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Authors: Ferreras, Pablo, Aldama, Juan J., Beltrán, Juan F., and Delibes, Miguel

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## IMMOBILIZATION OF THE ENDANGERED IBERIAN LYNX WITH XYLAZINE- AND KETAMINE-HYDROCHLORIDE

Pablo Ferreras, Juan J. Aldama, Juan F. Beltrán, and Miguel Delibes

Estación Biológica de Doñana, CSIC, Apdo. 1056, E-41080 Sevilla, Spain

**ABSTRACT:** A combination of the dissociative anesthetic ketamine hydrochloride (KH) and the sedative xylazine hydrochloride (XH) was used to immobilize 31 wild Iberian lynx (*Felis pardina*) 45 times at Doñana National Park, Spain. A mean ( $\pm$ SE) dose of 4.6 ( $\pm$ 0.2) mg/kg KH and 4.0 ( $\pm$ 0.2) mg/kg XH resulted in mean ( $\pm$ SE) induction time of 5.6 ( $\pm$ 0.3) min and mean ( $\pm$ SE) first reaction time of 59.3 ( $\pm$ 6.5) min. Convulsions occurred four times (9%), but with no noteworthy consequences.

**Key words:** *Felis pardina*, Iberian lynx, immobilization, ketamine hydrochloride, recommended doses, Spain, xylazine hydrochloride.

### INTRODUCTION

Chemical immobilization of wild animals often is necessary for research or management purposes (Kreeger et al., 1986). It is of special importance to know the suitable anesthetic combinations and the dosages that provide an immobilization of adequate length and safety for endangered species. We describe the successful use of a combination of ketamine hydrochloride and xylazine hydrochloride in the immobilization of the Iberian lynx (*Felis pardina*), recognized as the most threatened carnivore of Europe (Mallinson, 1978).

The dissociative anesthetic ketamine hydrochloride, used alone or with the alpha-2-adrenoceptor agonist xylazine hydrochloride, has been broadly recommended for the chemical immobilization of nondomestic felids (Logan et al., 1986; Seal & Kreeger, 1987). Ketamine hydrochloride produces a dissociative or cataleptoid anesthesia (Wright, 1982) whereas xylazine hydrochloride acts as a nonnarcotic sedative analgesic (Seal & Kreeger, 1987). The main disadvantage of ketamine is poor muscle relaxation, which is avoided when used in combination with xylazine (Amend et al., 1972).

### MATERIAL AND METHODS

Between April 1983 and March 1992, 31 different wild individuals of Iberian lynx (*Felis pardina*) were captured using box-traps and

coil-spring traps (Victor #2, Woodstream Co., Lititz, Pennsylvania, USA) with padded jaws, and immobilized in Doñana National Park, Spain (37°00'N, 06°30'W) as part of a study on radiocollaring (Delibes and Beltrán, 1986). Some animals were recaptured on successive occasions, but we have included here immobilization data for the same animal only if  $\geq 6$  mo had elapsed, in order to avoid pseudoreplication (Hurlbert, 1984). We classified lynx as adults and juveniles depending on their weight, dentition, and body measures (Beltrán and Delibes, 1993).

Once captured, the animals were put into a squeeze cage (100 cm  $\times$  75 cm  $\times$  40 cm) and given manual intramuscular injection of a combination of 8 to 70 mg of xylazine hydrochloride (23.32 mg/ml Rompún, Bayer, Barcelona, Spain) and 8 to 100 mg of ketamine hydrochloride (50 mg/ml Ketolar, Parke-Davis, El Prat de Llobregat, Spain) into their hindquarters. After anesthetic induction, animals were weighed and measured; samples of blood and parasites also were collected. Lynxes were placed on right lateral recumbency, and rectal temperature was monitored every 5 min. Eyes were covered with a piece of cloth to avoid corneal damage. Ambient temperature during immobilization ranged between 15 and 28 C. Animals recovered in the immobilization cage, in a quiet and dark room.

We designated induction time (IT) as the time elapsed from the injection of the anesthetics to the lack of response to external (tactile and auditory) stimuli. First recovery time (FRT) was defined as the time from the lack of response to external stimuli to the first movement of the head. The animals were held inside the immobilization cage for at least 12 hr after first reaction, and then released at the site of capture, fully recovered, and with no apparent adverse effects.

We employed the one-way ANOVA test with the variables that satisfied the requirements of

parametric statistical tests, and with the  $\log(X + 1)$  transformation of the variables that did not fit these requirements (dose of xylazine hydrochloride per weight, total anesthetics dose, ratio of anesthetics, IT and FRT), to compare the effects of different factors (Sokal and Rohlf, 1981). Single and multiple regression were conducted in order to evaluate the influence of doses on FRT and IT; hypotheses concerning partial regression coefficients were checked with a two-tailed  $t$ -test (Zar, 1984). Acceptance of significant differences was set at  $P < 0.05$ .

### RESULTS

The resulting sample of 45 independent immobilizations consisted of 14 adult males (mean weight = 12.6 kg; SE = 0.5; range = 9.3 to 15.9), nine adult females (mean weight = 9.8 kg; SE = 0.3; range = 8.1 to 11.0), 11 juvenile males (mean weight = 6.0 kg; SE = 0.7; range = 2.3 to 10.4) and 11 juvenile females (mean weight = 6.8 kg; SE = 0.6; range = 4.0 to 9.7).

The mean dose for KH was 4.6 mg/kg (SE = 0.21; range = 2.13 to 8.16;  $n = 45$ ) and the mean dose for XH was 4.0 mg/kg (SE = 0.17; range = 2.14 to 6.57;  $n = 45$ ). The mean IT achieved with these doses was 5.6 minutes (SE = 0.3; range = 1 to 11;  $n = 45$ ) and the mean FRT was 59.3 min (SE = 6.5; range = 15 to 120;  $n = 45$ ). There were no significant differences in the dosages, IT, or FRT among the sex-age classes considered.

In six additional cases, the initial dose did not effectively immobilize the animal, and an additional dose was injected that consisted of half the initial dose of the two anesthetics. Initial doses were not statistically different from the cases without second injection ( $F = 0.41$ ,  $P = 0.53$ , for KH;  $F = 0.06$ ,  $P = 0.81$ , for XH). These six cases were excluded from general analysis.

Convulsions occurred in four of 45 cases. The mean KH dose, though slightly greater in these four cases (mean ( $\pm$ SE) = 4.7 ( $\pm$ 0.3) mg/kg) than in the others (mean ( $\pm$ SE) = 4.5 ( $\pm$ 0.2) mg/kg) was not significantly different for this factor ( $F = 0.06$ ,  $P = 0.82$ ). However, the XH dose was greater ( $F = 3.99$ ,  $P = 0.052$ ) in the cases

with convulsions (mean ( $\pm$ SE) = 5.1 ( $\pm$ 0.6) mg/kg) compared to others (mean ( $\pm$ SE) = 3.8 ( $\pm$ 0.2) mg/kg). Convulsions lasted only a few minutes and were followed by a normal recovery. The FRT was longer in the cases with convulsions (mean ( $\pm$ SE) = 95.5 ( $\pm$ 23.3) min) than in cases without convulsions (mean ( $\pm$ SE) = 55.8 ( $\pm$ 6.6) min;  $F = 3.88$ ,  $P = 0.055$ ), but the IT, though shorter in animals with convulsions (mean ( $\pm$ SE) = 3.3 ( $\pm$ 0.8) min), did not differ significantly from the IT of animals without convulsions (mean ( $\pm$ SE) = 5.8 ( $\pm$ 0.4) min;  $F = 1.55$ ,  $P = 0.22$ ).

Variation in IT (log-transformation) had a negative and significant regression on the XH (log-transformation) dose ( $y = 2.53 - 0.44x$ ,  $r^2 = 0.102$ ,  $P = 0.04$ , where  $x = \log(XH + 1)$  and  $y = \log(IT + 1)$ ) but not on the KH dose ( $P = 0.64$ ,  $x = \log(KH + 1)$  and  $y = \log(IT + 1)$ ). The FRT (log-transformation) had a positive and very significant regression on the XH (log-transformation) dose ( $y = 2.0 + 1.22x$ ,  $r^2 = 0.201$ ,  $P = 0.002$ , where  $x = \log(XH + 1)$  and  $y = \log(FRT + 1)$ ) but not significant on the KH dose ( $P = 0.33$ ,  $x = \log(KH + 1)$  and  $y = \log(FRT + 1)$ ).

In order to evaluate the whole effect of KH and XH doses, we conducted multiple regressions of either IT and FRT on the variables sum of KH and XH administered per kg of body weight (KH + XH), and KH/XH ratio with all variables log-transformed. The multiple regression of IT on these two variables was not significant ( $F = 2.51$ ,  $P = 0.09$ , where  $y = \log(IT + 1)$ ,  $x_1 = \log[(KH + XH) + 1]$ , and  $x_2 = \log[(KH/XH) + 1]$ ), but the single regression on KH/XH ratio was nearly significant ( $y = 1.51 + 0.41x$ ,  $r^2 = 0.079$ ,  $P = 0.072$ , where  $x = \log[(KH/XH) + 1]$  and  $y = \log(IT + 1)$ ). The FRT multiple regression was significant ( $F = 5.54$ ,  $P = 0.0073$ ) and the fitted equation was:

$$y = 1.61 + 1.27x_1 - 0.71x_2$$

where  $x_1 = \log[(KH + XH) + 1]$ ,  $x_2 = \log[(KH/XH) + 1]$  and  $y = \log(FRT + 1)$ . The total anesthetic dose (KH + XH) had

a greater predictive effect ( $t = 3.09$ ,  $P = 0.004$ ) than the anesthetics ratio ( $t = -1.76$ ,  $P = 0.085$ ) in the multiple regression model of FRT. Single regression of FRT on total anesthetic dose (KH + XH) was very significant ( $y = 1.35 + 1.14x$ ,  $r^2 = 0.152$ ,  $P = 0.008$ , where  $x = \log[(KH + XH) + 1]$  and  $y = \log(FRT + 1)$ ).

Mean body temperature during immobilization was 37.9 C (SE = 0.1; range = 35.0 to 41.5) but appeared to vary with ambient temperature. Heart rate was measured only in four cases, the mean was 106 beats per minute (SE = 3.46; range = 96 to 112). Mean respiratory rate was 32 breaths per minute (SE = 2.83; range = 24 to 36;  $n = 4$ ).

### DISCUSSION

The advantages of immobilization with ketamine and xylazine have been well documented (Seal and Kreeger, 1987; Kreeger et al., 1990) and confirmed by this study: a high margin of security, quick induction, normal body temperature and profound analgesia and amnesia.

The need in some of our cases of a second dosage of anesthetics may be related to the injection site; some superficial injections may not have entered the muscle mass. Drugs injected subcutaneously or into fat deposits result in reduced degree of immobilization and a prolonged recovery time due to poor absorption (Pemberton and Gales, 1991). Also the wide spectrum of nutritional and physiological condition of animals in the wild may have been a contributing factor (Travaíni et al., 1992).

Convulsions could have been the result of individual variation in the sensitivity to ketamine, an agent described as the cause of such muscular alterations (Wright, 1982). Three of the four animals that suffered convulsions were direct relatives: a female, her son and her granddaughter. Thus, genetics may have been a significant factor.

The greater correlation between IT and FRT to xylazine than to ketamine, was consistent with Wright's (1982) findings.

This dependence may be due to the high doses of xylazine; our KH doses were two to three times less, and our XH were three times greater than those recommended by authors working with bobcats (*Felis rufus*) and Canada lynx (*Felis lynx*) (Seal and Kreeger, 1987). The mean doses employed in our study have been used since 1983, when they were recommended to us by S. Jonsson (University of Stockholm), who was using them successfully with the European lynx (*Felis lynx*). The high amount of XH in our doses did not appear to be dangerous for the animals immobilized, though a smaller dose could be suitable. Despite these apparently high XH doses we did not detect any respiration arrest, the more important side effect of XH (Haigh, 1982). The safety of the doses employed was also supported by subsequent radiotracking of all animals and successive recapture of some of them (Delibes and Beltrán, 1986). We are not able to say that the doses used were optimal, but in our study they successfully immobilized all the animals without apparent negative effects.

The greater predictive value of the KH/XH ratio compared to the total amount of KH + XH on the IT, means that in the dose ranges employed in this study, the IT was determined by the relative proportion of the anesthetics, with low values of KH/XH giving shorter IT. In contrast, FRT was predicted mainly by total KH + XH, though the ratio of anesthetics also had certain predictive value: lower KH/XH combinations gave longer FRT. Thus with the same total amount of KH + XH, we obtained shorter IT and longer FRT by increasing XH with respect to KH.

In summary we report the successful use of the following mean doses to immobilize captured free living Iberian lynx: 4.6 mg/kg of ketamine hydrochloride and 4.0 mg/kg of xylazine hydrochloride, providing a mean immobilization time of 59.3 minutes (SE = 6.51). With an adult animal of an unknown weight, we recommend 46 mg of ketamine hydrochloride and 40 mg of xylazine hydrochloride.

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