

Reversible chemical immobilization of wild red deer (*Cervus elaphus* L.) using tiletamine-zolazepam - xylazine hydrochloride mixture

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ABSTRACT

Fifteen live-trapped red deer (*Cervus elaphus* L.) were immobilized during January 2002 in the Baranja region of Croatia. The main purpose of the manipulation was the first individual treatment of red deer against trematode *Fascioloides magna*. Tiletamine-zolazepam - xylazine hydrochloride combination (Zo-Ro) was used in this trial for immobilization. Safe and efficient immobilization of red deer calves was achieved by administration of 2.0 ± 0.29 mg/kg of tiletamine-zolazepam (Mean \pm SD) and 2.76 ± 0.85 mg/kg of xylazine hydrochloride (Mean \pm SD). Mean doses (SD) for complete immobilization of adults were 1.9 (0.22) mg/kg of tiletamine-zolazepam and 2.24 (0.64) mg/kg of xylazine hydrochloride. Mean (SD) induction period was 5.88 (2.17) min for calves and 5.1 (2.6) min for adults. Recovery was induced by administration of atipamezole. Mean dose (SD) of atipamezole was 0.14 (0.04) mg/kg for calves and 0.10 (0.02) mg/kg for adults. Mean recovery time was 12.25 min for calves and 13.2 min for adults. There were no complication or death cases during the immobilization or after it.

Key words: red deer, chemical immobilization, tiletamine-zolazepam, xylazine hydrochloride, atipamezole

Introduction

In order to establish physical contact with wild animals, so essential for treatment, transportation and scientific purposes, humans have developed different methods for their physical restraint. Developing knowledge on this field has proved that capture of wild animals is an extremely stressful condition (DAWKINS and GOSLING, 1992), giving the advantage to methods that minimize stress. Appropriately used, chemical immobilization can be employed to safely restrain and capture any species, minimizing stress and the risk

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of injuries associated with other methods (NIELSEN, 1999). These characteristics enabled chemical immobilization to become a widely used tool of wildlife researchers. Different drugs can be used solely or in combination, the main demand being to achieve safe immobilization for both animals and humans. However, each drug carries a potential risk for animals. Many scientists have concluded that a combination of drugs is more effective and safer for immobilization, through a short and calm induction period, showing good myorelaxation and minimal influence on cardio-respiratory performance (KREEGER et al., 1996; FERNANDEZ-MORAN et al., 2001; ROFFE et al., 2001; CATET et al., 2003; ONUMA, 2003). Another important fact is that different species prefer different drug combinations. Since 1960, and nicotine-salicylate-gallamine mixture (SCHLOETH et al., 1960), several different drugs and their combinations have been used in the chemical immobilization of red deer with varying degrees of success. In this paper we present chemical immobilization of wild red deer with tiletamine-zolazepam - xylazine hydrochloride mixture (Zo-Ro), administered by two application methods. Similar research studies are encouraged in wildlife species to ensure the most appropriate drug(s) and dosages for safe immobilization.

Materials and methods

During January 2002 we trapped fifteen wild red deer (*Cervus elaphus* L.) near the village of Tikveš in the Baranja region of Croatia for individual therapy against large American liver fluke (*Fascioloides magna*). For trapping purposes we used a stable wooden facility (corral) for physical restraint and manipulation. Usually, the trapping object comprises two parts: the larger part of object is approximately 15 m in diameter, with fence reaching 2.5 m in height; the second part of object is made in a shape of a tunnel. Chemical immobilization of each deer was a prerequisite for the anthelmintic procedure. Drugs were administered manually to 14 animals while they were in narrow tunnel, at the muscles of upper hind quarters (i/m). Only in one case, a two-year-old male, we used a dart gun (Dist-Inject®, Peter Ott AG, Basel, Switzerland) with 3 ml darts (Dist-Inject®, Peter Ott AG, Basel, Switzerland), due to his aggressive behavioural display. This deer was also darted in the upper hind quarters. To achieve chemical immobilization as well as to test the safety of Zo-Ro mixture [Zoletil® (tiletamine-zolazepam, Virbac, Carros, France) and Rompun® 20% (xylazine-hydrochloride, Bayer, Leverkusen, Germany)] we used different dosages (mg/kg). We measured induction time as a period from drugs administration to complete immobilization (from injection to lying with head down). After the induction time each deer was taken from the tunnel and physically restrained. Following immobilization the animals were weighed with transportable scale (Pocket balance, Germany). To prevent corneal drying we applied an ophthalmic ointment (Chloramphenicol ung., Pliva d.d., Croatia) to deer's eye. To keep the animal calm a blindfold was used to cover the eyes. For reversal immobilization after therapy procedure each animal received a dose of atipamezole

(Antisedan[®], Pfizer, Karlsruhe, Germany) immediately after the anthelmintic procedure (approximately 20 min after drug administration). Therefore, the effect of atipamezole was monitored simply through safe recovery. In the case of protracted recovery time, antagonization was supported by administration of pentetrazole (Pentavet[®], Veterina d.o.o., Croatia). During therapy procedure and recovery time all animals were monitored visually. Recovery time was measured from atipamezole administration to first attempt at standing. Results were analysed using SPSS for Windows 6.1.

Results

Eight out of fifteen trapped animals were red deer calves aged 8 months, respectively. Body mass of adult deer ranged between 86 and 125 kg, while in calves it ranged from 60 to 84 kg. The solution of tiletamine-zolazepam and xylazine hydrochloride (Zo-Ro) was administered to 8 calves and 7 adult deer. Statistical data on different parameters of immobilized red deer calves are presented in Table 1. Mean (SD) dose to achieve an effective immobilization was 2.76 ± 0.85 mg of xylazine hydrochloride per kg of body mass (bm) and 2.0 ± 0.29 mg of tiletamine-zolazepam/kg of bm. Volume of the administered drug combination ranged from 0.8 to 1.6 mL per animal. The lower dose of drug combination and individual mass of animals corresponds with longer induction time, i.e. the lowest dose of drug combination (volume 0.8 mL) corresponds to longest induction time (9.0 min).

Table 1. Different parameters and statistical data on chemically immobilized red deer calves (n = 8)

N ^o	Sex	Mass (kg)	Xylazine-hydrochloride (mg/kg)	Tiletamine-zolazepam (mg/kg)	Induction time (min)	Atipamezole (mg/kg)	Recovery time (min)
1.	F	78	1.3	1.37	9	0.09	11
2.	M	84	2.3	2.12	8	0.11	18.5
3.	F	66	2.4	2.16	8	0.15	7.5
4.	F	68	2.3	2.1	6	0.14	10
5.	F	72	3.3	1.98	4	0.20	18
6.	F	60	4.0	2.38	4	0.16	10
7.	F	72	3.3	1.98	4	0.10	20
8.	M	74	3.2	1.92	4	0.13	3
Range (Min-Max)		60 - 84	1.3 - 4.0	1.37 - 2.38	4 - 9	0.09 - 0.2	3 - 20
Mean (SD)		71.75 (7.36)	2.76 (0.85)	2.0 (0.29)	5.88 (2.17)	0.14 (0.04)	12.25 (6.00)

Table 2. presents statistical data on different parameters of red deer adults immobilized with Zo-Ro mixture. Two additional animals, Nos. 6 and 7 required redosing and were not evaluated statistically. Mean (SD) dose to achieve effective immobilization was 2.24 ± 0.64 mg of xylazine hydrochloride/kg of bm and 1.9 ± 0.22 mg of tiletamine-zolazepam/kg of bm. Volume of administered drug combination ranged from 1.4 to 2.4 mL per animal. To antagonize effects of Zo-Ro mixture both categories received similar Mean dose of atipamezole (0.10-0.14 mg/kg). A significant ($P < 0.05$) reduction in induction time (Mean value) was observed in red deer calves immobilized with a higher content of xylazine (Table 3.). However, this was not confirmed in the case of adult animals.

Table 2. Doses of xylazine hydrochloride, tiletamine-zolazepam and atipamezole and their effect on induction and recovery time in red deer adults (n = 5)

N ^o	Sex	Mass (kg)	Xylazine-hydrochloride (mg/kg)	Tiletamine-zolazepam (mg/kg)	Induction time (min)	Atipamezole (mg/kg)	Recovery time (min)
1.	F	125	1.6	2.0	5	0.08	10
2.	F	86	2.3	1.66	9	0.11	12
3.	F	96	2.0	1.86	2.5	0.10	12
4.	M	96	3.3	2.23	3	0.13	12
5.	F	120	2.0	1.78	6	0.08	20
Range (Min-Max)		86 - 125	1.6 - 3.3	1.66 - 2-23	2.5 - 9	0.08 - 0.13	10 - 20
Mean (SD)		104.6 (16.9)	2.24 (0.64)	1.9 (0.22)	5.1 (2.6)	0.10 (0.02)	13.2 (3.89)

Table 3. Parameters on induction and recovery time in red deer calves and adults immobilized with low and high xylazine content

Parameter	Low xylazine group		High xylazine group	
	Calves	Adults	Calves	Adults
Induction time (min)	$7,75 \pm 1,26^a$	$4,50 \pm 1,80^a$	$4,13 \pm 0,25^b$	$6,00 \pm 3,00^a$
Recovery time (min)	$11,75 \pm 4,73$	$14,00 \pm 5,29$	$12,75 \pm 7,80$	$12,03 \pm 0,06$

a, b: significant differences were found between means with no common superscripts ($P < 0.05$)

Discussion

Chemical immobilization of large wild ungulates can be induced by different drugs individually or in combination. The main prerequisite for each manipulation with wild animals is to fulfil all safety requirements for both animals and personnel, with a sufficiently

long duration of chemical immobilization to ensure that animals will be found in forested areas and that some procedure can be performed on it (NIELSEN, 1999). One of the normally used and at the same time effective and safe drug combinations in deer game is 1:1 Zo-Ro solution, composed of equal quantities of tiletamine-zolazepam and xylazine hydrochloride (JANOVSKY et al., 2000). According to our results, even higher dosages of administered Zo-Ro mixture (mg/kg) in comparison to mean dose of $2.4 \text{ mg/kg} \pm 2.3 \text{ mg/kg}$ (Zo-Ro) used by JANOVSKY et al. (2000), had no negative effects on animals' condition. Furthermore, as opposed to etorphine-acepromazine (Immobilon®, C-Vet Ltd., Bury St. Edmunds, UK) or even medetomidine hydrochloride (Domitor®, Pfizer, Karlsruhe, Germany), xylazine hydrochloride and tiletamine-zolazepam are not lethal for humans in small dosages and also possess wide safety margins (CARRUTHERS et al., 1979; LIN et al., 1993), reducing the risk of drug accidents in humans.

It is also possible to alter the original 1:1 ratio of components in the Zo-Ro solution. Accordingly, MILLSPAUGH et al. (1995) used 2.5 mg/kg of Telazol® (in fact Zoletil®) plus 0.3 mg/kg of xylazine hydrochloride to achieve safe immobilization of Rocky Mountain elk. Our results, however, suggest that efficacy and safety of the Zo-Ro solution will be also preserved in the case where higher xylazine hydrochloride dosages are used. These higher xylazine hydrochloride doses are more suitable as they will reduce the cost of chemical immobilization (xylazine hydrochloride is less costly than tiletamine-zolazepam). Preserved efficacy of Zo-Ro solution in the case of an altered original ratio is visible mainly in the case of immobilization of red deer calves (Mean mass $71.75 \pm 7.36 \text{ kg}$). Mean value of induction period in calves was 5.88 min and is achieved by Mean doses of 2.76 mg/kg (xylazine hydrochloride) and of 2.0 mg/kg (tiletamine-zolazepam). Mean induction period for adults was 5.1 min. Both values are clearly shorter than those provided by JANOVSKY et al. (2000). Two adult females required redosing to achieve adequate immobilization, as they remained conscious even 12 minutes after first injection. As it was impossible to evaluate correctly the induction time for these animals we did not include them in the statistical analysis.

To antagonize the effects of Zo-Ro mixture, adult animals received 0.10 mg/kg of atipamezole, respectively. The antagonism effect was rapid in both categories of animal (Mean recovery time 13.2 in adults and 12.25 in calves). Higher dosages of atipamezole in calves (Mean 0.14 mg/kg) have reduced Mean recovery time in comparison with adult animals. Furthermore, one of the females that required redosing received a maximal volume of Zo-Ro mixture and recovered within 13 min after administration of 0.19 mg/kg of atipamezole. This, together with other results, indicates atipamezole as a good antagonist for Zo-Ro mixture. In the case of two protracted recovery times (one calf and one female) a dose of pentetrazole (5 mL) was given to each animal. This administration resulted in rapid antagonization.

Capture of free-ranging ungulates is usually followed by a mortality rate ranging from 0% to 26% (BERGERUD et al., 1964; HOUSTON, 1969; JOLICOEUR and BEAUMONT, 1986; PEREZ et al., 1997; KILPATRIK and SPOHR, 1999; JANOVSKY et al., 2000). The fact that we recorded no complications or death cases either during immobilization procedure or after it is partially attributable to the good characteristics of the Zo-Ro mixture for immobilization of wild ungulates, and partially to animals having been trapped by a stable wooden facility (corral), and physical restraint. A combination of these methods ensures better positioning for proper dose estimation, as well as a reduction in unnecessary chasing. Chasing, together with the capture and stress, is one of the most important factors included in initiation of so significant capture myopathy (WILLIAMS and THORNE, 1996).

Finally, we can conclude that Zo-Ro mixture is a safe and effective drug combination for immobilization of wild red deer. Different responses to administered dosages in wild animals have been reported previously (NIELSEN, 1999), which corresponds with our results (i.e. two females required redosing). Therefore, drug combinations with wide safety margins should be used. We recommend the use of Zo-Ro mixture with a higher content of xylazine hydrochloride, especially for short-term manipulation. A higher ratio of xylazine hydrochloride will not influence the efficacy of Zo-Ro mixture but will reduce the costs involved in chemical immobilization. Atipamezole is an effective antagonist for Zo-Ro mixture.

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SAŽETAK

Tijekom siječnja 2002. godine na području Baranje, Republika Hrvatska, kemijski je imobilizirano 15 prethodno uhvaćenih jelena običnih (*Cervus elaphus*). Jeleni su bili uhvaćeni radi pojedinačnog tretiranja protiv metilja *Fascioloides magna*. Kao sredstvo imobilizacije upotrijebljena je kombinacija tiletamin-zolazepama i ksilazin-hidroklorida (Zo-Ro). Sigurna i učinkovita kemijska imobilizacija jelenske teladi ostvarena je aplikacijom $2,0 \pm 0,29$ mg/kg tiletamin-zolazepama (srednja vrijednost \pm standardna devijacija) i $2,76 \pm 0,85$ mg/kg ksilazin-hidroklorida. Srednja doza (standardna devijacija) sredstva za potpunu imobilizaciju odraslih jelena iznosila je 1,9 (0,22) mg/kg tiletamin-zolazepama i 2,24 (0,64) mg/kg ksilazin-hidroklorida. Srednja vrijednost (standardna devijacija) vremena potrebnog za ulazak u imobilizaciju (indukcija) iznosila je 5,88 (2,17) minuta za telad i 5,1

(2,6) minuta za odrasle. Izlazak iz imobilizacije potaknut je aplikacijom atipamezola. Srednja doza (standardna devijacija) atipamezola bila je 0,14 (0,04) mg/kg za telad, odnosno 0,10 (0,02) mg/kg za odrasle jelene. Srednje vrijeme izlaska iz imobilizacije iznosilo je 12,25 minuta za telad, odnosno 13,2 minuta za odrasle. Tijekom postupka imobilizacije i neposredno nakon njega nisu uočene komplikacije niti uginuća.

Ključne riječi: jelen obični, kemijska imobilizacija, tiletamin-zolazepam, ksilazin-hidrokorid, atipamezol
