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RESEARCH PAPER

A comparison of sedative effects of xylazine alone or combined with levomethadone or ketamine in calves prior to disbudding

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Abstract

Objective To compare the sedative effects of intramuscular xylazine alone or combined with levomethadone or ketamine in calves before cautery disbudding.

Study design Randomized, blinded, clinical trial.

Animals A total of 28 dairy calves, aged 21 ± 5 days and weighing 61.0 ± 9.3 kg (mean ± standard deviation).

Methods Calves were randomly allocated to three groups: xylazine (0.1 mg kg^{-1}) and levomethadone (0.05 mg kg^{-1}; group XL), xylazine (0.1 mg kg^{-1}) and ketamine (1 mg kg^{-1}; group XK) and xylazine alone (0.2 mg kg^{-1}; group X). Local anaesthesia (procaine hydrochloride) and meloxicam were administered subcutaneously 15 minutes after sedation and 15 minutes before disbudding. The calves' responses to the administration of local anaesthesia and disbudding were recorded. Sedation was assessed at baseline and at intervals up to 240 minutes postsedation. Times of recumbency, first head lift and first standing were recorded. Drug plasma concentrations were measured.

Results Data were obtained from 27 animals. All protocols resulted in sedation sufficient to administer local anaesthesia and to perform disbudding. Sedation scores significantly correlated with drug plasma concentrations ($p \leq 0.002$). Times to recumbency did not differ among protocols (2.8 ± 0.3, 3.1 ± 1.1 and 2.1 ± 0.8 minutes for groups XL, XK and X, respectively), whereas interval from drug(s) administration until first head lift was significantly shorter in group XK than X (47.3 ± 14.1, 34.4 ± 5.3 and 62.6 ± 31.9 minutes for groups XL, XK and X, respectively). The area under the time-sedation curve was significantly greater in group X than XK or XL (754 ± 215, 665 ± 118 and 1005 ± 258 minutes for groups XL, XK and X, respectively).

Conclusions and clinical relevance Levomethadone or ketamine with a low dose of xylazine produced short but sufficient sedation for local anaesthesia and disbudding with minimum resistance.

Keywords calves, disbudding, ketamine, levomethadone, sedation, xylazine.

Introduction

Disbudding, or the removal of horn buds from calves, is a routine husbandry procedure on dairies in many countries. Nevertheless, the procedure causes severe acute pain and is associated with behavioural and physiological responses (Graf & Senn 1999; Stafford & Mellor 2011; Mirra et al. 2018). The American Veterinary Medical Association (AVMA) recommends pre-emptive analgesia with sedation or general anaesthesia, local anaesthesia, and preoperative and postoperative administration of non-steroidal anti-inflammatory drugs for disbudding (AVMA 2014). Subsequently, the Finnish Government proposed legislation prohibiting any painful animal procedures without adequate sedation and analgesia (Finnish Government 2018).

Xylazine, an α_2-adrenoceptor agonist, is the sedative agent most widely used before disbudding worldwide (Faulkner & Weary 2000; Stilwell et al. 2010). Pain is alleviated during the procedure when xylazine-induced sedation is combined with a local anaesthetic (Grøndahl-Nielsen et al. 1999; Reedman et al. 2021). Moreover, the analgesic effects of an α_2-adrenoceptor agonist may be increased beyond the disbudding

procedure if combined with ketamine or butorphanol (Coetzee et al. 2010; Baldridge et al. 2011). These drugs may improve pre-emptive analgesia and help prevent the development of hypersensitization and thus decrease postoperative pain (Gehring et al. 2009; Coetzee et al. 2010; Baldridge et al. 2011).

Moreover, combining sedative and analgesic agents allows use of a reduced dosage of individual drugs, thus reducing the incidence of side effects. Intramuscular (IM) xylazine (0.2 mg kg^{-1}) combined with butorphanol (0.1 mg kg^{-1}) 20 minutes before disbudding eliminated response to the administration of the local anaesthetic (Grøndahl-Nielsen et al. 1999). However, the scientific evidence of combining ketamine or opioids with an α_2-adrenoceptor agonist for sedation before disbudding is scarce. Therefore, the primary objective of this study was to identify any advantages of the combinations of levomethadone or ketamine with xylazine over xylazine alone for sedation of calves before disbudding. We hypothesized that: 1) both levomethadone and ketamine combined with a low dose of xylazine would produce brief but sufficient sedation for administration of a local anaesthetic and to perform disbudding with minimum resistance; and 2) the duration of sedation would be shorter with the combination drugs than with xylazine alone.

Materials and methods

The Animal Experiment Board of Finland (ESAVI/9272/2018) and the Finnish Medicines Agency approved the study. The license fulfils the requirements of the European Union legislation as well as the Animal Research: Reporting of *In Vivo* Experiments guidelines (ARRIVE) (Percie du Sert et al. 2018, 2020). This study was a part of a larger research project focusing on the administration of levomethadone or ketamine with xylazine to calves before disbudding.

The study was conducted between October 2018 and January 2020 at the Viikki Teaching and Research Farm, University of Helsinki, Finland. A total of 28 clinically healthy dairy calves (four males and 24 females, 27 Finnish Ayrshire and one Simmental), weighing (mean ± standard deviation, SD) 61.0 ± 9.3 kg and aged 21 ± 5 days, were enrolled in the study. The calves were owned by the University of Helsinki, and written information about the study was provided to the farm personnel taking care of the animals. A written consent was obtained. Based on the sample size calculations according to the estimated duration of sedation with xylazine alone (Cagnardi et al. 2017), seven calves per group would be needed to detect a 50% decrease in the duration of sedation with a power of 80% and an alpha level set at 0.05. Inclusion criteria were normal birth weight, no history of an infectious disease (such as diarrhoea or respiratory tract infection) in the 2 weeks before the study, an unremarkable clinical examination and normal blood haematology and chemistry values. The calf was excluded if it reacted vigorously to disbudding or developed clinical signs of disease during the study.

The calves were housed in 3.7 × 4.8 m pens of eight to 13 calves, with a soft insulated lying area with a mattress bedded with sawdust and slatted floors. Calves were fed with 12 L of whole milk per day through an automatic milk feeder identifying calves via individual transponders (Lely feeder; Förster Technik, Germany). Grass hay, commercial calf concentrate and water were offered *ad libitum*.

The calves were clinically examined 1 day before disbudding (heart and respiratory rates, rectal temperature, inspections of oral mucous membranes, auscultation of intestines, heart and lungs and palpation of the umbilicus). Subsequently, the hair around the horn buds was clipped and body weight was measured. For the purpose of another study, two accelerometers weighing 13 g were secured with bandages, one to the neck collar and the other to one metacarpus. On the morning of the study day, the group pen was physically divided into two to allow sedation of the calves to be disbudded without being disturbed by the other calves. Each study day started between 08:30 and 09:30 hours.

Prior to the trial, the calves were randomly (www.randomization.com) allotted into three groups: xylazine (0.2 mg kg^{-1}; Nerfasin vet, 20 mg mL^{-1}; Le Vet B.V., The Netherlands; group X, $n = 9$); xylazine (0.1 mg kg^{-1}) combined in the same syringe with ketamine (1 mg kg^{-1}; Ketador vet, 100 mg mL^{-1}; Richter Pharma AG, Austria; group XK, $n = 9$) or xylazine (0.1 mg kg^{-1}) combined in the same syringe with levomethadone (0.05 mg kg^{-1}; L-Polamivet, 2.5 mg mL^{-1}; Intervet International B.V., The Netherlands; group XL, $n = 10$). Drugs were mixed in the same syringe immediately before injection and administered by an investigator blinded to group assignment (T0). All drugs were administered IM in the biceps brachialis muscle through a 21 gauge, 40 mm needle. Negative pressure was applied before the injection to avoid intravascular injection.

Local anaesthetic for disbudding was injected 15 minutes (T15) after drug administration. A cornual nerve block midway between the horn base and the lateral canthus of the eye together with a ring block around the horn bud were applied bilaterally (Faulkner & Weary 2000) using subcutaneous (SC) injections of procaine hydrochloride (4.5 mg kg^{-1}; Procamidor, 20 mg mL^{-1}; Richter Pharma AG). The behavioural response (such as head movements) to the administration of procaine was recorded (Yes/No) by the researcher (KS) according to whether or not the administration of local anaesthetic was possible by a single person. Following standard treatment practice, the block quality was evaluated 10 minutes later (T25) with a needle prick from a 23 gauge, 30 mm hypodermic needle and performed in the same order as the local anaesthetic injections (i.e., right *versus* left).

Meloxicam (0.5 mg kg^{-1}; Metacam, 20 mg mL^{-1}; Boehringer Ingelheim, Germany) was administered SC at T15 to alleviate postoperative pain, and a second dose was injected after 48 hours.

Sedation was subjectively assessed using a descriptive sedation scoring system (Appendix A). Assessments were performed at baseline (T0) and at predetermined intervals up to 4 hours (T240) by an experienced investigator (KS) blinded to the group allotment. The times from drug administration until recumbency, the first head lift and standing were recorded.

An experienced veterinarian performed disbudding (T30) using a preheated (approximately 650 °C) butane gas cautery iron dehorner (Dehorner GasBuddex with a 20 mm head; Albert Kerbl GmbH, Germany) by placing the device over the horn bud for 3−5 seconds. The presence of any behavioural responses to disbudding (head movement or struggling) was recorded (Yes/No).

Blood (3 mL) was collected via direct venipuncture of a jugular vein at T20, T120 and T240. Plasma was separated by centrifugation (3000 ***g*** for 10 minutes) and stored at −20 °C until analysed for xylazine, ketamine and levomethadone concentrations. The samples were precipitated with 2× volume of acetonitrile (ACN), including 50 ng mL^{-1} propranolol, repaglinide and phenacetin as internal standards. Samples were mixed for 3 minutes and centrifuged at 2952 ***g*** for 20 minutes. Precipitated samples were diluted 1:1 with water and transferred to mass spectrometry analysis. Standards and quality controls were prepared by spiking blank calf plasma. Individual standard curves and quality controls were prepared for each sample matrix and treated similarly to the samples. Analyses were performed with liquid chromatography−tandem mass spectrometry (Waters Acquity UPLC and Waters TQ-S triple-quadrupole MS; Waters Corp., MA, USA) using a reverse-phase C18 column (Waters Acquity BEH C18, 2.1 × 50 mm, 1.7 µm; Waters Corp.). Column temperature was 40 °C, autosampler temperature was 10 °C and injection volume was 2 µL. The aqueous eluent was 0.5% formic acid in water and ACN was the organic eluent. The eluent flow rate was 0.5 mL $minute^{-1}$. Nitrogen was used as a solvent (flow rate, 900 L $hour^{-1}$) and cone gas (flow rate, 150 L $hour^{-1}$). Solvent temperature was 650 °C, and source temperature was 150 °C. The capillary voltage was 1000 V, and the polarity was set as positive ionization. The multireaction monitoring mode transition using mass-to-change ratios were 221 → 105, 238 → 125, 310 → 265, 260 → 116, 180 → 110 and 435 → 230 m z^{-1} for xylazine, ketamine, levomethadone, propranolol, phenacetin and repaglinide, respectively. The cone voltage was 25 V for xylazine, ketamine and levomethadone, whereas the cone voltage was 30, 20 and 20 V for propranolol, phenacetin and repaglinide, respectively. The collision energy was 20, 20, 48, 18, 17 and 35 V and retention times were 1.93, 1.79, 2.88, 2.38, 2.24 and 2.93 minutes for xylazine, ketamine, levomethadone, propranolol, phenacetin and repaglinide, respectively. Accuracy ranged from 83% to 117% and imprecision from 3% to 16% for all drugs. The limits of quantitation (LoQs) were 0.2 ng mL^{-1} for ketamine and 0.5 ng mL^{-1} for xylazine and levomethadone.

Statistical analysis

The area under the time-sedation curve ($AUC_{SED0-240}$) was calculated using the trapezoidal method. Analysis of variance was used for testing the differences between the treatments in AUC, time to recumbency, the first head lift and standing, weight and age, and xylazine concentrations. The sedation scores were nonparametrically analysed using Friedman's test for the time effect within each group and Kruskal-Wallis test for the effect of group at each time point. In addition, Spearman's correlations were performed to determine the associations between drug concentrations and sedation. The number of calves reacting to the administration of local anaesthetic and disbudding was tested for differences between groups with Chi-squared tests. Bonferroni adjustments were used for multiple comparisons where appropriate and the level of significance (p-value) was set at 0.05. All analyses were performed using IBM SPSS Statistics for Windows Version 25 (IBM Corp., NY, USA). Data are presented as mean ± SD except for sedation score as median and interquartile range.

Results

All calves completed the experiment and were successfully disbudded. All recoveries were without complications. However, one calf in group XK was excluded from the study as it reacted strongly to the disbudding attempt. This calf was successfully disbudded after new administration of local anaesthesia. There were no significant differences in the age and body weight among groups.

Deep sedation was achieved in all groups, with sedation scores significantly higher than baseline (T0) up to T150 in group X and up to T90 in groups XK and XL (Fig. 1). Overall times, mean ± SD, from T0 to lying down and first standing up were 2.7 ± 1.1 and 73.7 ± 19.3 minutes, respectively, with no significant differences among groups (Table 1). The mean latency from T0 to the first head lift was 48.1 ± 22.3 minutes, with a significant treatment effect (df 2, F 4.2, $p = 0.03$) as the latency was shorter for group XK than for group X (Table 1). $AUC_{SED0-240}$ was also smaller for groups XK and XL than for

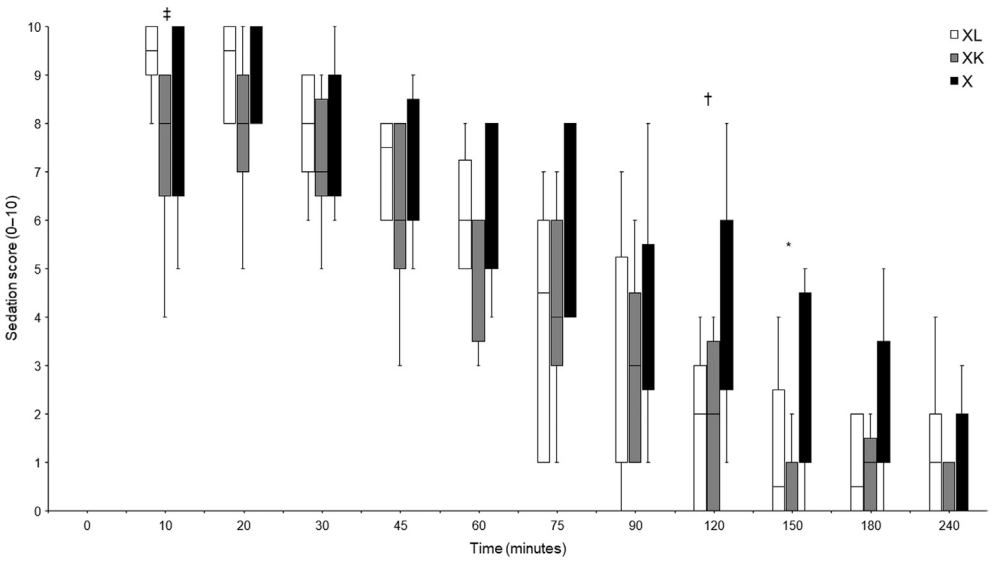

Figure 1 Sedation scores of 27 calves administered intramuscular xylazine (0.2 mg kg^{-1}; group X, $n = 9$) or xylazine (0.1 mg kg^{-1}) combined in the same syringe with levomethadone (0.05 mg kg^{-1}; group XL, $n = 10$) or xylazine (0.1 mg kg^{-1}) combined in the same syringe with ketamine (1 mg kg^{-1}; group XK, $n = 8$) at time 0 (T0). Cornual nerve blocks and ring blocks with procaine were performed at 15 minutes later (T15), followed by disbudding at T30. Data are presented as median (line), interquartile range (box) and range (whiskers). *Significant difference between groups X and XK ($p < 0.05$). †Significant difference between groups XL and X ($p < 0.05$). ‡Significant difference between groups XK and XL ($p < 0.05$).

Table 1 Mean ± standard deviation latencies from intramuscular sedative administration (T0) to lying down, first head lift, first standing up, from lying to standing and area under the time-sedation curve ($AUC_{SED0-240}$) of 27 calves: xylazine (0.2 mg kg^{-1}; group X, $n = 9$) alone or xylazine (0.1 mg kg^{-1}) combined in the same syringe with ketamine (1 mg kg^{-1}; group XK, $n = 8$) or xylazine (0.1 mg kg^{-1}) combined in the same syringe with levomethadone (0.05 mg kg^{-1}; group XL, $n = 10$). Disbudding was performed at T30. As some observations were missing, the number of observed calves is indicated at each time point

Variable	Group		
	X	XK	XL
Latency from T0 to lying down (minutes)	2.1 ± 0.8 ($n = 8$)	3.1 ± 1.1 ($n = 8$)	2.8 ± 0.3 ($n = 10$)
Latency from T0 to first head lift (minutes)	62.6 ± 31.9 ($n = 7$)	34.4 ± 5.3* ($n = 8$)	47.3 ± 14.1 ($n = 8$)
Latency from T0 to first standing (minutes)	80.3 ± 23.2 ($n = 9$)	67.1 ± 22.1 ($n = 8$)	73.1 ± 11.6 ($n = 10$)
Time from first lying down to first standing up (minutes)	78.8 ± 25.1 ($n = 8$)	64.4 ± 21.2 ($n = 8$)	70.3 ± 11.2 ($n = 10$)
$AUC_{SED0-240}$	1005 ± 258 ($n = 9$)	665 ± 118* ($n = 8$)	754 ± 215* ($n = 10$)

*Statistically different from group X ($p < 0.05$).

group X ($p = 0.008$ and $p = 0.045$, respectively). None of the calves substantially resisted local anaesthetic infiltration and four calves reacted slightly to disbudding (one animal from both groups X and XK and two from group XL); however, no significant differences were observed among the groups. It was possible to disbud all calves included in the study by one person applying the disbudder without any need for additional restraint.

Plasma xylazine, levomethadone and ketamine concentrations are presented in Table 2. No comparisons were performed at T240, as the xylazine concentrations in groups XK and XL were lower than the LoQ in all animals except one in each group. Sedation scores significantly correlated with drug plasma concentrations (Fig. 2).

Discussion

The results of the present study indicate that both levomethadone and ketamine combined with xylazine may be an alternative to xylazine alone for sedation of calves before cautery disbudding. Both drug combinations resulted in sufficient sedation for the administration of local anaesthetic and disbudding. The exclusion of one calf from group XK was probably the result of failure of the cornual nerve block.

Table 2 Mean ± standard deviation plasma concentrations of xylazine, ketamine and levomethadone in calves sedated with intramuscular xylazine (0.2 mg kg^{-1}; group X, $n = 9$) alone or xylazine (0.1 mg kg^{-1}) combined in the same syringe with ketamine (1 mg kg^{-1}; group XK, $n = 8$) or xylazine (0.1 mg kg^{-1}) combined in the same syringe with levomethadone (0.05 mg kg^{-1}; group XL, $n = 10$) at T0 and were disbudded at T30 minutes. n = number of calves at each time point. LLoQ, lower than the limit of quantification; na, not applicable

Time points (minutes)	Group	Plasma concentrations (ng mL^{-1})		
		Xylazine	Ketamine	Levomethadone
T20	X	33.56 ± 8.47 ($n = 9$)	na	na
	XK	17.3 ± 3.50* ($n = 8$)	142.0 ± 50.60 ($n = 8$)	na
	XL	17.82 ± 5.15* ($n = 10$)	na	4.37 ± 1.01 ($n = 10$)
T120	X	5.22 ± 1.81 ($n = 9$)	na	na
	XK	2.4 ± 1.14* ($n = 8$)	27.4 ± 17.66 ($n = 8$)	na
	XL	1.56 ± 0.36* ($n = 10$)	na	1.18 ± 0.2 ($n = 10$)
T240	X	1.29 ± 0.27 ($n = 6$)	na	na
	XK	LLoQ	5.4 ± 5.12 ($n = 8$)	na
	XL	LLoQ	na	0.66 ± 0.08 ($n = 7$)

*Statistically different from group X at the same time point ($p < 0.05$).

Fierheller et al. (2012) reported that cornual nerve block was 87.5% effective and attributed the failures to 'the incorrect needle placement, individual anatomical variations or sensory contributions from other nerves, such as the second cervical nerve'. However, in the calves of the present study, additional procaine was infiltrated caudal to the horn-bud to block innervation from the second cervical nerve, but this might have been insufficient to completely desensitize the entire bud. Recently, Thomsen et al. (2021) reported that 42% of calves sedated with IM xylazine (0.7 mg kg^{-1}) and administered a cornual nerve block with procaine showed behavioural responses such as kicking or head lift to the hot-iron disbudding.

Use of levomethadone combined with xylazine for sedation of calves before administering local anaesthesia for disbudding has not been previously published. One study reported that butorphanol (0.1 mg kg^{-1}) combined with IM xylazine (0.2 mg kg^{-1}) eliminated the response to the administration of the local anaesthetic before disbudding in calves aged 4–6 weeks (Grøndahl-Nielsen et al. 1999).

Levomethadone combined with xylazine to sedate young calves before disbudding has potential practical value, as opioids are commonly used in multimodal analgesic regimens in veterinary medicine to improve pain alleviation (Lamont 2008). Moreover, recent evidence indicates the need for more efficient options to treat disbudding-related acute pain (Adcock & Tucker 2020; Adcock et al. 2020) and also the need for options to prevent prolonged and persistent pain after disbudding (Casoni et al. 2019).

Time is often indicated as a major obstacle limiting the use of analgesic agents in cattle, as also discussed by Johnstone et al. (2021). In the present study, the time to recumbency did not differ between calves sedated with levomethadone and xylazine (0.1 mg kg^{-1}) and calves administered only xylazine (0.2 mg kg^{-1}). However, the physiological effects of xylazine combined with levomethadone or methadone in cattle have not been reported in the literature, and thus further investigation about both the efficacy and safety of levomethadone used to treat disbudding-related pain in young calves is warranted.

A combination of intravenous subanaesthetic doses of xylazine and ketamine has been previously studied in older calves (4–6 months of age) undergoing castration, but these studies were conducted without local anaesthesia (Gehring et al. 2009; Coetzee et al. 2010). In the present study, the sedation was less intense and of shorter duration with the combination of xylazine (0.1 mg kg^{-1}) and ketamine (1 mg kg^{-1}) than with xylazine (0.2 mg kg^{-1}) alone. An earlier study determined that an IM xylazine–ketamine combination (0.088 mg kg^{-1} xylazine and 4.4 mg kg^{-1} ketamine) produced lateral recumbency within 3–5 minutes and lasted for approximately 1 hour in six dairy calves aged from 3 weeks to 4 months and weighing 47–123 kg (Rings & Muir 1982).

The shorter sedation could be beneficial to calves, especially in a cold environment, by attenuating the decrease in body temperature (Vasseur et al. 2014). Avoiding the harmful effects of cold stress could help prevent calves from contracting illnesses such as respiratory infection or diarrhoea shortly after the disbudding procedure (Hulbert & Moisá 2016). However, the shorter sedation duration could cause practical problems if a large number of calves are sedated at the same time, administered local anaesthesia and then disbudded individually. Under these conditions, the first calves may recover from sedation before it is their turn to receive local anaesthesia or be disbudded. The effects of different sedation durations regarding practicality and the young calves' health and welfare in connection to disbudding should be studied further.

There was good correlation between plasma drug concentrations and sedation scores, which contrasts with previous

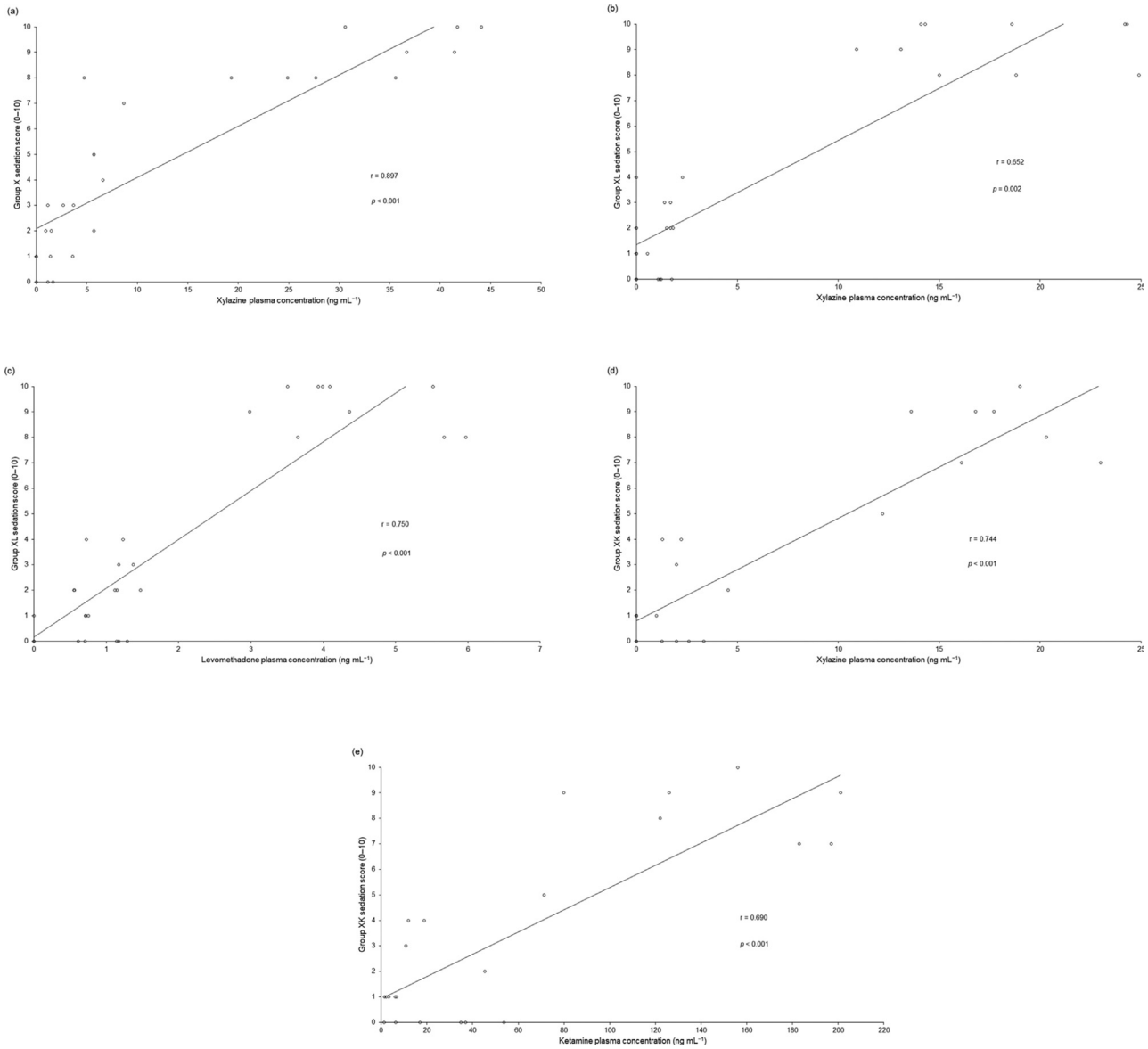

Figure 2 Plasma drug concentrations *versus* sedation scores in calves administered intramuscular (a) xylazine (0.2 mg kg^{-1}; group X, $n = 9$) or (b, c) xylazine (0.1 mg kg^{-1}) combined in the same syringe with levomethadone (0.05 mg kg^{-1}; group XL, $n = 10$) or (d, e) xylazine (0.1 mg kg^{-1}) combined in the same syringe with ketamine (1 mg kg^{-1}; group XK, $n = 8$) at (T0) and disbudding at (T30). The blood samples were collected at T20, T120 and T240. Numbers of measurements at T20 and T120 are the same as group numbers, numbers are less at T240 (Table 2).

studies in which no significant correlation was established between xylazine concentrations and the depth of sedation (Garcia-Villar et al. 1981). However, as blood samples were collected only three times in the current study, further investigations with more frequent sampling are necessary to fully elucidate this relationship.

Interestingly, the concentration of xylazine in plasma was unquantifiable at T240 in groups XK and XL. In one previous study (Baldridge et al. 2011), xylazine was quantifiable in the plasma of calves aged 2−4 months for up to 8 hours after IM injections of xylazine (0.05 mg kg^{-1}), ketamine (0.1 mg kg^{-1}) and butorphanol (0.025 mg kg^{-1}). The LoQ in our study (0.5 ng mL^{-1}) was comparable to the one used by Baldridge et al. (2011). This discrepancy could be attributed to differences in the calves' age [10−37 days in the present study *versus* 2−4 months in the previous study (Baldridge et al. 2011)]. In rats, both ketamine and xylazine have longer half-lives in older animals than younger ones after intraperitoneal

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coadministration; a difference that was reflected in the anaesthesia duration (Veilleux-Lemieux et al. 2013). Furthermore, in the present study, plasma xylazine concentrations at T120 tended to be higher in group XK than in XL, although the differences were not statistically significant. The number of samples was insufficient to calculate any pharmacokinetic variables. In another study, the coadministration of xylazine and ketamine in calves relatively reduced the elimination half-life and the volume of distribution and produced a higher AUC and concentration at time zero of xylazine compared with xylazine alone (Coetzee et al. 2010).

The present study had several limitations. The body temperatures of the calves were not measured during sedation although the study was performed during winter months. Previously, sedation with xylazine was reported to cause an immediate significant decrease in body temperature in calves, and approximately 4 hours to return to presedation temperature. The magnitude of variation in body temperature and its duration were more pronounced when the calves were disbudded in colder ambient temperatures (Vasseur et al. 2014). However, in the present study, the research barn was well insulated, and the sedated calves were kept on mattresses bedded with sawdust. Therefore, the body temperatures of the calves may not have been markedly affected.

A further limitation was the small number of blood samples collected for analysis of drug concentrations. Collection of the first blood sample was timed to coincide with the expected time of peak plasma concentration of xylazine before disbudding, whereas the second and third samples were predetermined to coincide with the expected recovery time and last sedation assessment, respectively. The sedation scores may have been influenced by the clinical procedure of disbudding despite use of local anaesthesia. When local anaesthesia was waning, nociceptive stimulus from disbudded horn buds may have induced a degree of arousal, leading to decreased latencies of head lift and standing. Nevertheless, the sedation scoring based on the behavioural responses showed that overall sedative effects were longer lasting with xylazine alone than with other treatments.

Conclusions

The results indicate that the combinations of xylazine (0.1 mg kg^{-1}) with either levomethadone (0.05 mg kg^{-1}) or ketamine (1 mg kg^{-1}) produced sufficient sedation to administer local anaesthesia and perform disbudding in calves with minimal resistance. With these dosages used in our study, the duration of sedation after disbudding was shorter after administration of xylazine–levomethadone or xylazine–ketamine than after a larger dosage of xylazine (0.2 mg kg^{-1}) alone. However, further research is warranted to elucidate the effects of these combinations on cardiopulmonary function, body temperature and postprocedural pain in calves.

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Authors' contributions

MA, KS and MR: study design, data acquisition, manuscript preparation. RA, ST and MN: data acquisition, manuscript preparation. LH: study design, funding acquisition, data acquisition, statistical analysis, manuscript preparation. A-HH: study design, funding acquisition, manuscript preparation. All authors read and approved the final version of the manuscript.

Conflict of interest statement

The authors declare no conflict of interest.

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Appendix A. Sedation score (0–10)[1]

Category	Score	Description
Spontaneous posture	0	Standing
	1	Sternally recumbent
	2	Laterally recumbent
Reaction to approaching/handling	0	Move away
	1	Stands up
	2	Lifts head (without efforts to stand)
	3	No reaction
Neck muscles relaxation	0	Normal
	1	Moderate, may move head when disturbed
	2	No resistance
Overall appearance	0	No sedation apparent
	1	Mild sedation, head drooping
	2	Moderate sedation, reacts slightly to surroundings, e.g. moves head or ears when approached/hearing sounds
	3	Deep sedation

[1]Modified from Hokkanen et al. (2014).