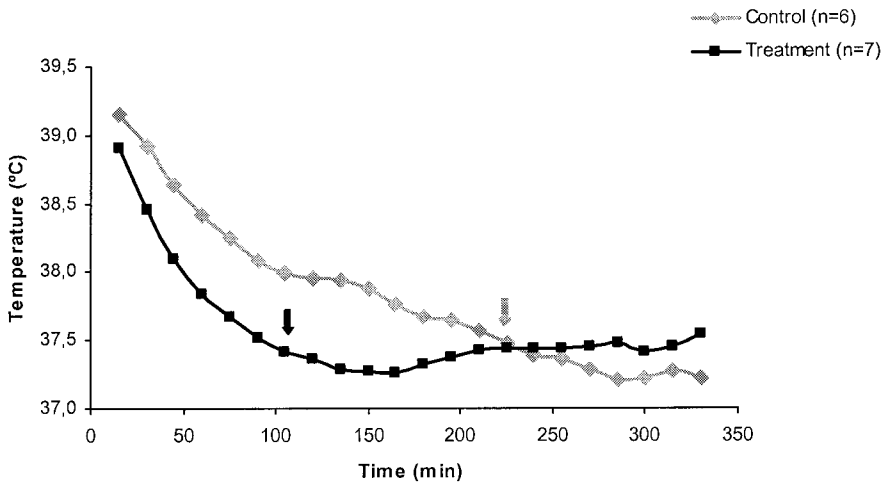

Figure 3 **Body temperature (°C) over transport from control and treated roe deer. Arrows indicate stabilisation of body temperature (ie no statistical differences were found between that measurement and those registered later).**

Haematological parameters are shown in Table 1. RBC count and haemoglobin concentration in samples taken from treated animals after transportation were significantly lower ($P < 0.05$) than those obtained from control animals. Lymphocyte count decreased over transport ($P < 0.05$) in roe deer treated with acepromazine. Eosinophil count also decreased over transport ($P < 0.05$) in treated animals.

Serum biochemical parameters are shown in Table 2. Serum CK, AST ($P < 0.05$) and ALT ($P < 0.001$) activities increased over transport in the control group, whereas in the treated animals they did not change. Serum lactate dehydrogenase (LDH) activity did not show statistical differences. Urea concentrations increased over transportation ($P < 0.05$) in both groups of roe deer. Serum creatinine concentrations decreased significantly over

transportation ($P < 0.05$) only in the control group. Serum glucose levels declined in both groups during transport. Serum chloride concentrations increased significantly ($P < 0.05$) during transportation in both groups. Serum potassium levels decreased over transportation ($P < 0.05$) only in the control group. No significant differences between groups were observed in serum cortisol levels.

Table 1 Haematological values (mean ± SEM) of control and treated animals before and after transportation.

	Before transport		After transport	
Parameters	***Control*** (n=12)	***Treatment*** (n=9)	***Control*** (n=12)	***Treatment*** (n=9)
Red blood cells (x 10^{12} l^{-1})	12.42 ± 0.35^{a}	$11.37 \pm 0.40^{a,b}$	12.40 ± 0.35^{a}	10.91 ± 0.40^{b}
Packed cell volume (l l^{-1})	0.44 ± 0.013	0.45 ± 0.015	0.46 ± 0.013	0.43 ± 0.015
Haemoglobin (g l^{-1})	$166.91 \pm 4.12^{a,b}$	$163.56 \pm 4.72^{a,b}$	169.98 ± 4.12^{a}	156.56 ± 4.72^{b}
MCV (fl)	35.94 ± 0.91^{a}	40.07 ± 1.05^{b}	37.15 ± 0.91^{b}	39.42 ± 1.05^{b}
MCHC (g dl^{-1})	37.62 ± 0.84	36.15 ± 0.96	36.98 ± 0.84	36.64 ± 0.96
MCH (pg)	13.45 ± 0.21^{a}	14.43 ± 0.25^{b}	13.52 ± 0.21^{a}	14.38 ± 0.25^{b}
White blood cells (x 10^{9} l^{-1})	4.817 ± 0.403	4.900 ± 0.465	4.533 ± 0.403	4.955 ± 0.465
Differential leukocyte count				
Lymphocytes (x 10^{9} l^{-1})	1.959 ± 0.168^{a}	2.037 ± 0.193^{a}	$1.612 \pm 0.168^{a,b}$	1.407 ± 0.198^{b}
Monocytes (x 10^{9} l^{-1})	0.083 ± 0.021	0.097 ± 0.025	0.084 ± 0.021	0.113 ± 0.025
Neutrophils (x 10^{9} l^{-1})	2.659 ± 0.371	2.609 ± 0.429	2.757 ± 0.371	3.431 ± 0.429
Eosinophils (x 10^{9} l^{-1})	0.159 ± 0.042^{a}	0.178 ± 0.048^{a}	$0.045 \pm 0.042^{a,b}$	0.016 ± 0.048^{b}
Basophils (x 10^{9} l^{-1})	0.004 ± 0.003	0.00 ± 0.004	0.00 ± 0.003	0.005 ± 0.004

^{a,b}Values with different superscripts are significantly different from each other ($P < 0.05$); values without superscripts are not significantly different from each other.

Table 2 Blood biochemistry (mean ± SEM) of control and treated animals before and after transportation.

	Before transport				After transport			
Parameters	***Control***		***Treatment***		***Control***		***Treatment***	
ALT (IU l^{-1})	$122.75 \pm 33.89^{a*}$	(n=12)	$89.07 \pm 39.45^{a,b}$	(n=8)	$173.42 \pm 33.89^{b*}$	(n=12)	$108.89 \pm 39.14^{a,b}$	(n=9)
AST (IU l^{-1})	1288 ± 1003^{a}	(n=12)	$2563 \pm 1177^{a,b}$	(n=8)	2104 ± 1003^{b}	(n=12)	$2317 \pm 1165^{a,b}$	(n=9)
CK (UI l^{-1})	$15\,084 \pm 4981^{a}$	(n=12)	5738 ± 5938^{a}	(n=8)	$22\,108 \pm 4981^{b}$	(n=12)	$8403 \pm 5784^{a,b}$	(n=9)
LDH (IU l^{-1})	1675 ± 477	(n=12)	2012 ± 593	(n=8)	2872 ± 477	(n=12)	1872 ± 553	(n=9)
ALP (IU l^{-1})	68.91 ± 15.84	(n=11)	92.40 ± 18.61	(n=8)	83.32 ± 15.49	(n=12)	92.59 ± 18.22	(n=9)
Urea (mmol l^{-1})	10.25 ± 1.60^{a}	(n=12)	$10.98 \pm 1.99^{a,c}$	(n=8)	$15.11 \pm 1.60^{b,c}$	(n=12)	16.81 ± 1.91^{b}	(n=9)
Creatinine (µmol l^{-1})	$121.11 \pm 5.75^{a,b}$	(n=11)	126.5 ± 6.74^{a}	(n=8)	$124.64 \pm 5.75^{a,b}$	(n=12)	118.37 ± 6.74^{b}	(n=8)
Lactate (mmol l^{-1})	2.97 ± 0.71	(n=11)	3.67 ± 0.83	(n=8)	5.39 ± 0.69	(n=12)	4.15 ± 0.83	(n=8)
Total bilirubin (µmol l^{-1})	4.87 ± 1.44	(n=12)	3.57 ± 1.44	(n=8)	6.58 ± 1.44	(n=12)	6.53 ± 1.44	(n=8)
Glucose (mmol l^{-1})	10.03 ± 0.95^{a}	(n=12)	11.75 ± 0.95^{a}	(n=8)	7.23 ± 0.95^{b}	(n=12)	6.81 ± 0.95^{b}	(n=8)
Total proteins (g l^{-1})	72.80 ± 2.74	(n=11)	71.81 ± 2.74	(n=8)	75.06 ± 2.84	(n=10)	74.21 ± 2.84	(n=9)
Cholesterol (mmol l^{-1})	1.20 ± 0.08	(n=12)	1.16 ± 0.08	(n=8)	1.28 ± 0.08	(n=12)	1.10 ± 0.08	(n=8)
Triglycerides (mmol l^{-1})	0.37 ± 0.06^{a}	(n=11)	0.59 ± 0.06^{b}	(n=8)	0.28 ± 0.06^{a}	(n=11)	0.30 ± 0.06^{a}	(n=8)
Sodium (mmol l^{-1})	148 ± 2.73	(n=10)	146 ± 2.73	(n=8)	145.8 ± 3.61	(n=6)	146.5 ± 3.61	(n=9)
Chloride (mmol l^{-1})	$104.37 \pm 1.12^{a,c}$	(n=11)	102.55 ± 1.12^{a}	(n=8)	107.74 ± 1.16^{b}	(n=10)	$106.46 \pm 1.16^{b,c}$	(n=8)
Potassium (mmol l^{-1})	6.27 ± 0.25^{a}	(n=11)	$5.55 \pm 0.25^{a,b}$	(n=8)	5.41 ± 0.27^{b}	(n=10)	5.26 ± 0.27^{b}	(n=9)
Cortisol log (ng ml^{-1})	1.71 ± 0.12	(n=11)	1.89 ± 0.12	(n=8)	1.62 ± 0.12	(n=10)	1.77 ± 0.12	(n=9)

^{a,b,c}Values with different superscripts are significantly different from each other ($P < 0.05$; $^{*}P < 0.001$); values without superscripts are not significantly different from each other.

Discussion

Heart rate is one of the most widely used acute stress indicators (Broom & Johnson 1993) and is considered to be an objective way of assessing the autonomic nervous system's response to psychological stressors (Hopster & Blockhuis 1994). Measurement of heart rate can be a useful measure of the emotional response of an individual to short-term challenges,

provided that a distinction is made between the metabolic and emotional effects and that the measurement itself does not cause too much disturbance (Broom & Johnson 1993).

The lack of difference in heart rate between the groups could be due to reflex tachycardia secondary to hypotension caused by acepromazine (Plumb 1995). Diverio *et al* (1996) found a greater increase in heart rate in red deer treated with a long-acting phenothiazine than in untreated deer during the 30 min immediately following stressor application, which was attributed to reflex tachycardia. Kock *et al* (1987) did not find significant differences in heart rate between normal bighorn sheep (*Ovis canadensis*) and those considered stressed.

The observed increases in heart rate during transportation are in agreement with results obtained by Horalek and Jones (1993) in red deer, who attributed such increases to the movement of the vehicle rather than noise or confinement. On the other hand, some researchers have reported decreases in red deer heart rates during transportation (Waas *et al* 1997; Grigor *et al* 1998), which were associated with their ability to adapt to the movement of the vehicle over time. This disparity could result from different road transport conditions (mainly road-type differences). Our results were obtained from transport conducted along a motorway, which can be considered the best road type in welfare terms, and, in spite of this, roe deer's heart rate increased during transport. In sheep, an increase in heart rate has been described after loading which remained high for the first 9 h of travelling (Parrot *et al* 1998).

Porges (1985) proposed that, instead of the basic heart rate, the variability in heart rate may be a better indicator of both the status of the individual's nervous system and its capacity to respond to environmental demands. The coefficient of variation of heart rate, used as a measure of heart-rate variability (Hopster & Blokhuis 1994), was not significantly altered by acepromazine (control group, $36.70 \pm 6.18\%$; treated group, $27.11 \pm 8.17\%$ [mean ± SEM]).

Increases in body temperature in certain stressful situations can be explained not only by physical activity, but also by stress-induced hyperthermia (SIH; Moe & Bakken 1997; Bakken *et al* 1999). SIH results from a regulated shift of the thermoregulatory set-point (Kluger *et al* 1987; Briese & Cabanac 1990) mediated by prostaglandin E and interleukins 1 and 6 (LeMay *et al* 1990; Kent *et al* 1993). Correlations have been found between SIH, the sympathoadrenal medullary system and the hypothalamic–pituitary–adrenal axis, which agrees with the proposal that SIH is a stress-mediated response (Groenink *et al* 1994). Zethof *et al* (1994) stated that, in mice, SIH is time-dependent, taking 10 min to reach a (stable) high level which is 1.0–1.5°C higher than baseline, and then taking 60 min to return to baseline. Moe (1996) found that SIH in farmed silver foxes (*Vulpes vulpes*) lasts 60–90 min after presentation of a short stressor. It has been suggested that SIH may be elicited in response to the anticipation of a known or unknown unpleasant event, indicating that SIH may reflect a state of anticipatory anxiety (Lecci *et al* 1990).

Although acepromazine can cause an alteration in thermoregulatory mechanisms (Plumb 1995), this cannot explain the differences between groups, as body temperature followed the same progression in both groups and then stabilised at the same baseline level. Therefore, it can be inferred that acepromazine does not suppress SIH, but improves adaptation to the new situation by accelerating temperature reduction. Taking into account that hyperthermia plays a very important role in the pathophysiology of capture myopathy (Chalmers & Barret 1982), it also can be suggested that acepromazine reduces the tissular necrosis risk induced by a prolonged augmentation of body temperature.

The first stage of the stress response is the activation of the sympathetic nervous system, stimulating the adrenal medulla and releasing catecholamines. Increases in RBC count,

haemoglobin concentration and PCV are associated with splenic contraction caused by the action of catecholamines on α-adrenergic receptors located in the splenic capsule (Ganong 1990), and are also partly attributable to a reduction in plasma volume (Wesson *et al* 1979; Cross *et al* 1988). In our study, changes in RBC count and haemoglobin concentration can be explained by acepromazine's α-adrenergic-blocking effect. This provokes relaxation of the spleen and the consequent splenic sequestration of erythrocytes (Turner & Hodgetts 1960; Jain 1993). Haemodilution caused by acepromazine due to lowering of blood pressure can be ruled out because, if this was the case, total proteins and sodium concentrations would also have decreased (Table 2). Although it has been reported that acepromazine decreases PCV (Bush 1993), no differences were found between groups in our study; this parameter did, however, show a decreasing trend similar to that shown by the RBC count and haemoglobin concentration (Table 1). The lack of significant effect could be attributable to the greater variability associated with PCV measurement using the microhaematocrit method, for which the coefficient of variation is 2% (Wintrobe 1974).

Total and differential leukocyte counts also respond to a variety of stimuli, including capture and transport. Catecholamines released during the alarm phase (the first stage of the stress response; see Selye 1946) may be responsible for the initial neutrophilia (increase in the number of neutrophils in the blood) and lymphocytosis (increase in the number of lymphocytes). Corticosteroids released during the resistance phase (the second stage of the stress response) contribute further to neutrophilia, but may cause a decrease in the lymphocyte count (lymphopenia). In domestic animals, the neutrophilia and lymphopenia peaks appear after 4–8 h exposure to stress (Duncan & Prasse 1986; Jain 1993). In red deer sedated with xylazine, Cross *et al* (1988) attributed the decrease in lymphocyte count to the splenic sequestration of lymphocytes, but in a later work (Cross *et al* 1989) they observed the same lymphopenia in splenectomised animals. The common mechanism of lymphopenia after corticosteroid exposure is redistribution of lymphocytes to bone marrow or body compartments. Long-term corticosteroid exposure may cause lympholysis (Schultze 2000). A possible explanation for the differences observed in lymphocyte count in our study is a delay caused by acepromazine in stress-induced lymphopenia. The mechanism of the decrease in eosinophil numbers is uncertain but is believed to be caused by intravascular lysis (steroid-induced apoptosis of eosinophils), decreased release from bone marrow, sequestration in organs such as the spleen and liver and increased tissue migration (Jain 1993; Young 2000). Therefore, the decrease in eosinophil count over transport observed in the treated animals is difficult to explain, although an alteration in eosinophil distribution could have occurred because of acepromazine. The great variability obtained in the eosinophil count could also be responsible for these results.

Muscle enzyme activity (CK, AST, LDH and ALT) increases during capture and handling operations because of increased muscle cell permeability or muscle cell damage (Duncan & Prasse 1986). These enzymes appear elevated in many stressed wild ungulates and in those suffering from capture myopathy (Kock *et al* 1987; Vassart *et al* 1992). Some authors have found that CK and AST levels are the most sensitive indicators of muscular disorders (Chapple *et al* 1991). When muscle activity begins, blood flow increases but is intermittent. Blood flow decreases as muscle contracts because of the compression of vessels, and increases during relaxation — a process called the 'muscle pump' (Guyton & Hall 1996). The muscle pump is active when the animal is running but it is inactive when it is immobilised by physical or chemical restraint or is standing in a crate. In most situations, the muscles of frightened animals that are not running are in a relatively high state of contraction, which hinders blood flow into the muscles. This leads to poor tissue perfusion,

decreased heat dissipation, and hypoxia (Spraker 1993). Catecholamines released in response to stress are potent vasoconstrictor agents that reduce the blood supply to muscles, thus causing hypoxia lesions. Results obtained from this study indicate that acepromazine exerts a protective effect against muscle damage because of its vasodilative effect and demonstrate its importance as a preventive treatment for rhabdomyolysis, as previously indicated for horses (Beech 1994).

Any process which increases protein catabolism will tend to result in increased levels of serum urea (Knowles & Warriss 2000). The increase of urea concentrations in our study was probably due to physical exercise, to the effects of glucocorticoids on protein catabolism, and also to food deprivation (Finco 1997).

Serum creatinine levels can be used to assess renal function. In some ungulates, however, increased creatinine concentrations, resulting from muscular activity and decreased renal excretion because of vasospasm in the kidney caused by catecholamines, have been described (Harthoorn 1976). The decrease in serum creatinine concentrations during transportation in the control group can be explained by the α-adrenergic-blocking effect of acepromazine on renal arterioles, where it promotes vasodilatation and thus allows the continued filtration and excretion of creatinine (Jarvik 1970). Moreover, this implies that oxygen supply to the kidneys was not impaired, thus reducing the risk of renal hypoxia and consequent renal necrosis.

Both increases and decreases in lactate concentrations have been reported as a result of transport. Because lactate concentrations vary greatly during the course of the transport period, measured values will depend on the timing of the blood sampling (Waas *et al* 1997). Our results did not show statistical differences in serum lactate levels between the treatment and control groups, nor over time; however, in a study of normal thoroughbred horses, 7 mg of acepromazine given intravenously 20 min before exercise resulted in lower serum lactate concentrations after exercise than in horses not administered the drug. This was attributed to a protective effect of acepromazine resulting from its vasodilative action (Freestone *et al* 1991; Beech 1994).

The decrease in serum glucose levels over transport in both groups may be due to food deprivation during transport (Knowles & Warriss 2000), which would correspond with the increased serum urea concentrations. The absence of group differences in serum glucose levels is in agreement with results obtained by Brearley *et al* (1990) comparing cattle treated with either saline or acepromazine submitted to 5 min of transport.

Serum sodium concentration did not change during transport (Table 2). This corresponds with results obtained by Grigor *et al* (1998) in red deer transported for 6 h. Changes in sodium levels are associated with variations in plasma volume, and, given that in this study total protein concentrations did not change either, it can be stated that there were no signs of dehydration. Changes in chloride concentration which are not associated with similar changes in sodium concentration are usually associated with acid–base imbalances. A disproportionate increase in chloride, as was seen in our study, is most commonly associated with normal to low hyperchloremic metabolic acidosis, and may be seen as a compensatory response to primary respiratory alkalosis (Carlson 1997).

Serum potassium levels decreased over transportation in the control group whereas no change was observed in the treated group. These differences cannot be explained by glucocorticoid action, which reduces plasma potassium levels (Verde & Gascón 1987), because there were no significant differences in cortisol concentrations between groups. It has been suggested that by influencing electrolyte movements, acepromazine may alter

neuromuscular excitability and impair the development of exertional rhabdomyolysis (Harris & Snow 1986; Freestone *et al* 1991).

Glucocorticoid levels rise in response to many short-term challenges in life and their measurement gives valuable information about the welfare of animals (Broom & Johnson 1993). Glucocorticoid hormones, produced in and released from the cortex of the adrenal glands in response to an extremely wide range of stressors, play a major role in mediating the physiological stress response and are widely interpreted as a measure of an animal's psychological perception of a situation. Many authors have reported significant increases in cortisol associated with transport (red deer [Smith & Dobson 1990; Brelurut 1991; Waas *et al* 1997; Grigor *et al* 1998]; cows [Kenny & Tarrant 1987]).

The lack of differences in serum cortisol levels in our study could be due to the fact that sedative effects are unrelated to plasma cortisol concentrations. Brearley *et al* (1990) found that at a similar depth of sedation, xylazine suppressed the cortisol response to stress whereas acepromazine had a slight potentiating effect. It has been suggested that chlorpromazine causes systemic release of adrenaline, which may result in an increase in adrenocorticotrophic hormone (ACTH) release and hence cortisol release (Bruss 1980). Other reasons for the lack of differences between groups could be the great inter-individual differences in stress-induced plasma cortisol concentrations (Moberg 1985) and the low number of individuals per group available in this study.

Animal welfare implications

Although this is a small study, the results show that during transport of roe deer, acepromazine is suitable for reducing the stress response and preventing its adverse effects. These benefits arise not only because of the sedative effect of acepromazine, but also because it causes peripheral vasodilation. This vasodilation has a protective effect against the muscular and renal damage which can arise from stress episodes in wild animals and which is directly involved in the pathogenesis of capture myopathy. Thus, animal welfare is improved by preventing and reducing the adverse effects of stress and by accelerating adaptation to new situations.

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