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Research Article

EVALUATION OF MIDAZOLAM-KETAMINE AS AN ANAESTHETIC COMBINATION IN BUFFALO CALVES

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ABSTRACT

The study was undertaken on six clinically healthy male buffalo calves to evaluate midazolam (0.3 mg/kg) and ketamine (6.0 mg/kg) combination as an anaesthetic. Ketamine was administered 10 min after the administration of midazolam. All six animals went into sternal recumbency immediately after intravenous administration of midazolam with drooping of lower lip at 1.27 min and lowering of head with chin on ground at 6.37 min. Animals appeared sleepy with eyes completely closed at 5.34 min. Animals went into lateral recumbency at 16.57 min of midazolam administration. There was copious watery salivation, slight drop wise lacrimation and complete relaxation of muscles of tail, anus, prepuce, neck, limbs, jaw and tongue. Swallowing reflex was abolished. Palpebral reflex was abolished at 24.30 min. There was complete analgesia at fetlock, base of tail, abdomen, ribs periosteum and base of horn. Movement of limbs at 39.30 min was indicating recovery, with return of head righting reflex at 41.78 min. All the animals returned to sternal recumbency at 43.29 min. All the animals stood up at 59.42 min with hind limbs held apart and head down. Complete recovery took 87.20 min. Plasma glucose level showed a non-significant increase after 10 min of midazolam administration. A significant increase in chloride and potassium was also observed. A non-significant increase in aspartate transaminase was observed after 10 min of midazolam and 5 min of ketamine administration as well as at recovery. Alkaline phosphatase showed a non-significant decrease at 5 min of ketamine administration and at complete recovery. The combination was found effective and safe for use in buffalo calves.

Key words: Midazolam, ketamine, buffalo calves

Midazolam is a water soluble imidazole benzodiazepine with sedative, hypnotic, anticonvulsant and muscle relaxant properties (Gross, 2001). It has short duration of action with rapid elimination half-life in humans (Reves *et al*., 1978). It has a rapid onset of action after intravenous administration (Crevoisier *et al*., 1981). In veterinary medicine, midazolam is not used as widely as other agents such as diazepam. It has been reported to produce profound central nervous system depression in dogs when administered in combination with xylazine and butorphanol (Tranquilli *et al*., 1991) and with ketamine in cats (Chamber and Dobson, 1989). Midazolam has minimal effect on cardiopulmonary system in pigs and dogs (Smith *et al*., 1991; Butola and Singh, 2007).

Ketamine has a wide range of effects including analgesia, anaesthesia, hallucination, arterial hypertension and bronchodilation. It is used for induction and maintenance of general anaesthesia in combination with some sedative drugs. The effect of ketamine on the respiratory and circulatory systems is different from that of other anaesthetics. When used at anaesthetic doses, it usually stimulates the circulatory system (Adams, 1997). Ketamine increases heart rate and produces anaesthesia and analgesia of short duration when given alone or after chlorpromazine medication in buffalo calves (Pathak *et al*., 1982). The effects of midazolam–ketamine combination need to be thoroughly investigated in buffaloes. Therefore, the present study was planned with the objective to evaluate efficacy and safety of midazolam–ketamine combination in buffalo calves.

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MATERIALS AND METHODS

This study was undertaken on six male buffalo calves of 6 to 12 months of age and weighing between 105 and 135 kg. Pilot trials were conducted to standardize the dose and route of administration of midazolam and ketamine. Midazolam @0.3 mg/kg was injected intravenously and ketamine was injected @6.0 mg/kg intravenously 10 min later. Behavioural characteristics, rectal temperature (RT), heart rate (HR), respiratory rate (RR), haemoglobin (Hb), packed cell volume (PCV), plasma glucose, urea nitrogen, creatinine, total

plasma proteins, albumin, inorganic phosphorus, calcium, magnesium, sodium, potassium, chloride, aspartate transaminase (AST), alanine transaminase (ALT), alkaline phosphatase (ALKP) and bilirubin were monitored before administration of drugs and at varying time intervals by standard techniques. Blood samples were collected from jugular venipuncture before administration of the diazepam, at 10 min of administration of midazolam, 5 min of administration of ketamine, at complete recovery and at 24 h of recovery. The statistical analysis of data was done using one way analysis of variance and Duncan's multiple range test (Duncan, 1955). Prior to the conduct of experiment, Institutional Animal Ethics Committee approval was obtained for this study.

RESULTS AND DISCUSSION

Effects of administration of midazolam-ketamine combination on behavioral parameters in buffalo calves are shown in Table 1. All the animals went into sternal recumbency immediately after intravenous administration of midazolam with drooping of lower lip at 1.27 min and lowering of head with chin on ground at 6.37 min after midazolam administration. A quick onset of action after intravenous administration of midazolam has been observed in cat and African buffalo calf (Ilkiw *et al*., 1996; Stegmanna, 2004). Animals appeared sleepy with eyes completely closed at 5.34 min. Depression of CNS is mediated through depression of limbic system and due to enhancement of gamma-aminobutyric acid (GABA) by binding of midazolam to GABA receptor, an inhibitory channel which, when activated, decreases nerve activity (Hall *et al.*, 2001). Animals went into lateral recumbency at 16.57 min of midazolam administration. There was copious watery salivation and slight drop wise lacrimation. Complete relaxation of muscles of tail, anus, prepuce, neck, limbs, jaw and tongue was also seen. Swallowing reflex was abolished. Palpebral reflex was abolished at 24.30 min. Relaxation of muscle is due to inhibition of internuncial neurons at spinal level by midazolam (Hall *et al.*, 2001). Similar findings were also observed by Nain *et al.* (2010) in buffalo calves after midazolam administration. There was complete analgesia at fetlock, base of tail, abdomen, ribs, periosteum and base of horn which remained for 10.85 min. Analgesia produced was due to antagonistic effects of ketamine on N-methyl-D-aspartate receptors (Kohrs and Durieux, 1998). Anaesthesia lasted for 17.47 min. Recovery was evident by the movement of limbs at 39.30 min with return of head righting reflex at 41.78 min. All the animals returned to sternal recumbency at 43.29 min and remained in milk-fever posture. Animals started nibbling of grass at 47.25 min. All animals stood up at 59.42 min with hind limbs held apart and head down. Complete recovery took 87.20 min (68 to 100 min).

No significant variations in rectal temperature, heart and respiration rates were observed during the entire period of observation (Table 2). However, there was a non-significant decrease in heart rate at 10 min of midazolam administration (51.67 beats/min) and at 5 min of ketamine administration (54.83 beats/min) as compared to the base value (60.00 beats/min). Kilic (2008) reported a significant decrease in heart rate within 5 min of detomidine-midazolam-ketamine

Table 1
Different behavioural characteristics induced by the intravenous administration of midazolam-ketamine combination in buffalo calves

Reflexes		Mean time ± SE (Min)
Drooping of lower lip*		1.27±0.23
Eyes closed*		5.34±0.59
Onset of salivation		17.74±0.71
Assumption of Lateral recumbency		16.57±0.56
Onset of lacrimation		16.94±1.39
Loss of palpebral reflex		24.30±2.47
Relaxation of muscle	Prepuce	14.02±2.01
	Tail	21.03±1.24
	Neck	16.37±1.35
	Limbs	21.63±1.22
	Jaw	25.13±1.65
Tongue reflex	Loss	20.97±0.74
	Regain	39.82±1.59
Swallowing reflex	Loss	29.13±2.83
	Regain	41.50±1.67
Onset of analgesia	Fetlock	26.81±0.74
	Abdomen	23.88±1.20
	Base of horn	24.03±1.06
	Ribs	28.53±0.61
Loss of analgesia	Ribs	35.57±0.97
	Base of horn	36.40±1.23
	Abdomen	36.22±1.16
	Fetlock	37.66±1.19
Regaining of muscle tone		39.30±1.40
Eyes open		39.30±1.40
Regaining of head rightening reflex		41.78±1.67
Return to sternal recumbency		43.29±1.94
Browsing time		47.25±1.14
Standing with ataxia		59.42±2.37
Complete recovery		87.20±4.77

*Minutes after midazolam administration

anaesthesia in cow calves. Decrease in heart rate in goats (Jangra *et al.,* 2008) and a significant decrease in heart and respiration rate in pigs (Smith *et al.,* 1991) has been reported after intravenous administration of midazolam. Midazolam decreases myocardial contractibility by direct action and by attenuating the catecholamine response to hypotensive state (Glisson *et al.,* 1983). Increase in heart rate after ketamine administration may be due to its action on sympathetic trunk and inhibition of neuronal uptake of catecholamine by sympathetic nerve endings (Tweed *et al.,* 1972; Ivankovitch *et al.,* 1974) or may be due to increase in central release of catecholamine resulting in tachycardia (Hardie and Lukasik, 2007). A slight decrease in both haemoglobin (Hb) and packed cell volume (PCV) was observed (Table 2). Pooling of circulating blood cells in the spleen and other reservoirs secondary to decreased sympathetic activity could be the reason for decrease in Hb and PCV (Pawde *et al.,* 2000; Kilic, 2004).

Effects of midazolam-ketamine combination on certain biochemical parameters are shown in Table 3. Plasma glucose level showed a non-significant increase at 10 min of midazolam (80.42 mg/dl) as compared to the base value (72.23 mg/dl). A significant increase in chloride concentration (103.39 mEq/L) at 5 min of ketamine administration was observed. Nain *et al.* (2010) also observed a significant increase in chloride

Table 2
Effects of midazolam-ketamine on certain clinico-haematological parameters in buffalo calves

Parameters (units)	Before administration of drugs	At 10 min of midazolam administration	At 5 min of ketamine administration	At recovery	At 24 hours of recovery
Ambient temperature (^{0}C)	27.08^a±2.21	27.83^a±2.09	28.75^a±1.96	31.92^a±1.69	31.08^a±1.84
Rectal temperature (^{0}C)	36.38^a ±0.22	36.20^a±0.21	36.13^a±0.27	36.88^a±0.19	36.88^a±0.19
Heart rate (beats/min)	60.00^a±2.5	51.67^a±2.6	54.83^a±4.5	60.50^a±1.3	60.33^a±2.2
Respiration rate (per min)	12.67^a±1.20	15.50^a±1.76	11.33^a±1.14	13.83^a±0.40	13.50^a±0.43
Haemoglobin (g/dl)	11.83^a±0.57	11.33^a±0.59	11.25^a±0.12	11.42^a±0.49	11.42^a±0.57
Packed cell volume (%)	28.33^a±0.49	27.83^a±0.88	27.42^a±0.93	27.67^a±0.95	28.42^a±0.92

Means with same superscripts do not vary significantly ($p>0.05$)

Table 3
Effects of midazolam-ketamine on blood biochemical parameters in buffalo calves

Parameters (units)	Before administration of drugs	At 10 min of midazolam administration	At 5 min of ketamine administration	At recovery	At 24 hours of recovery
Plasma glucose (mg/dl)	72.23^a±6.96	80.42^a±5.95	70.02^a±7.47	72.77^a±4.80	71.78^a±6.58
BUN (mg/dl)	32.92^a±5.64	31.92^a±5.90	35.42^a±5.64	33.53^a±5.79	26.88^a±7.99
Creatinine (mg/dl)	2.45^a±0.07	2.42^a±0.04	2.35^a±0.03	2.38^a±0.05	2.47^a±0.07
Total proteins (g/dl)	12.98^a±1.20	11.11^a±0.64	10.65^a±1.27	12.32^a±0.30	10.95^a±0.38
Albumin (g/dl)	6.32^{ab}±0.28	6.17^{ab}±0.22	6.34^{ab}±0.20	6.49^b±1.16	5.69^a±0.18
Globulin (g/dl)	5.83^a±0.29	5.55^a±0.26	5.48^a±0.31	5.77^a±0.19	5.26^a±0.22
Albumin:globulin ratio	1.08^a±0.01	1.10^a±0.03	1.16^a±0.04	1.13^a±0.03	1.08^a±0.03
Calcium (mg/dl)	7.22^a ±0.75	6.59^a±0.35	7.34^a±0.35	7.70^a±0.73	6.99^a±0.53
Phosphorus mg/dl)	5.28^a±0.63	4.37^a±0.48	4.17^a±0.79	4.26^a±0.70	5.69^a±0.51
Magnesium (mEq/L)	2.88^a±0.63	3.17^a±0.19	2.75^a±0.39	3.31^a±0.28	2.43^a±0.28
Chloride (mEq/L)	97.45^a±2.16	99.51^{ab}±1.21	103.39^b±1.03	101.21^{ab}±2.42	100.04^{ab}±1.24
Sodium (mmol/L)	134.77^a±1.46	132.33^a±2.93	135.12^a±2.58	139.47^a±1.67	137.80^a±3.04
Potassium (mmol/L)	7.15^a±0.16	7.25^a±0.30	7.22^a±0.40	8.00^{ab}±0.52	8.65^b±0.44
ALT (U/L)	49.60^a±2.88	50.14^a±2.96	49.94^a±2.47	47.27^a±1.86	47.85^a±1.67
AST (U/L)	173.08^a±4.68	179.40^a±6.35	182.73^a±4.51	178.07^a±4.84	172.73^a±5.19
ALKP (U/L)	146.50^a±12.45	148.00^a±6.67	124.33^a±13.16	135.17^a±9.09	146.00^a±11.35
Bilirubin (mg/dl)	0.13^a±0.01	0.10^a±0.01	0.08^a±0.02	0.10^a±0.02	0.10^a±0.03

Mean values with different superscripts vary significantly ($p<0.05$)
BUN=Blood urea nitrogen; ALT=Alanine transaminase; AST=Aspartate transaminase; ALKP=Alkaline phosphatase

value at peak effect of midazolam in buffalo calves. Hyperchloraemia may be the result of change in relative water content of body or may be associated with compensated respiratory alkalosis as well as compensated metabolic acidosis (Carlson, 1989). Potassium level also showed a significant increase at 24 hrs of recovery as compared to the base value. Increase in potassium concentration may be due to renal shutdown and metabolic acidosis induced by dehydration and sodium depletion with concomitant decrease in effective circulating fluid volume (Carlson, 1989). A non-significant increase was observed in AST activity after administration of midazolam and ketamine which remained elevated till complete recovery in comparison to base value. In contrast, alkaline phosphatase showed a non-significant decrease at 5 min of ketamine administration and at complete recovery as compared to the base value. The values of all other biochemical parameters fluctuated non-significantly within the normal physiological range. Based on the findings of the present study, it was concluded that midazolam-ketamine combination appears to be safe for general anaesthesia in buffalo calves with minimum cardio-pulmonary and biochemical changes.

REFERENCES

Adams, H. (1997). Circulatory interactions during total intravenous anesthesia and analgesia-sedation. *Anaesthesiol.* **46:** 1081-1087.

Butola, V. and Singh, B. (2007). Midazolam as tranquillizer in dogs. *Indian Vet. J.* **84:** 1141-1145.

Carlson, G. P. (1989). Fluid, Electrolyte and Acid-Base Balance. In: Clinical Biochemistry of Domestic Animals. (4th edn.). Academic Press, Inc. San Diego, California.

Chamber, J.P. and Dobson, J.M. (1989). A midazolam and ketamine combination as a sedative in cats. *J. Assoc. Vet. Anaesth*. **16:** 53-54.

Crevoisier, P.C., Eckert, M., Heizmann, P., Thurneysen, D.J. and Ziegler, W.H. (1981). Relation between the clinical effect and the pharmacokinetics of midazolam following IM and IV administration. *Arzeneim-Forsch/ Drug Reseach.* **31:** 2211-2214.

Duncan, D.B. (1955). Multiple range and multiple F test. *Biometrics* **11:** 15-18.

Glisson, S. N., Haddad, W., Kubak, M.A. and Hieber, M.F. (1983). Anti-adrenergic activity of midazolam and lorazepam in dogs. *Anaesthesiol.* **59:** 328.

Gross, M.E. (2001). Tranquillizers, α_2-adrenergic Agonist and Related Agents. In: Veterinary Pharmacology and Therapeutics. (8th edn.). Iowa State University Press.

Hall, L.W., Clarke, K.W. and Trim, C.M. (2001). Principle of Sedation, Analgesia and Premedication. In: Veterinary Anaesthesia. (10th edn.). W. B. Saunders, London.

Hardie, E.M. and Lukasik, V.M. (2007). Orthopedic Patients. In: Veterinary Anesthesia and Analgesia. Lumb and Jones (ed.). (4th edn.). Blackwell Publishing, Oxford.

Ilkiw, J.E., Suter, C.M., Faver, T.B., McNeal, D. and Steffey, E.P. (1996). The behaviour of healthy awake cats following intravenous and intramuscular administration of midazolam. *J. Vet. Pharmacol. Therap.* **19:** 205-216.

Ivankovitch, A.D., Melietch, D.J., Reinmnn, C., Albrecht, R.F. and Zahed, B. (1974). Cardiovascular effects of centrally administered ketamine in goats. *Anesth. Analg.* **53:** 924.

Jangra, S.K., Chawla, S.K., Peshin, P.K., Tayal, R., Singh, J. and Behl, S.M. (2008). Cardiopulmonary effects of midazolam in goats. *Indian J. Vet. Surg.* **29:** 20-24.

Kilic, N. (2004). Physiological and haematobiochemincal changes in ketamine/diazepam anaesthesia in horse. *Indian Vet. J.* **81:** 396-398.

Kilic, N. (2008). Cardiopulmonary, biochemical and haematological changes after detomidine-midazolam-ketamine anaesthesia in calves. *Bull. Vet. Inst. Pulawy*. **52:** 453-456.

Kohrs, R. and Durieux, M.E. (1998). Ketamine: Teaching an old drug new trick. *Anesth. Analg.* **87:** 1186.

Nain, V., Kumar, A., Singh, J., Singh, S. and Peshin, P.K. (2010). Evaluation of acepromazine, diazepam and midazolam as sedatives in buffalo calves. *Indian J. Vet. Res.* **19**: 22-36.

Pathak, S.C., Nigam, J.M., Peshin, P.K. and Singh, A.P. (1982). Anaesthetic and hemodynamic effects of ketamine hydrochloride in buffalo calves (*Bubalus bubalis*). *American J. Vet. Res.* **43:** 875-878.

Pawde, A.M., Amarpal, Kinjavdekar, P., Aithal, H.P., Pratap, K. and Bisht, G.S. (2000). Detomidine-diazepam-ketamine anaesthesia in buffalo (*Bubalus bubalis*) calves. *J. Vet. Med.* **47:** 175-179.

Reves, J.G., Corssen, G. and Holcomb, C. (1978). Comparison of two benzodiazepines for anaesthesia induction: Midazolam and diazepam. *Canadian Anaesth. Soc. J.* **25:** 211-214.

Smith, A.C., Zellner, J.L., Spinale, F.G. and Swindle, M.M. (1991). Sedative and cardiovascular effects of midazolam in swine. *Lab. Anim. Sci.* **41:** 157-161.

Stegmanna, G.F. (2004). Midazolam/ketamine induction and isoflurane maintenance of anaesthesia in a 2-month-old, hand-raised African buffalo (*Syncerus caffer*). *J. South African Vet. Assoc.* **75:** 43–44.

Tranquilli, W., Graning, L.M., Thurmon, J.C., Benson, G.J., Moum, S.G. and Lentz, E.L. (1991). Effects of midazolam preanaesthetic administration on thiamylal induction requirement in dogs. *American J. Vet. Res.* **52:** 662-64.

Tweed, W.A., Minuck, M. and Nymin, D. (1972). Circulatory response to ketamine anaesthesia. *Anesthesiol.* **37:** 613.