CHAPTER ONE

INTRODUCTION AND LETERITURE REVIEW

1.1 Introduction

Ketamine hydrochloride is the dissociative anesthetic drug of the phencyclidine group and can be used for application of anesthesia in rabbits, it has the properties of both anesthetic and analgesic drugs. (Udegbunam.R.I*et al* .,2015).It has an advantage during anesthesia that maintenance of spontaneous ventilation so this feature make it of value in veterinary use but the administration of ketamine is insufficient for surgical plane to induce the anesthetic effect so it can be used in combination with xylazine diazepam to provide a good anesthetic effect (Yershov.A.L et al .,2002) .

ketamine can cause effects on the different body systems including respiratory and cardiovascular systems depending on administrated dose. When administrated with a normal dose causes mild respiratory depression. And higher doses may cause a serious respiratory depression including apnea and death .The effects of ketamine in the cardiovascular system are changes in heart rate, cardiac output, and blood pressure. (Yershov.A.L et al .,2002).

1.2 Literature review

1.2.1. Economic values of rabbit:-

The socio- economic characteristics of rabbit production with respect to awareness and acceptability in relation to it is unique potentials and good attributes, which include high growth rate ,high efficiency in converting forage to meat, short gestation period, and high prolificacy, relatively low cost of production, high nutritional quality of the meet

which include low fat, sodium and cholesterol level, together with high protein level of about 20.8% (Biobaku and Oguntona,.1997 andAdedeji*et al.*,2015).

Rabbit production is small scale activity that produces innocuous and nutritive meat for families and local market, promoting direct working opportunities and economic income for individuals (Clavel,2004).Rabbit meat is vary essential in improving animal protein intake in developing countries (Orunmyi*et al.*,2006).

1.2.2Use of rabbit on scientific research :

Small animal in addition a clinical research on human beings have contributed to the understanding of various physiological and pathological processes that affect human beings (Ferreiea*et al* .,2005). Rabbits are used for experimental studies in various branches of medical dental sciences (Mapara*et al.*, 2012). It is being easily available and easy to handle and observed, less aggressive animal model, have a short vital cycle (puberty , gestation and lactation).

Rabbits are often used as live models in scientific research where changes in blood count occur, since they handle multiple blood sampling well, their surface veins are pronounced, and they are still and suitable for manipulation in delicate procedures, especially in the detection of health disorders in pet rabbits, where being familiar with reference values is extremely important (Milas, 2009).

1.2.3. Anesthesiain Rabbits:-

1.2.3.1 Injectable anesthesia

Injectable techniques have a long history of use in the rabbit. Ease of administration, predictability, reasonable efficacy, and avoidance of the technical demands of administrating inhalants (Gonzalez, *et al*., 2005)

1.2.3.2. Dissociative agent:

The most dissociative agent has been used in rabbit anesthesia is ketamine, it's should be restricted to minimally invasive procedures; both the IM and IV routes have been used. Constant-dose rate IV infusion of ketamine (30ml/h of a20mg/ml concentration). Intramuscular doses ranging from (20 to 60mg/kg) have been reported although doses greater than (50mg/kg) probably provide little additional restraint. Endo Tracheal intubation has been reported with ketamine alone (15–20mg/kg). (Reich and Silvay,1989).

1.2.3.3. Injection Sites

1.2.3.3.1 Intramuscular injection

The IM site is commonly used for the administration of a variety of parenteral agents used for tranquilization and anesthesia. As in other species, injections should be made into the body of large muscles, avoiding vessels and nerves. IM injections can be made into either the anterior or posterior aspect of the thigh or the lumbar epaixal musculature and also recommend administering drugs into the vastuslateralisAnd rectus femora's muscles of the anterior thigh atrightangles to the femur. Injections into the caudal thigh, specifically the biceps femoris, semi membranous, semitendinosus, and the adductor magnus, can be performed safely if the needle is directed away from the sciatic nerve and its branches. The needle should be inserted into the lateral aspect of the thigh at right angles to the femur or directed caudally away from the femur .(Gardner, 1964; Green, 1982).

1.2.3.3.2Sub cutaneous Injection:

Subcutaneous administration of anesthetics, although uncommon, may be useful with particular agents such as ketamine and medetomidine. The cervical region should be avoided, as rabbits are routinely handled by the scruff of the neck(Gardner, 1964; Green, 1982).

1.2.3.3.3. Intra venous injection :

Vascular access for IV administration of drugs is readily attainable. The lateral (marginal) auricular veins are preferred and easily accessed. Injection and catheterization techniques have been described that can also be used for constant IV or intermittent bolus infusion techniques .The cephalic and recurrent tarsal veins can also be utilized(Gardner, 1964; Green, 1982).

1.2.3.3.4. Intra peritoneal Injection:

Intra peritoneal administration of anesthetics is primarily of historical interest.(Gardner, 1964; Green, 1982)

1.2.4. Inhalation anesthesia :

The use of inhalants, while requiring special equipment and training, provides excellent reliability, efficacy, and anesthetic depth .These features are especially important in rabbit anesthesia where many injectable combinations that are adequate for procedures involving superficial structures are inadequate for more invasive manipulations. Reduction in recovery time is an additional benefit of inhalant anesthesia. The use of inhalants in specific experimental protocols maybe precluded, in that effects such as cardio protection and reduction of infarct size have been demonstrated With isoflurane or halothane .(Lipman,*et al.*, 2008).

1.2.5. Ketamine:

It is a cyclohexamine produce cataleptic state that inhibits movement, but does not provide adequate analgesia for the major surgical procedures .this drug is metabolized in the kidney and contraindicated in the patients with renal insufficiency(Kucuk*et al.,* 2005).

1.2.5.1. Pharmacology of ketamine:

Ketamine also known as Ketamine hydrochloride (Kucuk*et al*., 2005).

1.2.5.2. Chemical names:

The chemical name of ketamine is 2-(2-chlorophenyl)-2-(methyl amino) cyclohexene hydrochloride; 2-(o-chlorophenyl)- 2-(methyl amino) cyclohexene hydrochloride; 2-(methyl amino)-2-(2-chlorophenyl)cyclohexene hydrochloride; 2-(methyl amino)-2-(o-chlorophenyl) cyclohexene hydrochloride; cyclohexene, 2-(2-chlorophenyl)- 2-(methyl amino) hydrochloride; cyclohexene, 2-(o-chlorophenyl)- 2-(methyl amino) hydrochloride (Kucuk*et al.*, 2005).

1.2.5.3. Chemical formula:

Free base: $C_{13}H_{16}C_{l2}NO$

Hydrochloride salt: $C_{13}H_{17}C_{l2}NO$ (Kucuk*et al*., 2005)

1.2.5.4. Mode of action:

Ketamine are generally attributed to direct ketamine-induced inhibition of N-methyl-D-aspartate receptorsAnd also ketamine blocks dopamine uptake and they for elevate synaptic dopamine levels inhibition of central and peripheral cholinergic transmission could contribute to induction of antithetic state and hallucinations(Kucuk*et al*., 2005).

1.2.5.5.Clinical use of ketamine:

Ketamine hydrochloride used for chemical restraint, induction and maintenance of anesthesia and exerts analgesics and anti-inflammatory and antidepressant action(Zanos*et al* 2018).

1.2.5.6. Effects on cardiovascular system:

Ketamine differs from most anesthetic agents in that it appears to stimulate the cardiovascular system, producing changes in _heart rate, cardiac output, and blood pressure_ (Haas and Harper, 1992). Possibly reuptake inhibition of circulating catecholamine's may contribute to this phenomenon. On the other hand cardio depressant effects have been noted in critically ill patients. This may be due to chronic catecholamine depletion preventing any sympathomimetic effects of ketamine and unmasking a negative inotropic effect, which is usually overshadowed by sympathetic stimulation. (Kucuk*et al.*, 2005).

1.2.5.7. Effects on respiratory system

Ketamine is a mild respiratory depressant. It causes a shift of the CO2 dose-response curve to the right, in a dose-related manner, but does not change the slope of the curve. Respiratory drive to CO2 may be depressed as much as 15 to 22%. This effect is similar to that of opioids, but dissimilar from most sedative hypnotics and anesthetics, suggesting that opioid receptors may play a role in the respiratory depressant effect. In clinical studies, the effects were observed only at high doses.(Reich and Silvay, 1989). Some case reports describe respiratory depression after rapid intravenous injection, but also after routine pediatric use of ketamine administered intramuscularly. At recreational doses respiratory depression is not likely to occur, but cannot wholly be excluded. Ketamine has a bronchodilators effect and pharyngeal and laryncheal reflexes are maintained

1.2.5.8. Other pharmacological effects:

1.2.5.8.1Muscle:

Ketamine increases muscle tone. (Kucuk*et al.,* 2005).

1.2.5.8.2Hormones :

Plasma cortisol and prolactin are increased after ketamine administration (Kucuk*et al.,* 2005).

1.2.5.8.3.Eye:

Ketamine may decrease intraocular pressure (Reich and Silvay, 1989).

1.2.5.8.4. Neuro pharmacological actions:

An agonistic effect on α- and β-adrenergic receptors, an antagonistic effect at muscarinic receptors of the CNS, and an agonistic effect at the σ-receptor (Bergman, 1999).

1.2.5.8.5. Other effect of ketamine :

Effect physiological parameters include glucose, urea, creatinine, alkaline phosphatase ,AST, and ALT.and it effects have been widely studied in particular the influence of anesthetic molecules on plasma biochemical parameters in a variety of animal species (Bougherara.H.Bouaziz.O)

CHAPTER TWO

Material and methods

2.1:Experimental animals :

Four apparently healthy female local rabbits were used in the studyTheywere obtained from the local market(Aloasher) and were selected according to their morphological characteristics.

2.2: housing and management:

The experimental rabbits were kept in two cage throughout the study. The animal were fed Lucerne hay (*Medicago Sativa*) and water was available. The animals were kept in cage for an adaptation period of 7 days before experimentation so that they were accustomed to handling, experimental conditions.

2.3 Experimental plan:

A total number of four rabbit were used in the experiment .The experiment have been designed and conducted to investigate the effect of ketamine on the clinical parameters and some haematological parameters on rabbits. The rabbits were randomly assigned to two equal groups anesthetic and control group .The anesthetic group were subjected to 1ml of ketamine injection (5mg/kg). The clinical parameters and blood samples were taken after 30 minutes from the ketamine injection.

2.4 Clinical measurements :

2.4.1 Rectal temperature (Tr):

The measurement of Tr of the experimental animals were made to nearest ± 0.1 C° using electronic clinical thermometer. Thermometer was inserted into the rectum inrotated manner until the thermometer attach rectal mucosa andleft to about one minute before the reading was obtained.

2.4.2 Respiratory rate (breath/min):

The respiratory rate was measured by auscultation from lung the values were taken for one minute.

2.4.3 Heart rate (HR)(beat/min):

The heart rate was measured by monitoring the rate of a stethoscope and stop watch .The value were taken for one minute.

2.5Haematological parameters:

2.5.1 Laboratory investigation

Complete blood count was investigation using haematologicalanalysersysmex KX-21N-Japan.

2.5.2Quality Control of automated Haematologysysmex Kx-21:

This program is used to set Qu method and output method of QC data.They are two kinds: X control (control blood is subjected to two consecutive analysis and the mean of them is used as the QC data.This method causes little influence on reproducibility in analysis).The other is L-J control (this control use data form a single analysis of control blood as QC data.the control width in this method is prone to influence in reproducibility in analysis ,so that the control width is wider then in the first method) (sysmex corporation ,1998).

2.5.3Analysis parameters

This instrument analysis the following parameter using three detectors block and two kind of reagent:

WBCs analysis parameters: DC detedtion method WBCs count in 1ml of whole blood:

LYM% (W-SRC) (WBC-small cell ratio) , ratio % of small lymphocyte to whole blood.

LYM (W-SCC) (WBC-Small cell count).

Absolute count of small lymphocyte in one micro liter of whole blood.

NEUT (W-LCC) (WBC-large cell count).

Absolute count of the neutrophils in one micro liter of whole blood.

HBG (Haemoglobin) analysis principle DC detection method:

HBG (haemoglobin) analysis principle nonocyanide haemoglobin analysis method volume (gram) of haemoglobin in 1dl of whole blood.

2.5.4 Reagent

Instrument reagent system is composed of 5 reagent for testing and cleaning:

Cell pack---------- dilute sample.

WDTM lyses----------WBC dilution.

Stromatolyser CTM---- haemoglobin dilution.

20% Clorox---------- cleaning.

2.6. Statistical analysis:

The obtained data were subjected to standard methods of statistical analysis using SPSS(version 24)The data were performed according to analysis of variance the independent –sample T-test. A level of P value of less than 0.05 was considered statically significant.The value of the parameters measured are expressed as means ± standard deviation (SD).

CHAPTER THREE

Results

The study aimed to investigate the effect of the ketamine on the clinical parameters and some hematological parameters

3.1. Rectal temperature (Tr):-

Fig.3.1shows the effect of injectable ketamine on rectal temperature (Tr). The control values of (Tr) of the rabbit was (39.45) c° the treated group had lower(Tr) mean values (38.1) c° than the mean measured for control rabbit($p \leq 0.03$)

3.2. Respiratory rate (RR):-

Fig 3.2.show the effect ofinjectable ketamine on (RR), lower in anesthetic rabbits (33.5 breath/minute) than control rabbits (61.5 breath/minute)

3.3.Heart rate (HR):-

The effect of anesthesia on heart rate is shown in (Fig3.3) The control values of Heart rate were (137 beat/minute) comparing to (103 beat / min) in anesthetic rabbit ($p \leq 0.01$).

3.4.Packed cell volume (PCV):-

The result of effect of anesthesia on the PCV are shown in Fig3.4There was a slight not significant increase in the PCV% of the anesthetic rabbit.

3.5. Hemoglobin concentration :

Fig3.5.shown the effect of anesthesia on hemoglobin concentration there was a slight not significant increase in the hemoglobin concentration of anesthetic rabbit (10.5 g/dl) compare to (9.5 g/dl) for normal rabbit.

.3.6.Total leukocytes count :-

The effect of anesthesia on total leukocytes count show significantdecrease($P \leq 0.05$) in anesthetic group compare to control groups inFig(3.6).

3.7.Diffrential leukocyte count:-

Injection of ketamine did not significantly affects the differential leukocytes count (table3.1.).

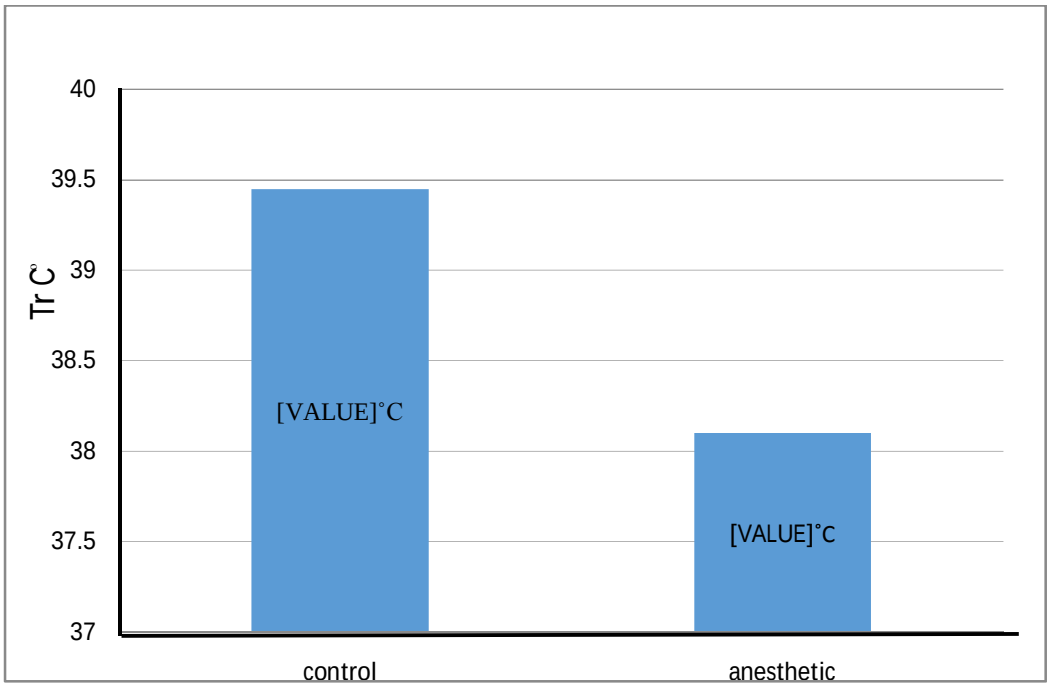

Fig (3.1): Effect of anesthesia (ketamine) on rectal temperature (Tr) inrabbit .

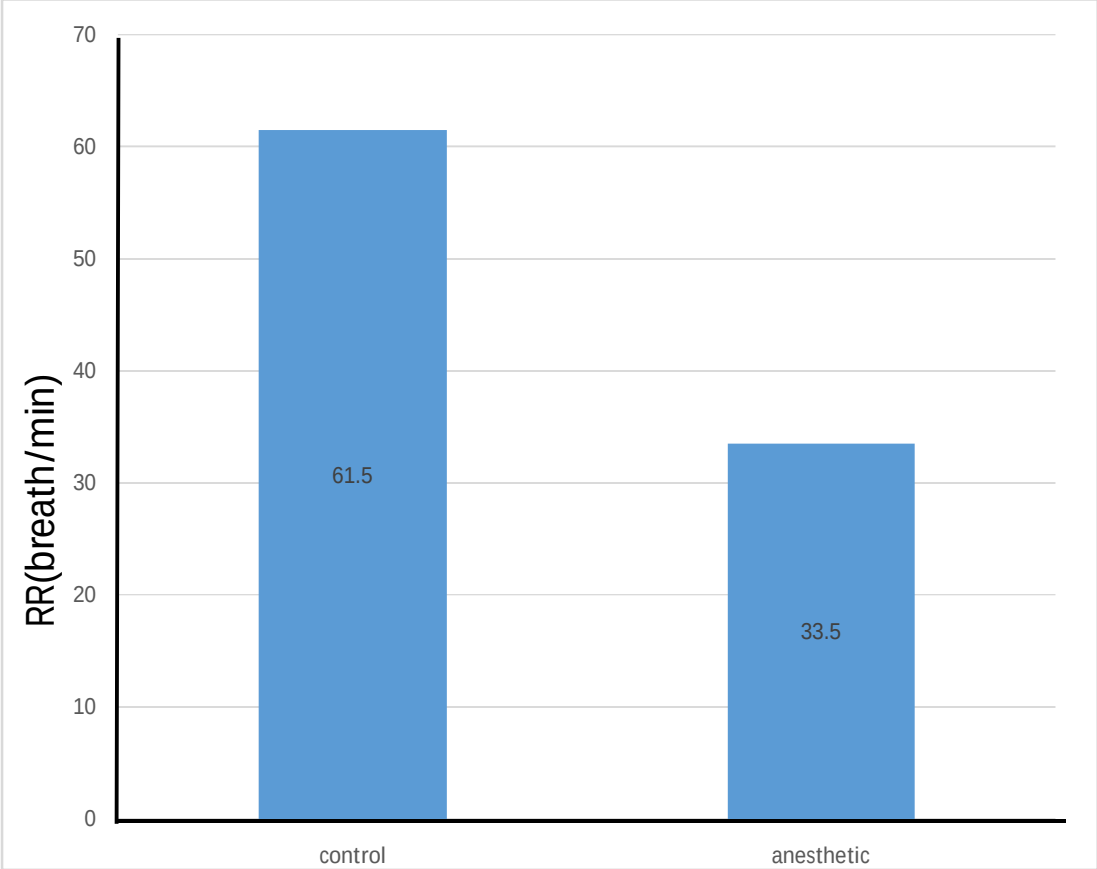

Fig (3.2): Effect of anesthesia (ketamine) on respiratory rate (breath/min) in rabbit

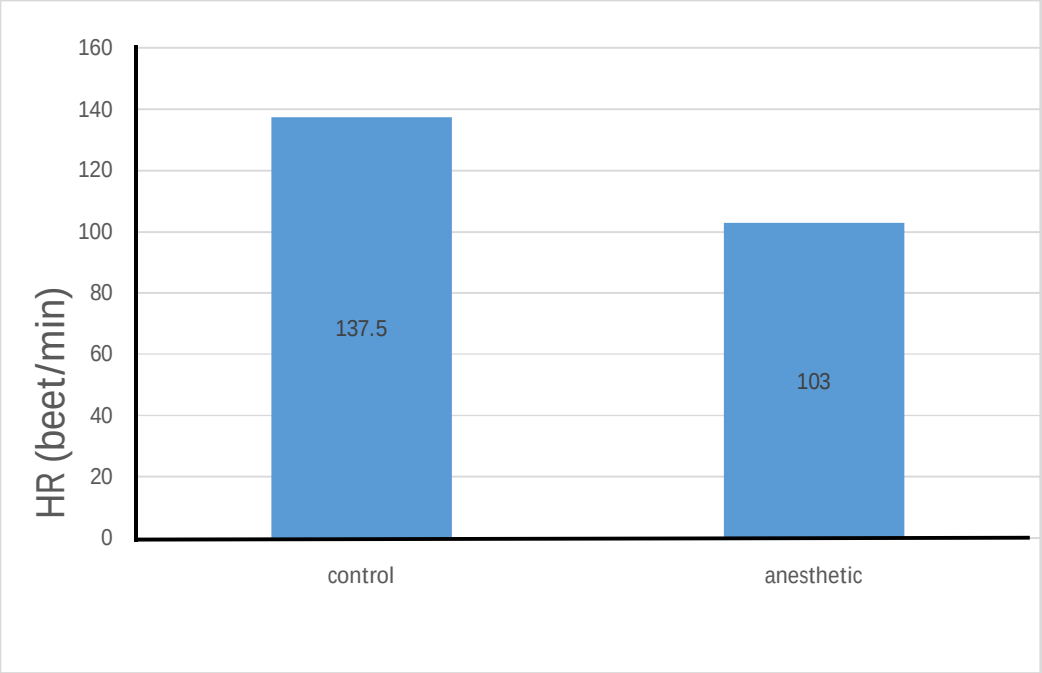

Fig (3.3): Effect of anesthesia (ketamine) on heart rate (HR) in rabbit

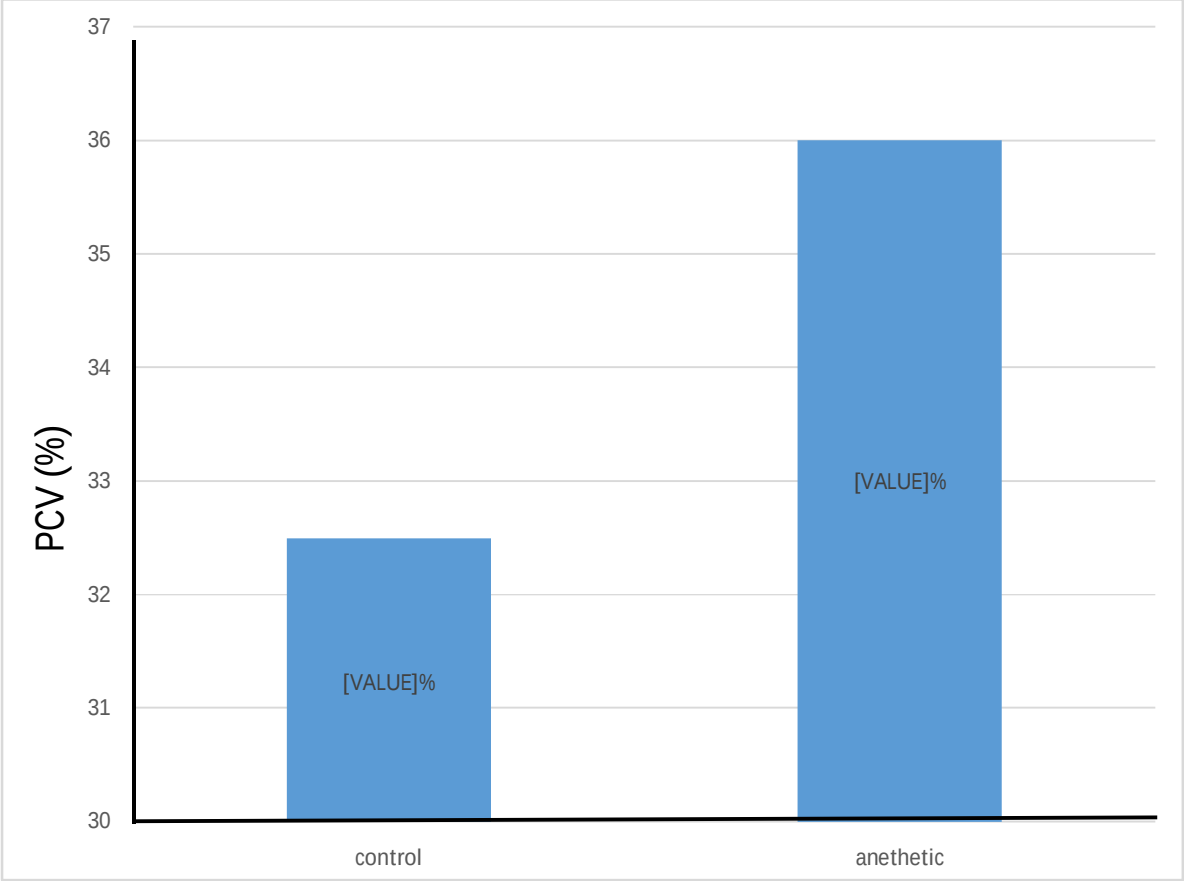

Fig (3.4): Effect of anesthesia on the PCV in rabbit

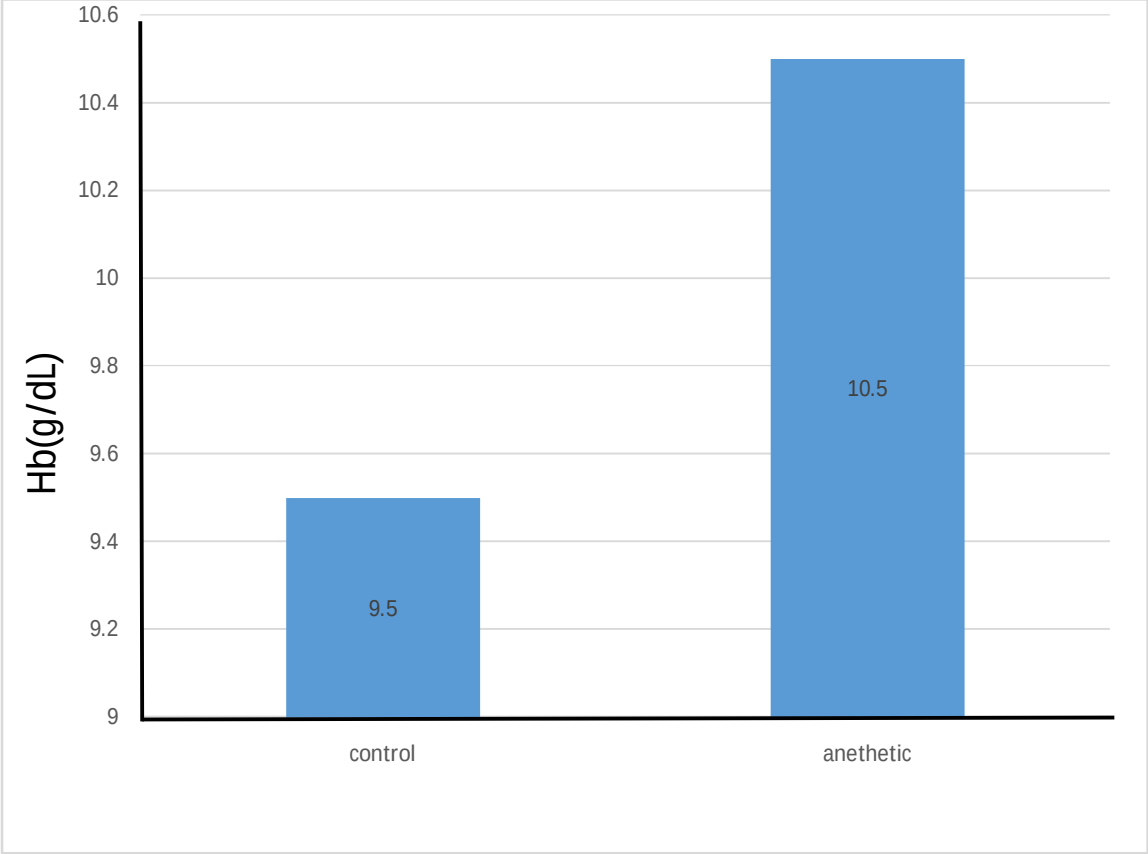

Fig (3.5): Effect of anesthesia on the hemoglobin concentration in rabbit

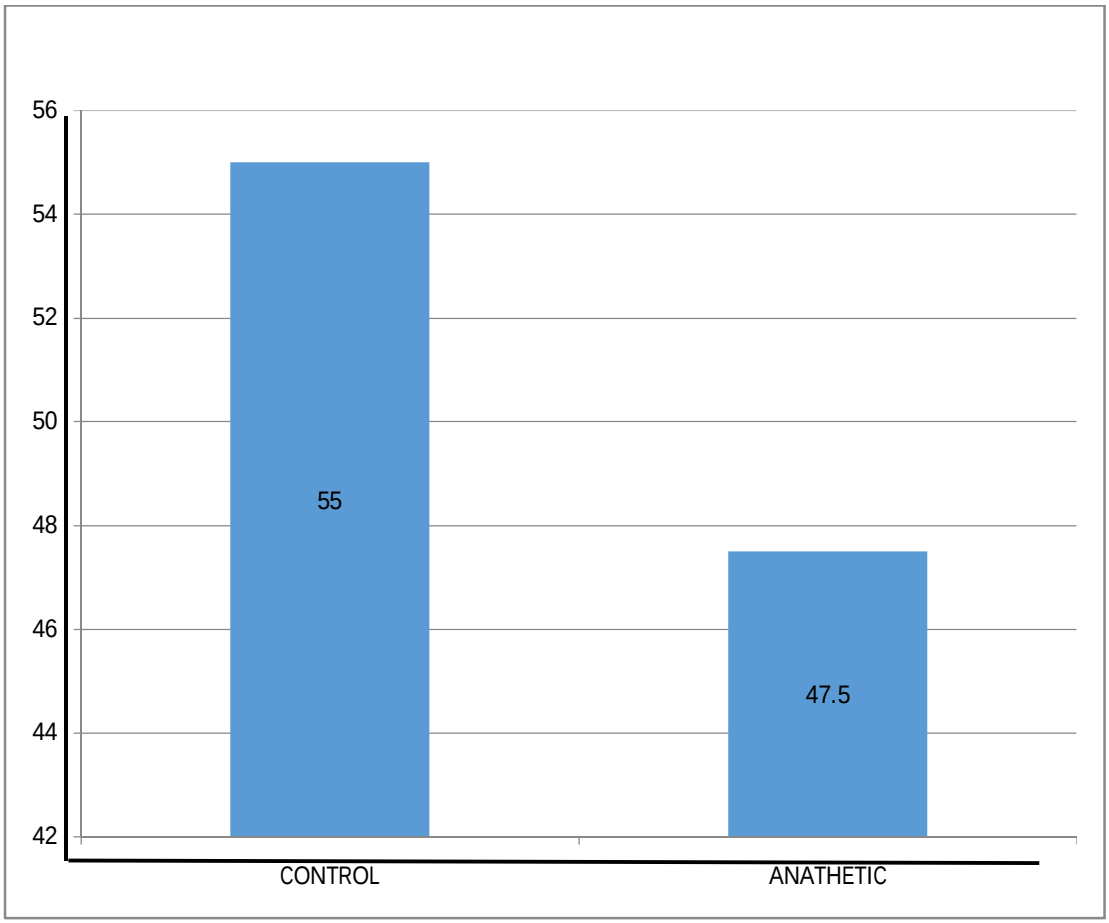

Fig (3.6) Effect of anesthesia on total leukocytes count (TLC) in rabbit.

Table (3.1) Effect of ketamine on differential count in rabbit.

Leukocytes series	Control mean ± S/td. Deviation	Anesthetic mean ± Std. Deviation	Significant
Lymphocyte (%)	60.±1.4	61.0±1.4	N.S
Neutrophils (%)	27.5±0.7	25.5±0.7	N.S
Monocytes (%)	5.±0.0	5.5±0.7	N.S
Eosinophil (%)	7.5±0.7	7.5±0.7	N.S
Basophils (%)	0.0±0.0	0.5±0.7	N.S

Result are M ± SD.

N.S: Non Significant (P>0.05).

Discussion

In this experiment, the effects of ketamine (5mg/kg) were investigated in rabbits .The current results indicate that the anesthesia with ketamine influence the clinical parameters and some hematological parameters in rabbits. This was manifested by the decrease in body core temperature Trin anesthetic rabbit.The decrease in Trcould be attributed with the blocking of hypothalamus regulatory center in the brain after injection of ketamine. Generally anesthesia causes decrease in body temperature due to reduction in metabolic rate and limited movement of animal (Hall *et al*.,2001). The result agrees with Azari*et al*,(2012) andYohannes (2018) .

ketamine also resulted in decrease of RR value of rabbit . These responses may be associated with depression of respiratory center in brain (Walter, 2008). Depression of RR after ketamine injection has was reported by lee *et al* .,(2010) Generally all anesthetic techniques commonly used in rabbit anesthesia depress ventilation (Grint and Murison., 2008). The present result is in agreement with findings of Yohannes , (2018) who reported a decrease in Rr and Tr in rabbits injected by ketamine alone .

The cardiovascular response to ketamine administration were manifested by the decrease of heart rate (Hr) value in rabbits. these response may be associated inhibition the release of central noradrenaline neurotransmitter or depression of sympathetic activity. Similar result have being reported indicating lower(Hr) value in (lee *et al* .2010) , (yohannes,2018) reported that heart rate decreased from (155 to 127) beats/minutes after 20 minutes. in contrast, (Singh *et al*., 2006) noted that dose(2 mg/kg) of epidural ketamine increased heart rate in camel , suggesting an increase

the differences in result may be associated with breed, dose, and mode of administration .

In this study is 1ml (5 mg/kg) of ketamine induce slight change on hematological parameters in the present study. The Hbconcentration and PCV% value were slightly increased an anesthetic rabbit as responses may be associated with stressful condition of the research carried out during summer season and sampling method . Previous studies pointed to contrasting findings that dose of ketamine caused reduction in Hb concentration and PCV due to relaxation of the spleen and consequent splenic sequestration of erythrocyte (Singh *et al.*, 2006). TLC was significantly decrease in anesthetic rabbit. The TLC during the period of anesthesia on sedation might be attributed to shifting of the fluid from extravascular compartment to intravascular compartment (Wagner et al., 1991.,Singh*et al.*, 2006) in dog .

Conclusion and Recommendation

Conclusion:

Used of ketamine in the rabbit effected on the thermoregulation and hematological parameters it is decrease in the rectal temperature, respiratory rate and heart rate.

Ketamine causes slight increase on hemoglobin concentration and PCV%and significantly decrease on TLC values with no significant change on DLC.

Recommendation:

Today the use of ketamine alone is rareUsed of combination such as (xylazine and dizpam) with ketamine to decrease of the side effect of the ketamine and to increase the powerful of anesthesia.

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