 DOI: https://doi.org/10.3126/jiaas.v38i1.73078

EVALUATION OF CLINICAL AND HEMATOLOGICAL EFFECT OF XYLAZINE-KETAMINE AND XYLAZINE-ATROPINE IN RABBIT

Pradeep Kumar Sah[1], Manish Gautam[2]*, Swastika Sapkota[3] and Shambhu Shah[2]

[1]B.V.Sc. & A.H., Institute of Agriculture and Animal Science, Paklihawa, Rupandehi, Nepal

[2]Assistant Professor, Department of Theriogenology and Physiology, Institute of Agriculture and Animal Science, Paklihawa, Rupandehi, Nepal

[3] B.V. Sc & A.H., Nepal Polytechnic Institute, Purbanchal University, Nepal

*Corresponding author: manish.gautam@pakc.tu.edu.np

Pradeep Kumar Sah: ORCID iD: https://orcid.org/0009-0001-6322-906X

Manish Gautam: ORCID iD: https://orcid.org/0000-0002-8129-9109

Swastika Sapkota: ORCID iD: https://orcid.org/0009-0009-8745-7794

Shambhu Shah: shambhu.shah@pakc.tu.edu.np ORCID iD: https://orcid.org/0000-0002-7675-8657

ABSTRACT

This study was conducted to evaluate the clinical and hematological changes during Xylazine-Ketamine (XK) and Xylazine-atropine (XA) anesthesia in rabbits. Ten healthy and well-fed Rabbits of 1.6 to 2.0 kg BW were selected and divided into two groups: Group I (XK) and Group II (XA). Rabbits in Group I (n=5) were anesthetized with a combination of Xylazine HCl @ 5 mg/kg BW (IM) and Ketamine HCl @ 35 mg/kg BW (IM), whereas Xylazine HCl @ 5 mg/kg BW (IM) and Atropine Sulphate @ 0.2 mg/kg BW (IM) were used for anesthesia in Group II (n=5). In both groups, various clinical parameters like heart rate, respiration rate and temperature were collected from the rabbits before induction of anesthetic agents (control) and thereafter on 15, 30, 45, 60,75, and 90 minutes of post-induction and blood parameters at 0 minute and 30 minutes to evaluate hematological changes. In addition, clinical changes in temperature, respiration and heart rate were also assessed in both groups. RBC, Hb, MCV, MCH and MCHC were decreased significantly ($p<0.05$) in Group I(XK), temperature, respiration and heart rate were decreased significantly ($p<0.05$) in both groups during the experiment. There were no significant differences in lymphocytes, monocytes, granulocyte, HCT, platelets values from baseline in group I(XK) and Group II(XA). The rabbits recovered from anesthesia uneventfully. This result showed that the combination of Xylazine and Ketamine would give better anesthetic results in terms of clinical and hematological parameter and general anesthesia compare to Xylazine and Atropine in healthy rabbits. Rabbits should be carefully monitored by the veterinarians during surgical interventions to avoid anesthesia-related risks and complications.

Keywords: Atropine, Hematology, Ketamine, Rabbit, Xylazine

INTRODUCTION

Rabbits are one of the mostly used experimental lab animals which are generally used in biomedical researches and experimental surgeries aswell (Brodbelt, 2009; Greene and Thurmon, 1988). While conducting surgery and/or other medical operation, pain management is an important factor in rabbit and safe aesthesia is a challenging thing in rabbit (Peeters et.al, 1988). Many research institutions across the world employ rabbits for experimental purposes, and they usually require anesthesia. Preanesthetic mortality in rabbits (1 in 72) have been reported (Khalaf et al., 2014) which might be due to inappropriate anesthetic protocol. It is important to look at the hematological and clinical consequences of regularly used anesthetic combinations. Xylazine and Ketamin are commonly used to sedate the rabbits (Holve et al 2013). Xylazine hydrochloride (HCl) is an alpha-2 agonist which inhibits catecolamine and dopamine, causing inhibition in nerve signals to central nervous system which results in sedative and analgesic along with relaxation of striated muscles effects (Chao et al., 1984; Nesgash et al., 2016). Ketamine hydrochloride is a dissociative

drug that causes deep analgesia, increases arterial pressure and pulse rate, and stimulates the heart. Ketamine usage alone may result in convulsions, myoclonus, and hypertonicity of the muscles (Nesgash et al., 2016). Atropine sulphate keeps the heart rate of animals under sedation at baseline while xylazine acts as a sedative.

There is no any study on effect of xylazine and atropine combination in rabbits. This investigation was designed to compare the sedative effects of combination xylazine-ketamine and xylazine-atropine. Additionally, changes in several blood parameters and clinical manifestations were examined. To lessen the undesirable sensory, motor, sympathetic, and parasympathetic effects, balanced anesthesia is required. Thus, the analysis of several significant hematobiochemical alterations linked to Xylazine-Ketamine and Xylazine-atropine anesthesia in rabbit was the main goal of this study**.**

MATERIALS AND METHODS

This research work was carried out from September to November 2023 in the Veterinary Teaching Hospital of Institute of Agriculture and Animal Science, Paklihawa, Rupandehi, Nepal, to find the effects of Xylazine-Ketamine and Xylazine-Atropine anesthesia on some clinical and hematological indices in rabbits.

Experimental animals

The study was conducted on ten apparently healthy adult rabbits of both sexes weighing 1.6-2.0 kg, two female and eight male and none of female rabbit were pregnant, The rabbits were housed indoors at same condition, fed concentrated food and green grass. Before anesthesia, the animals had free access to water but feed was withheld for 12 hours prior to the experiment. Baseline data of respiratory rate, heart rate and temperature were obtained prior to injection of any drugs.

Study Design

A randomized, controlled experimental design were employed for this study. Each group had four male and one female. Group I (XK) was treated with 5 mg/kg Xylazine and 35 mg/kg Ketamine and Group II (XA) was treated with 5 mg/kg Xylazine and 0.2 mg/kg Atropine. All drugs were administered intramuscularly.

Anesthesia evaluation

Vital signs including heart rate, body temperature and respiration were observed before administration of anesthesia and after 15, 30, 45, 60, 75 and 90 minutes of administration of the anesthesia combinations.

Blood collection and hematological analysis

Blood sample were taken before sedation and after thirty minutes of sedation. Blood samples was collected in (K3-EDTA) vial. 27 G, 1ml syringes was used to collect 1 ml blood from marginal ear vein. Collected blood samples were analyzed in blood hemato-analyzer machine (Mindray BC-2800 Vet) available in Veterinary Teaching Hospital (VTH), Paklihawa campus.

Statistical Analysis

The data was collected in Microsoft Excel sheet and compared with paired t-test using an online graph pad calculator. The data obtained from this research work were calculated as mean ± standard error for all the cases in both groups. Statistical significance was set at $p< 0.05$.

RESULTS AND DISCUSSION

Effect on clinical parameters

The effects of Xylazine-Ketamine (XK) and Xylazine-Atropine (XA) anesthesia on temperature, respiration and heart rate in both groups I and II are shown in Table 1. There was decreased in the physiological parameters (rectal temperature, heart rate, and respiratory rate) in both the experimental group in comparison to baseline value (as in 0 minute)**.** A decrease in the physiological parameters can be related to a reduction in the metabolic rate, relaxation of the muscles, and a direct depressive effect on the central nervous system.(Kandpal et al., 2005). Following anesthesia, there was a substantial drop-in respiratory activity at the $p<0.05$ level. This decline continued from 15 to 90 minutes, however in group I (XK) it was below baseline. Throughout the experiment, both Group I (XK) and Group II (XA) showed significant decreases in respiration at 15 minutes, 45 minutes, 60 minutes, 75 minutes, and 90 minutes at the level of ($p<0.05$). This could be because ketamine depresses the brain's respiratory centers (Afshar et al., 2005; Luna et al., 1997; Dittmar et al., 2004; Mostachio et al., 2008). After 60 minutes of anesthesia, the heart rate was considerably lower ($p<0.05$) than the baseline value (207.20± 43.21 beats/min). This could be because XK causes deep bradycardia that affects loading conditions and ventricular function which is supported by study of (Dittmar et al., 2004; Mostachio et al., 2008 Santosh et al., 2013).

Table 1: Effect of general anesthesia regime induced by xylazine-ketamine and xylazine-atropine on clinical parameters in rabbits

Parameter	Combinations	0 min	15 min	30 min	45 min	60 min	75 min	90 min
		Mean± SD	Mean± SD	Mean± SD	Mean± SD	Mean± SD	Mean± SD	Mean± SD
Temperature (°F)	XK	103.300± 1.631[a]	102.980± 0.795[a]	101.8± 0.604[a]	102.280± 0.396[a]	102.00± 0.587[a]	101.66±0.499[a]	101.980± 0.912[b]
	XA	103.24± 0.986[a]	104.100± 1.120[a]	103.58± 1.564[a]	103.920± 1.496[a]	103.560± 1.126[b]	103.440± 0.841[b]	103.700± 0.866[a]
Respiration/ min	XK	206.40± 45.13[a]	60.80± 18.63[b]	63.20± 17.06[b]	70.40± 11.87[b]	78.40± 12.84[b]	75.60± 310.6[b]	112.80± 5799[b]
	XA	204.80± 22.7[a]	140.00± 49.23[b]	140.00± 62.67[a]	212.00± 87.59[b]	187.6± 65.24[b]	180.00± 48.39[b]	186.80± 34.11[b]
HR (beats/ min)	XK	207.20± 43.21[a]	182.40± 40.65[a]	172.80± 47.78[a]	167.60± 43.51[a]	160.80± 25.98[b]	170.40± 31.95[a]	211.20± 21.80[a]
	XA	202.40± 32.20[a]	131.20± 67.55[a]	155.60± 36.34[a]	268.00± 70.54[a]	113.20± 80.87[a]	209.60± 52.35[a]	221.60± 33.66[a]

*Values bearing common superscript in columns and in rows do not differ significantly from each other ($p<0.05$).

Effect on hematological parameters

Table 2 displays the effects of Xylazine-Atropine (XA) and Xylazine-Ketamine (XK) anesthesia on several hematological markers. Hemoglobine (Hb) and Red blood cells (RBC) were found to be decreased in both the group but it was significantly low only in

Group I at the P<0.05 level. It may be due to the pooling of circulating blood cells in the spleen or other reservoirs as a result of decreased sympathetic activity because of anesthesia (Biswas et al., 2018; Kanu et al., 2018). The anesthetic drug causes vasodilation of smooth muscles resulting in decrease in RBC and Hb in blood (Wilson et al., 2004). MCH and MCV were significantly lower after administration of anesthetic combination in Group I. It can be related to microcytic hyperchromic anemia (Chao et al.,1984). The changes in group II remain insignificant. MCHC was found to be significantly lower at the P<0.05 level than their control values in Group I(XK) combination. Previous studies have stated that IM/IV application of Ketamine result in decrease in the hematocrit and RBC count (Frankel and Hawkey, 1980; Pfeil and Dyesterberg, 1987). There was insignificant increase in the total WBC count in both the group. It can be related to the stress in the rabbits. Studies have suggested that increase in stress causes increase in the cortisol level leading to increasing in the WBC (Mazaheri-Khameneh et al., 2012; Toth and January, 1990)

Table 2: Effect of general anesthesia regime induced by xylazine-ketamine and xylazine-atropine on hematological parameters in rabbits

	Combinations	0 Min	30 Min
		Mean± SD	Mean± SD
WBC(*10^9/L)	XK	8.6800± 1.1323^a	8.7620± 1.1662^a
	XA	7.1080± 0.7538^a	7.2620± 0.7178^a
Lymphocyte(%/L)	XK	29.3900± 1.3413^a	29.2540± 2.1879^a
	XA	30.5040± 1.8063^a	30.5980± 1.8379^a
Monocyte (%/L)	XK	6.480± 0.4087^a	6.5040± 0.4221^a
	XA	5.5020± 0.5442^a	5.500± 0.5116^a
Granulocyte (%/L)	XK	63.7240±1.4448^a	64.2420± 2.1585^a
	XA	63.9140±2.4647^a	63.9020± 2.3210^a
RBC (X10^{12}/L)	XK	7.0920± 0.2862^a	7.0560± 0.2954^b
	XA	7.15± 0.1603^a	7.11± 0.1409^a
Hb(gm/dl)	XK	9.9400± 1.1992^a	9.6760± 1.160^b
	XA	10.4600± 0.7266^a	10.4340± 0.7080^a
HCT%	XK	39.8540± 2.6833^a	39.7940± 2.7223^a
	XA	36.7880± 4.6792^a	36.7960± 4.6779^a
MCV (fL)	XK	55.3140± 6.4390^a	55.2740± 6.4571^b
	XA	55.4160±6.2439^a	55.440± 6.2741^a
MCH (pg)	XK	20.9640± 1.7171^a	20.9280± 1.17276^b
	XA	21.8200± 0.9757^a	21.8220± 0.9805^a
MCHC (mg/dL)	XK	30.6640± 0.92527^a	30.6400± 0.9364^a
	XA	31.8100± 0.8656^a	31.7860± 0.8494^a
Platelets(*10^9/L)	XK	52.0540± 7.1841^a	52.0440± 7.2011^a
	XA	55.8420± 3.9627^a	55.8340± 3.9623^a

*Values bearing common superscript in columns and in rows do not differ significantly from each other (p<0.05).

CONCLUSION

Combination of Xylazine and Ketamine showed the better anesthetic effect with less impact on clinical and hematological parameters in healthy rabbits. Monitoring of rabbits during general anesthesia by Veterinarian is required to avoid anesthesia related risks and complication during surgical interventions.

RECOMMENDATIONS

Further research should be done to identify combinations that provide effective sedation and anesthesia while minimizing undesirable effects on clinical and hematological parameters. Investigate species-specific responses to these drugs to tailor approaches in veterinary medicine and research. Conduct long-term studies to understand sustained effects and establish safe protocols for repeated anesthesia.

ACKNOWLEDGEMENT

The authors are heartily thankful towards Asst. Prof. Dr Dipendra Mishra, Asst. Prof. Dr. Nishant Shah, Asst. Prof. Dr Ganga Prashad Yadav, Asst. Prof. Dr. Suman Kumar Singh, Asst. Prof. Dr. Nishant Shah, Dr. Indresh Jha, Dr. Keshav Lamsal, Mr. Amlesh Yadav, Mr. Krishna Kumar Singh, Anish Balchhaudi, Sachin Shrestha, Dipak Acharya, Saurabh Bastola, Minu Gupta, and everyone who supported during the research period.

Declaration of Conflict of Interest

All the authors have read the manuscript and declare no conflict of interest.

Ethical Approval

This experimental methodology was ethically approved by the Nepal Veterinary Council (Ref. No.34/2080/81).

Funding

The paper was written without any external funding.

REFERENCES

Afshar F. S., Baniadam, A., and Marashipour, S. P. (2005). Effect of xylazine-ketamine on arterial blood pressure, arterial blood pH, blood gases, rectal temperature, heart and respiratory rates in goats. Bulletin of Veterinary Institute in Pulawy, 49, 481-484.

Biswas, D., Hasan, M., Mallick, S., Juyena, N., Shoriotullah, M., and Alam, M. (2018). Clinical and hematological changes upon administration of Xylazine-Ketamine and Xylazine-Thiopentoneanaesthetic combinations in ewes. Bangladesh Veterinarian, 34(1), 9–19. https://doi.org/10.3329/bvet.v34i1.38708

Brodbelt, D. (2009). Perioperative mortality in small animal anaesthesia. The Veterinary Journal, 182(2), 152–161. https://doi.org/10.1016/j.tvjl.2008.06.011

Casas-Diaz, E., Marco, I., Lopez-Olvera, J. R., Mentaberre, G., and Lavin, S. (2011). Comparison of xylazine-ketamine and medetomidine-ketamine anaesthesia in the Iberian ibex (Capra pyrenaica). European Journal of Wildlife Research, 57(4), 887–893. https://doi.org/10.1007/S10344-011-0500-7/METRICS

Chao, C. C., Brown, R. D., and Deftos, L. J. (1984). Effects of xylazine immobilization on biochemical and endocrine values in white-tailed deer. Journal of Wildlife Disease, 20(4), 328–332.

Dittmar, M. S., Fehm, N. P., Vatankhah, B., and Horn, M. (2004). Ketamine/Xylazine Anesthesia for Radiologic Imaging of Neurologically Impaired Rats: Dose Response, Respiratory Depression, and Management of Complications. Comparative Medicine, 54(6):652-655.

Frankel, T., and Hawkey, C. (1980). Haematological changes during sedation in cats. Vet Rec, 107: 512-513.

Greene, S. A., and Thurmon, J. C. (1988). Xylazine – a review of its pharmacology and use in veterinary medicine. Journal of Veterinary Pharmacology and Therapeutics, 11(4), 295–313. https://doi.org/10.1111/j.1365-2885.1988.tb00189.x

Holve, D. L., Gum, G. G., and Pritt, S. L. (2013). Effect of sedation with xylazine and ketamine on intraocular pressure in New Zealand white rabbits. Journal of the American Association for Laboratory Animal Science, 52(4), 488–490.

Kandpal, M., Jadon, N. (2005). Studies on reversal of clinico-physiological effects of xylazine with yohimbine in dogs. Indian Journal of Veterinary Surgery, 26(1), 39-40.

Kanu, S., Alam, M., Hoda, N., and Acharya, K. (2018). Effects of Ketamine and Xylazine-Ketamine Combination in Clinical and Haemato-biochemical Parameters in Captive Rhesus Monkey (Macaca mulatta). Nepalese Veterinary Journal, 35, 150–162. https://doi.org/10.3126/nvj.v35i0.25252

Khalaf, F. H., Al-Zuhairi, A. H., and Al-Mutheffer, A. (2014). Clinical and hematological effect of acepromazine, midazolam, ketamine as general anesthetic protocol in rabbits, 5(2), 55-59.

Lin, J.-H., Shih, C.-H., Kaphle, K., Wu, L.-S., Tseng, W.-Y., Chiu, J.-H., Lee, T.-C., and Wu, Y.-L. (2008). Acupuncture effects on cardiac functions measured by cardiac magnetic resonance imaging in a feline model. Hindawi.Com, 7(2), 169–176. https://doi.org/10.1093/ecam/nem187

Luna, S. P. L., Taylor, P. M., and Massone, F. (1997). Midazolam and ketamine induction before halothane anaesthesia in ponies: cardiorespiratory, endocrine and metabolic changes. Journal of Veterinary Pharmacology and Therapeutics, 20(2), 153–159. https://doi.org/10.1046/J.1365-2885.1997.00049.X

Mazaheri-Khameneh, R., Sarrafzadeh-Rezaei, F., Asri-Rezaei, S., and Dalir-Naghadeh, B. (2012). Evaluation of clinical and paraclinical effects of intraosseous vs intravenous administration of propofol on general anesthesia in rabbits. Veterinary research forum: an international quarterly journal, 3(2), 103–109.

Mostachio, G. Q., De-Oliveira, L. D., Carciofi, A. C., and Vicente, W. R. R. (2008). The effects of anesthesia with a combination of intramuscular xylazine–diazepam–ketamine on heart rate, respiratory rate and cloacal temperature in roosters. Veterinary Anaesthesia and Analgesia, 35(3), 232–236. https://doi.org/10.1111/J.1467-2995.2007.00381.X

Nesgash, A., Yaregal, B., Kindu, T., and Hailu, E. (2016). Evalution of General Anesthesia Using Xylazine-Ketamine Combination with and without Diazipam for Ovariohysterectomy in Bitches. Journal of Veterinary Science and Technology, 7(6):376.

Peeters, M. E., Gil, D., Teske, E., Eyzenbach, V., Brom, W. E. v. d., Lumeij, J. T., and de Vries, H. W. (1988). Four methods for general anaesthesia in the rabbit: a comparative study. Laboratory Animals, 22(4), 355–360. https://doi.org/10.1258/002367788780746197

Pfeil, R., and Duesterberg, J. (1987). Effects of immobilization with ketamine on hematologic values of cats. Zeitschrift fur Versuchstierkunde, 29, 271–276.

Santosh, K. M., Amarpal, Ahmad, R. A., Kinjavdekar, P., Aithal, H. P., Pawde A. M., Kumar, D. (2013). Evaluation of midazolam-ketamine with valuation of midazolam-ketamine with dexmedetomidine and fentanyl for injectable anaesthesia in dogs exmedetomidine and fentanyl for injectable anaesthesia in dogs. Veterinary Archives, 83(5), 509-523.

Toth, L. A., and January, B. (1990). Physiological stabilization of rabbits after shipping. Lab Animal Science, 40, 384–387.

Varghese, P. (2006). Clinical evaluation of the comparative effect of xylazine and xylazlne-ketamine premedication in thiopentone anaesthesia in dogs.(PhD Thesis), College of Veterinary and Animal Science, Mannuthy.

Weinstein, J., and Ralphs, S., C. (2004). External coaptation. Clinical Techniques in Small Animal Practice, 19(3), 98–104. https://doi.org/10.1053/J.CTSAP.2004.09.001

Wilson, D. Evans, A. Carpenter, R. and Mullineaux, D. (2004). The effect of four anesthetic protocols on splenic size in dogs. Vet Anaesth Analg, 31,102-108.

Yucel, U., and, M. L.-E. J. of B., (2019),. The Effect of Repeated Doses of Ketamine on Hematological Parameters in Rats. Dergipark.Org.Tr. Retrieved November 27, 2023, from https://dergipark.org.tr/en/pub/ejbcs/issue/50501/5998