



Responses of Rabbits to Concurrent Administration of Furosemide and Xylazine-Ketamine

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SUMMARY

The intramuscular (IM) administrations of 5 mg/kg xylazine, followed 30 min later by IM of 35 mg/kg ketamine alone (XK) or concurrently with IM of 1 mg/kg furosemide (XKF), were assessed in six healthy rabbits (4 bucks and 2 does) using selected anaesthetic indices (time to onset of anaesthesia, time to onset of analgesia, duration of analgesia, duration of recumbency/anaesthesia, and time to standing) as well as changes in heart rate (HR), respiratory rate (RR) and rectal temperature (RT) immediately following the loss of righting reflex and subsequently at 10 min intervals over a 60 min period of anaesthesia.

Time to onset of anaesthesia with XKF group (4.4±1.5 min) was not significantly ($P>0.05$) different from that with XK group (4.6±0.9 min). Time to onset of analgesia with XKF rabbits (11.6±4.1 min) was similar to that with XK rabbits (11.2±2.0 min). Duration of analgesia with XKF group (20.0±1.4 min) was significantly shorter than that with XK group (29.6±4.0 min). The respective duration of recumbency/anaesthesia and time to standing with XKF group (79.6±7.7 min and 26.2±1.5 min) were significantly longer than those with XK group (61.4±7.5 min and 6.6±2.2 min). With XKF group, respective ranges of the mean HR, RR and RT were from 110.4±5.5 to 130.4±10.0 beats/min, 78.4±16.5 to 112.0±6.2 breaths/min and from 39.9±0.2 to 40.1 ± 0.2°C; while

respective values with XK group were from 92.4±1.0 to 98.0±2.6 beats/min, 49.6±11.1 to 74.3±7.8 breaths/min and from 39.4±0.6 to 40.0±0.3°C. The mean values of HR and RR with XKF were significantly higher than those with the XK, whereas values of RT with both groups were similar throughout the period of the trials.

It was concluded that administration of furosemide concurrently with xylazine-ketamine anaesthesia in healthy rabbits prolonged the duration of anaesthesia though it shortens the duration of analgesia.

KEY WORDS: anaesthesia, concurrent, furosemide, ketamine, rabbits, xylazine.

INTRODUCTION

The rabbit has been bred for several purposes inclusive of which are: as a source of cholesterol-free animal protein; as a domestic pet; and as a laboratory animal for use in biomedical research (Brodbelt et al, 2005). Therefore, there is a need for the use of anaesthetics by research scientists and veterinary surgeons that frequently see rabbits in their practice.

However, rabbits are often considered difficult to anaesthetise for a number of reasons. First, rabbits have a high surface area volume ratio that makes them more prone to the development of hypothermia under anaesthesia (Flecknell, 1991). Second, tracheal intubation in rabbits and

the use of inhalation anaesthetic agents will be too complicated and time-consuming for the veterinarian in general practice and for the scientist who often conducts research without skillful assistant (Flecknell et al, 1996). Third, venous access can also be a challenge, thus intramuscular injectable anaesthetic techniques are commonly used in the rabbit (Flecknell, 2009).

Accordingly, ketamine-based combinations are the most popular in rabbit anaesthesia (Dupras et al, 2001; Hedenqvist et al, 2001; and Orr et al, 2005). As a sole anaesthetic agent, administered ketamine tends to cause hypertonus, poor muscle relaxation, persistent pain reflex response and violent recovery from anaesthesia (Green et al, 1981; Wright, 1982). For this reason, various sedative drugs including xylazine have been used to counteract these undesirable side effects (Green, 1975; Muir, 1985).

At times, rabbits that are on current drug therapy have to be anaesthetised. For instance, a rabbit with head trauma, congestive heart failure or fluid overload that is on furosemide therapy may need to be given general anaesthesia for some reasons. It has been reported that ketamine is predominantly excreted in the urine as an unchanged drug or as an active metabolite in the cat (Heavner and Bloedow, 1979). In a previous similar study of drug interaction in cats, Adetunji and others (2010) reported reduced duration of recumbency and time to standing, thus faster recovery following concurrent administration of furosemide on diazepam-ketamine anaesthesia. If this is also true for the rabbit, then a clinically significant drug interaction might be expected to occur between a diuretic and ketamine. However, this supposition is yet to be supported in the literature.

The aim of this study therefore, was to

determine the response of rabbits to concurrent administration of furosemide - a loop diuretic, on xylazine-ketamine anaesthesia using selected anaesthetic indices (time to onset of anaesthesia, time to onset of analgesia, duration of analgesia, duration of recumbency/anaesthesia, and time to standing) and some physiological variables such as heart rate (HR), respiratory rate (RR) and rectal temperature (RT).

MATERIALS and METHODS

Animals

Six adult New-Zealand white X American chinchilla rabbits comprising of 4 intact bucks and 2 intact does with the body weight range between 1.1kg and 1.9kg (1.5 ± 0.8 kg, mean \pm SEM), were used for the study. They were acquired from and housed in the Experimental Animal Unit of the Faculty of Veterinary Medicine, University of Ibadan.

The rabbits were housed in pairs in a 60 x 60 x 60 cm dimension wire netted cages with perforations that allow aeration on the sides and floor thereby preventing them from getting in contact with their droppings. Each cage was equipped with concrete feeders and drinkers. The rabbits were fed ad-libitum with 18% crude protein growers' mash and drinking water was provided free choice in their cages.

The animals were allowed two weeks to get accustomed to their new feeding regime and constant human handling. Just before the start of the trial, they were judged to be in good health based on findings at complete physical examination.

Design of the Study

Two series of the trials were carried out on each rabbit at 1 week interval. The first series (test group) consisted of premedication with xylazine, followed 30 min later by intramuscular (IM) administration of ketamine and furosemide concurrently (XKF). The second series (control group) was similarly

treated but without furosemide (XK). The rabbits were randomly assigned to either series of the study. For each group, selected anaesthetic indices were calculated and some physiological variables measured.

Study Procedure

The rabbits were allowed access to food and water up to the time of the trial. The test group was premedicated with IM injection of 5 mg/kg xylazine (Xylax®20 mg/ml, Farvet Laboratories, Holland), followed 30 min later by the concurrent IM injections of 35 mg/kg ketamine (Ketamine®50mg/ml, Rotexmedica, Germany) and 1 mg/kg furosemide (L o p e n ® 1 0 m g / m l , HuayuanZhangshorgPharma Co. Ltd, China) on either thigh. The control group of the trial was similarly treated but without furosemide injection. The anaesthetised rabbits were placed on right lateral recumbency on a foam padded table. Analgesia was tested using the paw pinch withdrawal reflex at 2 min intervals during anaesthesia by clamping haemostat to the first ratchet at the interdigital spaces of the hindlimbs. Withdrawal of the clamped limb indicates presence of pain and non-withdrawal of the clamped limb indicates analgesia.

Calculations

The following selected anaesthetic indices were calculated:

- i. Time to onset of anaesthesia: time interval (in min) between ketamine injection and loss of righting reflex by the rabbit.
- ii. Time to onset of analgesia: time interval (in min) between ketamine injection and loss of pedal withdrawal reflex by the rabbit.
- iii. Duration of analgesia: time interval (in min) between loss and return of pedal withdrawal reflex by the rabbit.
- iv. Duration of recumbency/anaesthesia: time interval (in min) between loss of righting reflex and

assumption of sternal posture by the rabbit.

- v. Time to standing: time interval (in min) between the assumption of sternal and standing postures by the rabbits.

Measurements

The baseline HR, RR, and RT were measured (time 0 min) immediately following the loss of righting reflex and subsequently at 10 min intervals over a 60 min period of anaesthesia. The HR (beats/min) was evaluated with the aid of a precordial stethoscope. The RR (breaths/min) was determined by counting the costo-abdominal movements. The RT (°C) was measured using digital clinical thermometer with the sensor inserted into the rectum.

Data Analysis

All data were expressed as means \pm SEM. The means of the anaesthetic indices were compared using student's t test for paired data. The mean values of the measured physiological variables were compared using analysis of variance for repeated measures followed by appropriate Dunnett test as post test (NCSS 2004, Lange/McGraw-Hill, New-York; Dawson and Trapp; 2004). A probability level of less than 0.05 was accepted for statistical significance in all comparisons.

RESULTS

Calculated Anaesthetic Indices

Time to onset of anaesthesia with XKF group (4.4 ± 1.5 min) was not significantly ($P > 0.05$) different from that with XK group (4.6 ± 0.9 min). Time to onset of analgesia with XKF rabbits (11.6 ± 4.1 min) was similar to that with XK rabbits (11.2 ± 2.0 min). Duration of analgesia with XKF group (20.0 ± 1.4 min) was significantly shorter than that with XK group (29.6 ± 4.0 min). The respective duration of recumbency/anaesthesia and time to standing with XKF group (79.6 ± 7.7 min and 26.2 ± 1.5 min) were significantly

longer than those with XK group (61.4 ± 7.5 min and 6.6 ± 2.2 min).

Measured Physiological Variables

The mean values of the HR, RR and RT for XKF and XK groups were compared as shown in Table 1. The range of mean HR for the XKF group was between 110.4 ± 5.5 and 130.4 ± 10.0 beats/min while that for XK group was between 92.4 ± 1.0 and 98.0 ± 2.6 beats/min. The values for the XKF group were significantly higher than those for the XK group throughout the period of the trial. The mean RR ranged from 78.4 ± 16.5 to 112.0 ± 6.2 breaths/min for the XKF group while for the XK group, it ranged from 49.6 ± 11.1 to 74.3 ± 7.8 breaths/min. The mean RR values for the first 10 min were significantly higher than the values for the rest of the trial period. Also, the mean RR values for the XKF group throughout the trial period were significantly higher than those for the XK group. The mean RT for the XKF group ranged between 39.9 ± 0.2 and 40.1 ± 0.2 °C and it was similar to that for the XK group which ranged between 39.4 ± 0.6 and 40.0 ± 0.3 °C.

TABLE I

The heart rate, respiratory rate and rectal temperature responses of rabbits to intramuscular administration of xylazine-ketamine alone and concurrently with furosemide.

Time interval (min)	HR (beats/min)		RR (breaths/min)		RT (°C)	
	XK	XKF	XK	XKF	XK	XKF
0 ^c	96.8±0.8	110.4±5.5*	50.4±9.0	78.4±16.5*	39.9±0.3	40.1±0.1
10	93.6±2.0	120.0±3.3*	49.6±11.1	81.6±20.2*	40.0±0.3	40.1±0.2
20	93.6±5.2	115.2±3.4*	56.0±12.3	104.8±15.7*	40.0±0.3	39.9±0.2
30	92.4±1.0	114.0±3.0*	54.0±17.3	112.0±6.2*	39.8±0.4	40.0±0.2
40	98.0±2.6	113.6±4.3*	66.7±14.7	104.4±14.1*	39.9±0.6	40.0±0.1
50	94.6±1.3	119.2±2.9*	74.3±7.8	104.0±6.6*	39.9±0.3	40.0±0.2
60	98.0±2.0	130.4±10.0*	72.0±16.0	105.0±6.6*	39.4±0.6	40.0±0.3

Data were processed as means \pm SEM of six rabbits

- 5 mg/kg xylazine followed 30 min later by 35 mg/kg ketamine intramuscularly (XK)
- 5 mg/kg xylazine followed 30 min later by 35 mg/kg ketamine injected concurrently with 1 mg/kg furosemide intramuscularly (XKF)
- Baseline data obtained immediately following loss of righting reflex

* $P < 0.05$ versus control

DISCUSSION

The results of this study showed that the co-administration of furosemide with xylazine-ketamine anaesthesia in the rabbit was associated with a longer duration of recumbency, longer time to standing and shorter duration of analgesia than the control values. The drug combination also caused higher mean heart and respiratory rates than the control values.

It was ascertained that the experimental

rabbits were in good hydration status so that they could withstand drug-induced diuresis. Food and water were provided up till the time just before the procedure because anaesthetized rabbits are not known to vomit (Flecknell, 1991).

The dose rates of both xylazine and ketamine used were those recommended in the literature (White and Holmes, 1976; Lipman et al, 1990; Plumb, 2002; Harcourt-Brown, 2002b). The dose rate used for furosemide was selected from the

dose range of 0.3 to 2.0 mg/kg recommended by Harcourt-Brown (2002a). Since the usual practice in most veterinary clinics is to leave anaesthetized rabbits unintubated (Flecknell et al, 1996), the rabbits used in this study were not intubated. An anticholinergic drug was not administered to counteract the increased secretions caused by xylazine and ketamine because the high atropinesterase level in the plasma of rabbits (Flecknell, 1991; Harcourt-Brown, 2002a) rapidly degrades atropine. Moreover, glycopyrrolate, a potent alternative anticholinergic drug, was not available. A week interval was allowed between each trial to allow for the complete recovery from the effects of drugs administered in the preceding trial.

The duration of analgesia which represents the duration of surgical anaesthesia in the rabbit was significantly shorter with the XKF group than the XK group. This is not unexpected because xylazine is known to have strong hyperglycemic effects (Illera et al, 2000) which results in osmotic diuresis coupled with its inhibitory effect on anti-diuretic hormone (ADH) production (Hall et al, 2001), and so it is expected to exaggerate or add to the diuretic effect of furosemide, thus causing a faster renal clearance of ketamine. The mean duration of recumbency and the time to standing that marked the sleeping time were however significantly longer with the XKF than their respective values with the XK group. This suggests that furosemide potentiates the hypnotic effects of ketamine. Although the mechanism by which this was done was not clear. It was reported that furosemide increased the excretion of K⁺ ions in the proximal tubule (Alexander, 1976; Plumb, 2002), decreased serum potassium levels and thereby causing muscle weakness (Hall et al, 2001). This might probably lead to prolonged sleeping time observed in the XKF group.

The mean heart rates reflected a more

marked bradycardia in the XK group than the XKF group for the duration of the experiment. This finding with the XK group is consistent with earlier findings with this combination by Karl and others (1979) and Amarpal and others (2010), and there were significantly higher heart rates with XKF group. The mechanism behind this is however not understood. It might not be unconnected with the furosemide's effect on decreased serum potassium level, and subsequent muscle weakness and cardiac arrhythmias (Hall et al, 2001).

The mean respiratory rates were markedly increased from 20-60 min in the XKF group and from 40-60 min of the trial in the XK group. The finding of decreased mean respiratory rate observed with the XK group conforms to earlier findings using xylazine-ketamine combination in rabbits as reported by Kilic (2004) and Amarpal and others (2010). There was however a generally higher respiratory rate ($P < 0.05$) in those treated with furosemide. The mechanism behind this finding with the XKF group is however not obvious. This finding may be a response to hypercapnia or hypoxaemia or both in the anaesthetized rabbits.

The mean rectal temperature in the XKF group remained steady till 60 min after an initial decrease at 20th min time interval while for the XK group, that was steadily decreasing till 60 min after an initial increase in the first 20 min of readings (Table 1). This decrease in mean rectal temperature observed with XK group of rabbits also conforms to earlier findings using the same drug combination as reported by Amarpal and colleagues (2010). This is probably due to the CNS depressant effect of ketamine (Hall et al, 2001), or prolonged depression of thermoregulation by xylazine as reported in cats (Ponder and Clarke, 1980). But it is noteworthy that it conforms to the earlier findings that mere xylazine-ketamine combination causes reduced temperature in rabbits (Karl et al, 1979).

In conclusion, administration of furosemide concurrently with xylazine-ketamine combination in rabbits prolonged the duration of anaesthesia though it shortens the duration of analgesia. It is also found to have increased the respiratory and heart rates beyond basal levels of depression of these vital signs noticed with xylazine-ketamine anaesthesia alone.

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