EVALUATION OF SELECTED ANAESTHETIC PROTOCOLS FOR TOTAL INTRAVENOUS ANAESTHESIA IN PREGNANT EWES

A thesis submitted
By
RASHA YASSIN FADL ELMOLA ELKHIDR
B.V.M. October 2004
U of K

Supervisor
Dr. ABAS TAHA HAMZA SOBAIR
B.V.Sc., Ph.D.

A thesis submitted to University of Khartoum in fulfillment of the requirements for the degree of Master of Veterinary Science (M.V.Sc.)

Department of Surgery and Anaesthesia
Faculty of Veterinary Medicine
University of Khartoum

May 2010
DEDICATION

TO MY LOVELY FATHER AND MOTHER

TO MY DEAR BROTHERS, SISTERS AND FRIENDS

WITH ALL MY LOVE
ACKNOWLEDGEMENTS

A very special thanks goes to my supervisor Dr. Abbas Taha Hamza Sobair and very instructive auctioneer who donated his time and was a significant part of the success.

Also I am grateful to my co-supervisor Dr. Mohammed Said Mohammed, Department of Obstetric & Gynecology, Faculty of Veterinary Medicine, University of Khartoum for his directions, guidance throughout the work of this study and helping with the statistical analysis of the data.

Special gratitude is extended to Dr. Bushra Husain and Dr. Rehab Mohamed Abd Elgafar for their help with the diagnosis of pregnancy by using Ultrasonography.

Special thanks are to Mokhtar Ali, Abd Allah Mohamed, Mustafa and all the staff members in the department of Surgery and Anaesthesia for their assistance and care for animals throughout the study.

My special thanks is extended to my family, friends and all colleagues for their help and encouragement.

Above all, I thank Allah for giving me strength to finish this work.
Abstract

This study was conducted using sixteen pregnant, clinically healthy ewes to investigate the reliability and safety of the following anaesthetic protocols:

1. Xylazine HCL (2%) + Ketamine HCL (5%), injected at early stage of pregnancy.
2. Xylazine HCL (2%) + Ketamine HCL (5%), injected at late stage of pregnancy.
3. Diazepam (0.5%) + Thiopentone sodium (2.5%), injected at early stage of pregnancy.
4. Diazepam (0.5%) + Thiopentone sodium (2.5%), injected at late stage of pregnancy.

The pregnant ewes at early stage of pregnancy were randomly divided into two groups, eight ewes in each group. The first group at early and later late stage of pregnancy was subjected to protocol 1. The second group at early and later late stage of pregnancy was subjected to protocol 2. Xylazine HCL at a dose rate of 0.1mg/kg body weight and diazepam at a dose rate of 0.4mg/kg body weight were used at pre anaesthetic medications and were injected intravenously.

Fifteen minutes after injection premedication, the pregnant ewes at early and later late stages of pregnancy were injected intravenously with Ketamine HCL at a dose rate of 22 mg/kg body weight. The pregnant ewes at early stage of pregnancy were injected with Thiopentone sodium at a dose rate of 7.5 mg/kg body weight and at late stage of pregnancy at a dose rate of 10 mg/kg body weight.

Throughout the anaesthetic procedures, the major physiological parameters, such as respiratory rate, heart rate and rectal temperature,
were monitored using standard methods. Also, the anaesthetic parameters (anaesthetic phase, lateral recumbancy phase, sternal recumbancy phase and standing phase) were monitored and recorded.

The heart rate was significantly decreased 15 minutes after premedication with Kylazine and restored to normal immediately after application of Ketamine HCL at early and late stages of pregnancy.

There was a significant decrease in the heart rate which occurred at 20, 25 minutes after Thiopentone injection sodium and there was no significant difference in heart rate observed 30 minutes after onapplicati of Thiopentone sodium in ewes at early stage of pregnancy. There was no significant difference in heart rate in ewes at late stage of pregnancy.

There was no significant difference in the respiratory rate in pregnant ewes at early stage of pregnancy when subjected to anaesthetic protocol 1 or anaesthetic protocol 2. However, the respiratory rate was significantly increased up to 15 minutes after premedication with Xylazine or diazepam and restored to normal immediately after application of Ketamine HCL or Thiopentone sodium in ewes at late stage of pregnancy. Apnea was reported only with this anaesthetic protocol 2

Induction of anaesthesia with all the tested anaesthetic protocols resulted in a no significant difference in the rectal temperature in pregnant ewes at early and late stage of pregnancy.

The anaesthetic protocol 2 resulted in a significant prolongation of the durations of lateral and sternal recumbancy phase and in a significant decrease in standing phase compared with anaesthetic protocol 1 in ewes at late stage of pregnancy.

The chosen anaesthetic protocols used for total intravenous anaesthesia proved to be safe and reliable for pregnant ewes at early and late stage, of pregnancy since not a single death case was reported.
List of Contents

<table>
<thead>
<tr>
<th>Description</th>
<th>Pag</th>
</tr>
</thead>
<tbody>
<tr>
<td>DEDICATION .............................................</td>
<td>i</td>
</tr>
<tr>
<td>ACKNOWLEDGEMENT .......................................</td>
<td>ii</td>
</tr>
<tr>
<td>ENGLISH ABSTRACT .................................</td>
<td>iii</td>
</tr>
<tr>
<td>List of Contents ..................................</td>
<td>v</td>
</tr>
<tr>
<td>List of Tables ....................................</td>
<td>ix</td>
</tr>
<tr>
<td>List of Figures ..................................</td>
<td>x</td>
</tr>
<tr>
<td>Introduction ......................................</td>
<td>1</td>
</tr>
<tr>
<td>CHAPTER ONE: .......................................</td>
<td>5</td>
</tr>
<tr>
<td>1. REVIEW OF THE LITERATURE .......................</td>
<td>5</td>
</tr>
<tr>
<td>1.1. Total intravenous anaesthesia (T.I.A) ........</td>
<td>9</td>
</tr>
<tr>
<td>1.1.1. Justification for total intravenous anaesthesia</td>
<td>9</td>
</tr>
<tr>
<td>1.1.2. Technique of total intravenous anaesthesia</td>
<td>11</td>
</tr>
<tr>
<td>1.2. Anaesthetic premedication ....................</td>
<td>11</td>
</tr>
<tr>
<td>1.2.1. Chemistry and pharmacokinetics of anaesthetic premedication</td>
<td>12</td>
</tr>
<tr>
<td>1.2.1.1. Xylazine (Rompun, Anased) ................</td>
<td>12</td>
</tr>
<tr>
<td>1.2.1.2. Diazepam (Valium) ........................</td>
<td>13</td>
</tr>
<tr>
<td>1.2.2. Pharmacodynamics and side effects of anaesthetic premedications</td>
<td>14</td>
</tr>
<tr>
<td>1.2.2.1. Xylazine ..................................</td>
<td>14</td>
</tr>
<tr>
<td>1.2.2.2. Diazepam ..................................</td>
<td>18</td>
</tr>
<tr>
<td>1.3. Anaesthetic Induction ........................</td>
<td>20</td>
</tr>
<tr>
<td>1.3.1. Chemistry and Pharmacokinetics of anaesthetic induction agents</td>
<td>20</td>
</tr>
<tr>
<td>1.3.1.1. Ketamine (Vetalar, Ketalar, Ketaset, Ketaject)</td>
<td>20</td>
</tr>
</tbody>
</table>
1.3.1.2. Thiopentone sodium (Thiosol) ........................................... 22
1.3.2. Pharmacodynamics and side effects of anaesthetic induction agents ................................................................. 24
  1.3.2.1. Ketamine ............................................................... 24
  1.3.2.1. Thiopentone sodium ................................................. 27
1.4. Monitoring ........................................................................ 29
1.5. Recovery from anaesthesia .................................................. 30

CHAPTER TWO: ........................................................................ 31
2. MATERIALS AND METHODS .................................................. 31
  2.1. Materials ......................................................................... 31
    2.1.1. Experimental animals .................................................. 31
    2.1.2. Drugs ...................................................................... 31
      2.1.2.1. Preanaesthetic medications ..................................... 31
      2.1.2.2. Anaesthetic induction ............................................. 32
    2.1.3. Injection set ................................................................ 32
    2.1.4. Monitoring tools ......................................................... 32
  2.2. Methods .......................................................................... 33
    2.2.1. Anaesthetic protocols and methods of injection ............. 33
    2.2.2. Experimental work ..................................................... 34
      2.2.2.1. Experiment (1) ...................................................... 36
      2.2.2.2. Experiment (2) ...................................................... 36
      2.2.2.3. Experiment (3) ...................................................... 36
      2.2.2.4. Experiment (4) ...................................................... 37
    2.2.3. Assessment of reflexes ............................................... 37
      2.2.3.1. Palpebral reflex ...................................................... 37
      2.2.3.2. Jaw reflex ............................................................ 37
      2.2.3.3. Tongue reflex ....................................................... 38
      2.2.3.4. Cough reflex ....................................................... 38
2.2.3.5. Swallowing reflex ................................................................. 38
2.2.3.6. Pedal (digital withdrawal) reflex ............................................. 38
2.2.4. Definitions and monitoring of different anaesthetic parameters and phases: ................................................................. 38
2.2.4.1. Injection time ........................................................................ 38
2.2.4.2. Induction of anaesthesia ......................................................... 39
2.2.4.3. Anaesthetic phase ................................................................. 39
2.2.4.4. Lateral recumbancy phase ..................................................... 39
2.2.4.5. Sternal recumbancy phase ..................................................... 39
2.2.4.6. Standing phase ..................................................................... 39
2.2.4.7. Recovery phase ..................................................................... 39
2.2.4.8. Total recovery time ............................................................... 40
2.2.5. A pilot study ............................................................................. 40
2.2.6. Statistical Analysis ................................................................. 42

CHAPTER THREE: ............................................................................. 43
3. RESULTS ....................................................................................... 43
3.1. Effect of anaesthetic protocol 1 (xylazine-ketamine HCl) on the heart rate in early and late stage of pregnant ewes ......................... 43
3.2. Effect of anaesthetic protocol 2 (diazepam-thiopentone sodium) on the heart rate in early and late stage of pregnant ewes .............. 43
3.3. Effect of anaesthetic protocol 1 (xylazine-ketamine HCl) on the respiratory rate in early and late stage of pregnant ewes .............. 47
3.4. Effect of anaesthetic protocol 2 (diazepam-thiopentone sodium) on the respiratory rate in early and late stage of pregnant ewes .............. 47
3.5. Effect of anaesthetic protocol 1 (xylazine-ketamine HCl) on the rectal temperature in early and late stage of pregnant ewe ............ 51
3.6. Effect of anaesthetic protocol 2 (diazepam-thiopentone sodium) on the rectal temperature in early and late stage of pregnant ewes .............. 51
3.7. Durations of the different phases following the induction of anaesthesia with selected anaesthetic protocols…………………..55
3.7.1. Duration of anaesthesia………………………………………………55
3.7.2. Lateral Recumbancy………………………………………………56
3.7.3. Sternal Recumbancy …………………………………………………57
3.7.4. Standing Phase ………………………………………………………58
3.8. General description of sedation, anaesthesia and recovery……..60
3.8.1. Sedative effects of the selected premedication…………………...60
3.8.1.1. Xylazine…………………………………………………………60
3.8.1.2. diazepam………………………………………………………..60
3.8.2. Induction of Anaesthesia…………………………………………60
3.8.3. Regurgitation………………………………………………………..60
3.8.4. Ruminal Tympany……………………………………………………60
3.8.5. Respiratory Emergencies…………………………………………61
3.8.6. Recovery…………………………………………………………….61

CHAPTER FOUR: ………………………………………………………62
4. Discussion …………………………………………………………...62

CONCLUSIONS……………………………………………………………71

FUTURE PROSPECTS……………………………………………………73

REFERENCES…………………………………………………………….74

ARABIC ABSTRACT……………………………………………………91
# List of Tables

<table>
<thead>
<tr>
<th>Table</th>
<th>Page</th>
</tr>
</thead>
<tbody>
<tr>
<td>Table 1: Doses for the selected anaesthetic premedication</td>
<td>41</td>
</tr>
<tr>
<td>Table 2: Doses for the selected anaesthetic induction</td>
<td>41</td>
</tr>
<tr>
<td>Table 3: Effect of anaesthetic protocol 1 (xylazine-ketamine HCL) on the heart rate in early and late stage of pregnant ewes</td>
<td>44</td>
</tr>
<tr>
<td>Table 4: Effect of anaesthetic protocol 2 (diazepam-thiopentone sodium) on the heart rate in early and late stage of pregnant ewes</td>
<td>45</td>
</tr>
<tr>
<td>Table 5: Effect of anaesthetic protocol 1 (xylazine-ketamine HCL) on the respiratory rate in early and late stage of pregnant ewes</td>
<td>48</td>
</tr>
<tr>
<td>Table 6: Effect of anaesthetic protocol 2 (diazepam-thiopentone sodium) on the respiratory rate in early and late stage of pregnant ewes</td>
<td>49</td>
</tr>
<tr>
<td>Table 7: Effect of anaesthetic protocol 1 (xylazine-ketamine HCL) on the rectal temperature in early and late stage of pregnant ewes</td>
<td>52</td>
</tr>
<tr>
<td>Table 8: Effect of anaesthetic protocol 2 (diazepam-thiopentone sodium) on the rectal temperature in early and late stage of pregnant ewes</td>
<td>53</td>
</tr>
<tr>
<td>Table 9: Durations of the different phases resulted following induction of anaesthesia with different selected anaesthetic protocols in pregnant ewes at early and late stages of pregnancy</td>
<td>59</td>
</tr>
</tbody>
</table>
# List of Figures

<table>
<thead>
<tr>
<th>Figure</th>
<th>Description</th>
<th>Page</th>
</tr>
</thead>
<tbody>
<tr>
<td>Fig 1</td>
<td>Effect of xylazine-ketamine Hcl (protocol1) and diazepam-thiopentone sodium (protocol2) on heart rate of ewes at early and late stages of pregnancy</td>
<td>46</td>
</tr>
<tr>
<td>Fig 2</td>
<td>Effect of xylazine-ketamine Hcl (protocol1) and diazepam-thiopentone sodium (protocol2) on respiratory rate of ewes at early and late stages of pregnancy</td>
<td>50</td>
</tr>
<tr>
<td>Fig 3</td>
<td>Effect of xylazine-ketamine Hcl (protocol1) and diazepam-thiopentone sodium (protocol2) on rectal temperature of ewes at early and late stages of pregnancy</td>
<td>54</td>
</tr>
<tr>
<td>Fig 4</td>
<td>Comparison between the duration of the anaesthetic phase resulted after application of xylazine-ketamine Hcl on ewes at early (p1-early) or late (p1-late) stages of pregnancy and diazepam-thiopentone sodium on ewes at early (p1-early) or late (p1-late) stages of pregnancy</td>
<td>55</td>
</tr>
<tr>
<td>Fig 5</td>
<td>Comparison between the duration of the lateral recumbancy phase resulted after application of xylazine-ketamine Hcl on ewes at early (p1-early) or late (p1-late) stages of pregnancy and diazepam-thiopentone sodium on ewes at early (p1-early) or late (p1-late) stages of pregnancy</td>
<td>56</td>
</tr>
<tr>
<td>Fig 6</td>
<td>Comparison between the duration of the sternal recumbancy phase resulted after application of xylazine-ketamine Hcl on ewes at early (p1-early) or late (p1-late) stages of pregnancy and diazepam-thiopentone sodium on ewes at early (p1-early) or late (p1-late) stages of pregnancy</td>
<td>57</td>
</tr>
<tr>
<td>Fig 7</td>
<td>Comparison between the duration of the standing phase resulted after application of xylazine-ketamine Hcl on ewes at early (p1-early) or late (p1-late) stages of pregnancy and diazepam-thiopentone sodium on ewes at early (p1-early) or late (p1-late) stages of pregnancy</td>
<td>58</td>
</tr>
</tbody>
</table>
late (p1-late) stages of pregnancy and diazepam-thiopentone sodium on ewes at early (p1-early) or late (p1-late) stages of pregnancy.............58
Introduction

At present and world wide, sheep are becoming very important as a source of meat, milk, wool, additional income as well as a sign of tribal pride.

In Sudan quite a number of sheep breeds such as Desert, Nilotic, Zaghawa, Toposa and Fellata are very well known and famous for their high quality meat for local use and export (Alsayed A, 2001).

Like other animals, sheep are quite often exposed to injuries and other surgical affections in both confined and grazing environments.

Surgery is increasingly becoming carried out in sheep for both treatment and research purposes. The major surgical operations in sheep include Laparotomy, Rumenatomy, Caesarean Section, Castration, Laparoscopy as well as operations on head and limbs. It is now commonly agreed and adopted that successful surgical operations require carefully selected and well planned anaesthetic procedures.

In fact safe and reliable anaesthetic procedures are considered to be invaluable and indispensible prerequisites for a successful surgery.

It is now commonly accepted that ideal veterinary anaesthesia is carried out in three integrated phases, namely: pre-anaesthetic medication, anaesthetic induction and anaesthetic maintenance. Quite a number of drugs are now tested and licenced for use to bring about the mentioned phases.

Preanaesthetic medication is usually carried out using specific drugs to calm the animal before anaesthetic induction, to reduce pain and to counteract the
undesired anaesthetic related side effects as well as the reduction of the anaesthetic dose thereby increasing the margin of safety of the anaesthetic. Examples of such kind of drugs include: Phenothiazine derivatives, $\alpha_2$ adrenoceptor agonists, Benzodiazepines and Butyrophенones (Hall, Clarke and Trim, 2001).

Anaesthetic induction is usually brought about using rapidly acting intravenous anaesthetics such as Barbiturates, Ketamine HCL, Saffan and Propofol which are capable of rapidly inducing unconsciousness usually in one site-brain circulation time, with the aim of reducing the struggle and its consequent physical and physiological harmful effects (Hall, Clarke and Trim, 2001).

Anaesthetic maintenance is usually carried out using inhalation anaesthetics such as Halothane, Enflurane and Isoflurane particularly for long duration operations due to their advantages of being safe, with fast and smooth recovery and allowing easy control over the depth of anaesthesia (Hall, Clarke and Trim, 2001).

In Sudan and many other third world countries, inhalation anaesthesia is still lagging behind in veterinary medicine because of inavailability of anaesthetic drugs, anaesthetic machines and their accessories, lack of a continuous cash flow to support anaesthetic practice and training as well as the lack of servicing facilities.

Taking into consideration the above mentioned problems facing inhalation anaesthesia in Sudan and many other third world countries, total intravenous anaesthesia will still have to play a vital role particularly under field conditions where no inhalation anaesthetic facilities are available.
Total intravenous anaesthesia in pregnant sheep must be carried out using combination of drugs that guarantee the reliability and safety for both mother and fetus. Taking into considerations the diversity of currently available drugs, the selection of excellent combinations of drugs for a safe total intravenous anaesthesia in pregnant sheep still represents a real challenge for veterinary anaesthetists in many third world countries.

**Objectives:-**

Because of the diversity of the newly presented drugs in veterinary anaesthesia and the scanty information available in the literature, the objectives of this investigation are as follows:

1. To assess by comparing and contrasting the effects of selected anaesthetic protocols on selected physiological and anaesthetic parameters.

2. To determine the reliability and safety of the selected anaesthetic protocols on the basis of their effects on pregnancy in ewes at early and late stages of pregnancy.

The selected anaesthetic protocols for total intravenous anaesthesia to be investigated in this study are:

1. Xylazine HCL (2%) + Ketamine HCL (5%) were injected in ewes at early stage of pregnancy.

2. Xylazine HCL (2%) + Ketamine HCL (5%) were injected in ewes at late stage of pregnancy.

3. Diazepam (0.5%) + Thiopentone sodium (2.5%) were injected in ewes at early stage of pregnancy.
4. Diazepam (0.5%) + Thiopentone sodium (2.5%) were injected in ewes at late stage of pregnancy.
CHAPTER ONE

1. Review of Literature

The ewe's reproductive activity is controlled by photoperiod (the number of hours of daylight). In temperate climates, when day length becomes shorter and temperatures become cooler, this change triggers the ewe's brain to release hormones to begin the reproductive process (Susan Schoenian, 2005). The further away from the equator that a sheep breed originated, the more likely they are to exhibit seasonal breeding patterns. Conversely, sheep developed in the tropics or sub-tropics are likely to exhibit estrus behavior throughout the year. Through selection and management, the sheep's natural breeding patterns have been altered (Susan Schoenian, 2005).

During their fertile period, ewes will come into estrus (heat) every 17 days until they are bred or their fertile period is over. Only during the estrus the ewe will allow a ram to mate with her. Usually the ewe is pregnant for 142 to 152 days (about five months). Pregnancy is also called gestation. Since ewes are only pregnant for five months, it is possible for ewes to give birth to lambs at an interval of every six to eight months. However annual lambing (every 12 months) is most common (Susan schoenian, 2005).

The close contact between the developing embryo and the mother results in an opportunity for the conceptus to influence maternal physiology through endocrine mechanisms. In both ruminants (sheep, cattle) and nonruminants, the blastocyst, before it attaches to the endometrium, secretes substances which prolong the life span of the cyclic corpus luteum (CL) beyond the
period of the estrous cycle. The time at which it occurs is known as maternal recognition of pregnancy (MRP) (B. Hafez; E.S.E. Hafez, 2000).

In ruminants, the trophoblast of the developing conceptus blocks luteal regression by the production of interferons (IFNs) ensuring MRP (Flint APF, 1995).

During the latter half of gestation, changes occur in the genital tract, particularly in the vulva and vagina. The vulva becomes highly edematous and vascular. The vaginal mucosa is pale and dry during most of gestation but it becomes edematous and pliable towards the end of pregnancy (B. Hafez; E.S.E. Hafez, 2000).

As pregnancy progresses, the developing fetus is retained within the uterus by tight closure of the external os of the cervix. Highly viscid mucus seals the cervical canal. This so-called mucous plug of pregnancy liquefies and is discharged in strings immediately before parturition (B. Hafez; E.S.E. Hafez, 2000). The uterus undergoes gradual enlargement to accommodate the growing fetus, but the myometrium remains quiescent, thereby preventing premature expulsion. The mechanisms that permit the enormous increase in size are unknown but are probably hormonal (B. Hafez; E.S.E. Hafez, 2000).

Also during the course of pregnancy, the mother makes metabolic and growth adjustments to provide an adequate supply of nutrients for the development of the fetus. Maternal body composition, feed intake, energy consumption, and metabolism are altered during pregnancy, but the mechanisms responsible are not fully established. Recent evidence has implicated insulin-like growth factors and their binding proteins as playing
important roles in maternal adaptation, which guarantees an adequate supply of substrates of the developing fetus (Owens J.A., 1991).

The key hormone necessary for maintenance of pregnancy is progesterone and the corpus luteum (CL) persists throughout pregnancy in all farm animals except the horse. The source of progesterone during the latter half of pregnancy may be from placenta in ewes (B. Hafez; E.S.E. Hafez, 2000).

Various practical methods have been used for pregnancy diagnosis in sheep. Both pregnancy and fetal numbers are accurately diagnosed by using radiography after day 70 of the gestation. Rectal abdominal technique detects pregnancy with an accuracy of (66 to 100%) from day 49 to day 109 of gestation, however it has a low accuracy (17 to 57%) for determining multiple fetuses. Progesterone assays have a high sensitivity (88% to 100%) and a low specificity (60% to 72%) at day 16 to day 18. Estrone sulphate assay accurately detects pregnant ewe at day 30 to day 35 (Karen A, et al. 2001).

Ovine pregnancy specific protein B (OPSPB) assay accurately (100%) detects pregnancy from day 26 after breeding onwards. The accuracy of progesterone, estrone sulphate and OPSPB assays for determining fetal numbers is relatively low. A-mode and Doppler ultrasonic techniques accurately detect pregnancy during the last half of gestation. Fetal numbers can not be determined by A-mode ultrasound, while the Doppler technique needs experience to achieve high accuracy. Transrectal B-mode, real time ultrasonography identifies the embryonic vesicles as early as the day 12.8 after mating, but the sensitivity of the technique for pregnancy diagnosis is very low (12%) at less than 25 day after mating. Transabdominal B-mode
ultrasonography achieved high accuracy for pregnancy diagnosis (94% to 100%) and the determination of fetal numbers (92% to 99%) at day 29 to day 106 of gestation. Real-time, B-mode ultrasonography appears to be the most practical method. However, the price of the scanner may constraint its wide use all over the world (Karen A, et al. 2001).

Relaxation of the pelvic ligaments occurs gradually during the course of pregnancy but becomes more rapid with approaching parturition. This relaxation is more noticeable in the cow and ewe than in the mare, and is related to high levels of estrogens in late pregnancy and to the action of relaxin (B. Hafez; E.S.E. Hafez, 2000).

A unique feature of early mammalian development is the provision of nutrients from the maternal organism by way of the placenta. The placenta is an apposition or fusion of the fetal membranes to the endometrium to permit physiologic exchange between fetus and mother. The placenta differs from other organs in many respects. It originates as a result of various degrees of fetal-maternal interactions and is connected to the embryo by a cord of blood vessels. The size and functions of the placenta change continuously during the course of pregnancy, and the organ is eventually expelled. For the fetus, the placenta combines in one organ many functional activities that are separate in the adult (B. Hafez; E.S.E. Hafez, 2000).

Ewes usually give 1 to 3 lambs at each birth. Twin birth (two babies) is most common in well-managed flocks, though first time mothers are more likely to have single lambs. Ewes produce their largest litters of lambs when they are between 3 and 6 years of age (Susan Schoenian, 2005).
Because sheep are often raised in harsh environments, sometimes it is more desirable for ewe to have just one lamb. This is because there may not be enough food for the ewe to carry two lambs or produce milk for two lambs. Also if the flock has to travel far for food and water, it is better to have one big strong lamb, than two or three smaller lambs that may struggle to keep up and eventually die (Susan Schoenian, 2005).

1.1. **Total intravenous anaesthesia (T.I.V.A):**

Intravenous anaesthetics have been used for decades in animal species as part of a general anaesthetic technique, most commonly to induce general anaesthesia prior to maintenance with inhalational anaesthetic agents. In recent years the use of intravenous anaesthetic agents to maintain general anaesthesia has become more commonplace because of the development of intravenous anaesthetic agents with pharmacokinetic profiles suitable for use by infusion. Total intravenous anaesthesia (TIVA) is now a popular technique in humans, as it offers advantages over inhalational anaesthesia, eliminating the hazard of atmospheric pollution, and thus reducing operator exposure and environmental exposure. TIVA has also been developed and used in animals and several combinations of anaesthetic, analgesic and hypnotic drugs have been described for use clinically (Andrea Nolan, 2004).

1.1.1. **Justification for total intravenous anaesthesia:**

Anaesthetic induction is usually brought about using rapidly acting intravenous anaesthetics capable of rapidly inducing unconsciousness usually in one site-brain circulation time, with the aim of reducing the struggle and its consequent physical and physiological harmful effects (Hall, Clarke and Trim, 2001). Anaesthetic maintenance is usually carried out using inhalation
anaesthetics particularly for long duration operations taking into considerations their advantages of being safe and with fast and smooth recovery (Hall, Clarke and Trim, 2001). But in Sudan and many other third world countries, inhalation anaesthesia is still lagging behind in veterinary medicine because of inavailability of anaesthetic drugs, anaesthetic machines and their accessories, the lack of a continuous cash flow to support anaesthetic practice and training as well as the lack of servicing facilities.

General anaesthesia induces immobilization, relaxation, unconsciousness, and freedom from pain. It is indeed one of the miracles of medicine, without which modern surgical techniques could never have developed (Lumb & Jones, 1996). One of the main stimuli to the development of TIVA was an awareness that chronic exposure to tract to amounts of gaseous or volatile anaesthetic agents might have an adverse effect on the health of personal working in an operating theatre environment. Complete elimination of all anaesthetic molecules from the operating theatre area is impossible so it is necessary to know the ‘safe’ level of contamination. But pollution and toxicity are by no means the only indications for use of the intravenous route (Morgan, M, 1983). Taking into consideration the above mentioned problems facing inhalation anaesthesia in Sudan and many other third world countries, TIVA will still have a vital role to play particularity under field conditions, where no inhalation anaesthetic facilities are available.

It is therefore obvious that the TIVA has an important part to play in the armamentarium of the anaesthesitist and it was fortuitous that the main stimulus for increasing use of the method coincided with the introduction of suitable drugs and with the growth of the clinical science of pharmacokinetics (Morgan, M, 1983).
1.1.2. Techniques of total intravenous anaesthesia:

The aim of total intravenous anaesthesia is to achieve a concentration of the drug in the blood stream, which will result in the required depth of anaesthesia, and then to maintain that level of anaesthesia until the end of the procedure. The method of achieving this procedure is similar to that used during inhalational anaesthesia, in which, a relatively high concentration of anaesthetic drug is given initially and then it is reduced to maintenance level until the end of surgery. In TIVA, the above aims are achieved by an initial bolus injection of the agent (Morgan, M.1983). Doses of induction agents can be adjusted through the use of the appropriate preanaesthetic medications with the objectives of increasing the margin of safety and counteracting undesirable side-effects associated with the use of such induction agents (Hall, Clarke and Trim, 2001).

1.2. Anaesthetic premedication:

Premedication helps both the anaesthetist and the animal, for it makes induction and maintenance of anaesthesia easier for the anaesthetist while at the same time rendering the experience safer and more comfortable for the patient. It implies the administration, usually before, but sometimes at or immediately after, the induction of anaesthesia, of sedatives, anxiolytics and analgesics, with or without anticholinergics (Hall, Clarke and Trim, 2001). The classic aims of premedication are to relieve anxiety thus apprehension, fear and resistance to anaesthesia, to counteract unwanted side effects of agent used in anaesthesia. Such desirable effects may require modifications depending on the species of animal and on
the drugs used, to reduced the dose of anaesthetic and to provide extra analgesia (Hall, Clarke and Trim, 2001).

1.2.1. Chemistry and pharmacokinetics of the selected anaesthetic premedications:

1.2.1.1. Xylazine (Rompun, Anased)):

Xylazine is a 2-(2, 6-dimethylphenylamino)-4H-5,6 dihydro-1,3-thiazine (Lumb and Jones, 1996). It was developed in 1962 by Bayer (Leverkusen, Germany) as an antihypertensive agent. During clinical studies in people, xylazine was found to have excessive central nervous system depressant effects and it was subsequently introduced for veterinary use as a sedative, analgesic and muscle relaxant. Early accounts of its actions and uses in animals were reported in the late 1960's and early 1970's (Clarke & Hall, 1969; Kerr et al., 1972; Antonaccio et al., 1973; Klein & Baetjer, 1974). Today xylazine is commonly used alone or in combination with other anaesthetic drugs in many species. The use of an antagonist (e.g. yohimbine, atepamizole, idazoxan, or tolazoline) to reverse xylazine’s sedative effect has become popular and is a valuable tool in the management of anaesthesia where xylazine is used as an adjunct (S.A. Greene & J.C. Thurmon, 1988).

Xylazine was enthusiastically received as a sedative and over the past 20 years it has maintained its popularity as a generally reliable sedative and premedicant in a wide range of animal species (Clarke & Hall, 1969).

Xylazine is rapidly absorbed after intravenous injection and undergoes extensive biotransformation (Garcia-Villar et al., 1981). Different routes were used for the administration of xylazine including deep intramuscular route in cattle (England and Kock, 1988; Kitzman et al., 1982; Hall and
Clarke, 1992). Xylazine has also been administered epidurally in cattle as reported by (Tamara, L, et al., 2002; Lee, I, et al, 2003) and in ponies (Mervyn Maze et al., 1991). Xylazine is rapidly absorbed after i.m. injection and undergoes extensive biotransformation (Garcia-Villar et al., 1981). About 20 metabolites have been identified (Duhm, et al. 1969). Xylazine metabolism is rapid and its half life t ½ β ranges from 23 min in sheep to 50 min in horses (Garcia-Villar et al., 1981). The sensitivity of cattle to xylazine is not explained in terms of plasma kinetics of xylazine as the t ½ β is short (36 min). Sensitivity of xylazine may be related to metabolism resulting in long-acting metabolites or species difference in the number of alpha-receptors (Garcia-Villar et al., 1981). Xylazine takes place in the liver and its metabolites are excreted through the kidney (Hall, L.W and Clarke, K.W. 1982).

1.2.1.2. Diazepam (Valium)

Diazepam is a benzodiazepine derivative developed through original Roche research. Chemically, diazepam is 7-chloro-1, 3-dihydro-1-methyl-5-phenyl-2H-1,4-benzodiazepin-2-one. It is a colourless crystalline compound, insoluble in water and has a molecular weight of 284.74. Intravenous injection may give rise to thrombophlebitis and this is thought to be due to solvents rather than to diazepam itself. An emulsion specially prepared for intravenous injection is claimed not to be irritant to veins, but the bioavailability of this preparation is reduced compared with that of the other formulations. Because of the problems of solubility, diazepam should not be diluted with water or mixed with solutions of other drugs (Hall, Clarke and Trim, 2001). A rise in plasma concentration, coupled with a return of clinical effects, occurs 6-8 hours after administration and is thought to be due to
Diazepam rapidly passes the blood-brain and placental barriers. Ninety-six percent of the drug is protein-bound (van der Kleijn E, et al. 1971). Diazepam is metabolized by demethylation and hydroxylation in people, dogs and rats to N-desmethyldiazepam, 3-hydroxydiazepam, and oxazepam. These metabolites are pharmacologically active (Schallek W, Schlosser W, Randall LO, 1972). Approximately 70% of diazepam is excreted in urine as conjugated metabolites and 10% in feces.

Premedication with diazepam increase the length of action of other anaesthetic agents and the drug is particularly useful prior to ketamine anaesthesia to reduce the hallucinations which were seen to occur with this dissociative anaesthetic agent (Hall, Clarke and Trim, 2001).

1.2.2. Pharmacodynamics and side effects of the selected anaesthetic premedications:

1.2.2.1. Xylazine:

Xylazine provide light to heavy sedation according to the dose rate administered. The use of this agent alone provides satisfactory sedation for restraint or it can be used for premedication prior to induction of anaesthesia with other anaesthetic agents. Dose rates for xylazine in sheep is 0,02 to 0,2 mg/kg, the largest dose producing profound sedation for many hours (Hall, Clark and Trim, 2001).
Xylazine HCl has variable effects on the cardiovascular system. In many species intramuscular or intravenous injection of xylazine induces hypotension and bradycardia (Adams, 2001) and temporary hypertensive in camels (Ramadan, 1994). Xylazine administered intravenously significantly decreases heart rate in the horse reversed by injection of thiopentone sodium with the ultimate result of an increase in the heart rate as recovery from anaesthesia is approached (Brower et al., 1980). Also the intravenous administration of xylazine in goat kids resulted in an immediate bradycardia at 5 minutes post injection (Nuha O.M., 2004; Ghurashi, 1999).

Xylazine induced decrease in heart rate and cardiac output are moderated by ketamine’s sympathomimetic action, while blood pressure and systemic vascular resistance are increased. When injected intravenously as a bolus, xylazine induces bradycardia and a brief period of hypertension (5-10 minutes), followed by a longer-lasting decrease in cardiac output, a decreased blood pressure and a decreased respiratory rate in horses (lumb and Jones, 1996; S.A.Greene & J.C.Thurmon, 1988). Also the administration of anaesthetic protocol xylazine-ketamine intravenously caused bradycardia in horses (Muir, W.W.et al 1977). Waterman, A.E. (1981) and Rings D.M. and Muir W.W. (1982) who studied the cardiopulmonary effects of xylazine-ketamine in calves also they found decreasd in heart rate.

There was a significant decrease in the heart rate which occurred at 5, 15, 20 and 25 minutes post anaesthetic induction after administration of protocol xylazine-ketamine HCl in sheep lambs (Thoria E.H.R., 2005).

A range of xylazine doses alters respiratory mechanics and gas exchange, causing tachypnoea, increased airway pressures and respiratory resistance,
decreased lung compliance, pulmonary oedema and hypoxaemia with or without hypercapnia (Mitchell & Williams 1977; Aziz & Carlyle 1978; Uggla & Lindquist 1983; Jansen et al. 1984; Doherty et al. 1986; Waterman et al. 1987; Hsu et al. 1989; Nolan et al. 1990; Papazoglou et al. 1994; Celly et al. 1997b; Bacon et al. 1998). When xylazine at a dose rate of 0.02-0.05mg/kg, was given intravenously to sheep during halothane anaesthesia, airway pressure increased and PaO₂ values markedly decreased and when xylazine administered to conscious sheep (0.05mg/kg,i.v) has resulted in hypoxemia (S.A.Greene & J.C.Thurmon, 1988).

Administration of anaesthetic protocol l xylazine-ketamine in goat kids resulted in a non significant increase in respiratory rate (Nuha M.O., 2004). Xylazine caused excessive salivation in ruminants due to decreased swallowing (Knight, 1980; Nowrouzian et al. 1981; Aminkov & Hubenov 1995; Nuha, 2004).

Xylazine resulted in none significant changes in rectal temperature in sheep lambs (Thoria, E.H.R., 2005).

Administration of xylazine decreased the rectal temperature in different animal species (Robertson Carter et al 1990). The pre-administration of xylazine had no significant effect on rectal temperature in buffaloes (Kumar and Sharma, 1986). Apparently, the effect of xylazine on the body temperature of cattle was variable and might depend on the size of the dose administered (Adams, 2001).
The effects of xylazine on the digestive tract are variable and include reduced rumen motility (Toutain et al. 1982; Deghani et al. 1991; Mohammad et al. 1996) and ruminal tympany (Ruckebush & Allal 1987).

Xylazine has been given to mares during all stages of pregnancy and has resulted in normal foals with no abortions (Tronicke & Vocke, 1970). However, xylazine caused increased myometrial tone and increased intra-uterine pressure in cows (LeBlanc et al., 1984). Xylazine-induced increased intra-uterine pressure is likely to be mediated by the $\alpha_2$ adrenoceptor stimulation as this effect was abolished by administration of yohimbine but not by prazosin (an $\alpha_1$ adrenoceptor antagonist) (Rodriguez-Martinez et al., 1987). The manufacturer warns that xylazine may precipitate early parturition or abortion in cattle when used in the last trimester of pregnancy (S.A. Greene & J.C. Thurmon, 1988).

Xylazine caused decreased insulin concentration in horses (Thurmon et al., 1982; Tranquilli et al., 1984a; Greene et al., 1987); in cattle (Symonds & Mallinson, 1978; Eichner et al., 1979); in sheep (Brockman, 1981) and in dogs (Goldfine & Arieff, 1979; Benson et al., 1984). This effect was apparently mediated by $\alpha_2$ receptors in pancreatic beta cells which inhibit insulin release (Hsu & Hummel, 1981). Xylazine caused increased plasma glucagons concentrations in sheep (Brockman, 1981), but did not change glucagons concentrations in dogs (Goldfine & Arieff, 1979).

Increased urinary output following xylazine occurs in cattle (Thurmon et al., 1978), cats (Hartsfield, 1980), ponies (Trim & Hanson, 1986), and in horses (Greene et al., 1986; Thurmon et al., 1984). Glucosuria has been reported in cattle (Thurmon et al., 1978) and ponies (Trim & Hanson, 1986), but not in...
horses when a qualitative urinary glucose determination technique was used (Greene et al., 1986; Thurmon et al., 1984).

Other side effects of xylazine include: muscle twitching when sedation is deep; sweating in horses at the time sedation is diminishing; vomiting at the onset of sedation in dogs and cat; hyperglycaemia; decreased intraocular pressure and gut motility and increased urine production. Xylazine also causes uterine contractions and should not be used in late pregnancy for it may induce premature labour. Increase in uterine tone may contraindicate it in cattle or horses receiving ovum transplants since this may reduce the chance of implantation (Hall, Clark and Trim, 2001).

The degree of sedation or restraint produced by xylazine depends on the route of injection, dosage, and animal’s temperament (Riebold, 2001). Xylazine at a dose rate of 0.1mg/kg administered i.v produced deep sedation in goats that lasts for 30-35 minutes (Adams, 2001).

Xylazine administration caused sudden death in a stallion suffering from colic (Fuentes, 1978).

1.2.2.2. Diazepam:

The use of diazepam as a premedicant, sedative and tranquillizer is less well documented. Diazepam alone has not been widely used in large animals and in horses its muscle relaxing properties may be associated with induced panic (Muir et al., 1982; Rehm & Schatzmann, 1984). At clinical doses, it causes only minimal respiratory and cardiac depression. High intravenous doses cause a slight decrease in respiration, blood pressure, and left ventricular stroke work. An increase in heart rate and a decrease in cardiac
output can also occur. In horses, cardiopulmonary dynamics were unchanged with doses of 0.05 to 0.4 mg/kg i.v injection. However, doses exceeding 0.2 mg/kg frequently produced recumbency. Smaller doses resulted in muscular fasciculations, weakness, and ataxia. Diazepam half-life values ranged from 6.94 to 21.6 hours (Muir WW, Sams RA, Huffman RH, Noonan JS, 1982).

The effects of diazepam in domestic animals have been poorly documented and the sedative and hypnotic effects appear to be minimal or absent in fit healthy dogs. Attempts to use it for hypnosis or as an intravenous induction agent in this species of animal have been unsuccessful. Lees, P, (1979) recorded a personal communication from Mackenzie and reported that the administration of doses of 2mg/kg given intravenously to greyhounds resulted in no obvious sedation, marked ataxia, and violent struggling when restrained. It has proved to be useful for postoperative sedation in dogs and provided pain (Hall and Clark, 1991).

Diazepam has a major role in veterinary practice in control of convulsions of any origin. Averill (1970) recommenda that dogs in status epilepticus should be given 5 mg by slow intravenous injection followed, if necessary, by a further 5 mg, and more recently doses of 10-35 mg have been recommended for this purpose (Hall, Clarke and Trim, 2001).

The toxicity of diazepam is relatively low. No acute hepatotoxic or nephrotoxic effects being apparent. In rabbits the LD50 for intravenous diazepam is 60 mg/kg. Overdoses should be treated with the benzodiazepine antagonist flumazenil and supportive measures including intravenous fluids and an adequate airway (Tranquilli WJ, Lemke K, Williams LL, Ballard G, Ko JCH, Benson GJ, Thurmon JC, 1992).
Hypotension may be alleviated by the use of levarterenol or metaraminol. Physostigmine (0.25 to 1.50 mg/kg IV or IP) has been reported as an antidote in rats, rabbits, and cats (Nagy J, Decsi L, 1979).

1.3. Anaesthetic Induction:

General anaesthesia is a state of unconsciousness produced by a process of controlled, reversible intoxication of the central nervous system whereby the patient neither perceives nor recalls noxious stimuli. Thus, an anaesthetic agent may be defined as a substance which, produces loss of consciousness in a controllable manner. It may be a volatile or gaseous substance inhaled into the lungs, or a non-volatile compound usually administered by intravenous injection but which may be given by mouth, per rectum or by intraperitoneal or intramuscular injection (Hall and Clark, 1991). Intravenous agents are popular in veterinary anaesthesia for all species of animal and this is undoubtedly partly due to the speed and pleasantness with which they produce unconsciousness (Hall and Clark, 1991).

1.3.1. Chemistry and Pharmacokinetics of the selected anaesthetic induction agents:-

1.3.1.1. Ketamine (Vetalar, Ketalar, Ketaset, Ketaject):

The term dissociative anaesthesia is used to describe an anaesthetic state induced by drugs that interrupt ascending transmission from the unconscious to conscious parts of the brain, rather than by generalized depression of all brain centers (Corssen G, Miyasaka M, Domino EF, 1968). Phencyclidine, ketamine, and tiletamine are used to induced taming and immobilization as

Ketamine is a 2-(0-chlorophenyl)-2-(methylaminocyclo-hexanone). The molecule exists as two optical isomers and the racemic mixture is currently used clinically. It is water soluble and has a lipid solubility 5 to 10 times that of thiopental (White PF, Way WL, Trevor AJ, 1982; Corssen G, Reves JG, Stanley TH, 1988; Cohen ML, Trevor AJ, 1974; Hall and Clark, 1991). It is available in 10, 50 and 100 mg/ml strengths and is suitable for intramuscular or intravenous injection. The 10 mg/ml solution is made isotonic with sodium chloride. Ketamine rapidly crosses the blood-brain barrier, quickly entering the brain and the brain plasma concentration ratio becomes constant in less than 1 minute. Liver metabolism produces at least four metabolites which are excreted in urine; they may have some slight additive effect of the action of the parent drug (Hall and Clarke 1991).

Taylor et al. (1972) considered that 2mg/kg produced ideal anaesthesia for intrauterine surgery in sheep.

Ketamine is safer than thiopentone sodium in sick animals. When used alone, ketamine fails to produce good skeletal muscle relaxation but the addition of a sedative or a tranquillizer was reported to improve muscle relaxation quality of anaesthesia. Most commonly xylazine or diazepam is recommended as a preanaesthetic medication (Riebold, 2001).
Ketamine induced anaesthesia by functional disruption (dissociation) of the CNS through marked CNS stimulation or induction of a cataleptoid state (Adams, 2001).

1.3.1.2. Thiopentone sodium (Thiosol):

Thiopentone sodium is a mixture of monosodium salt of 5-ethyl-5-(1-methylbutyl)-2-thiobarbiturate and exsiccated sodium carbonate (6 part w/w) (Vickers, Schnieden and Woodsmith, 1984). It is one of the barbiturates that are all derivatives of barbituric acid, which has a short anaesthetic duration in almost all animal species and man (Brander, Pugh, Bywater and Jenkins, 1993). In UK thiopentone sodium was introduced into veterinary practice in 1937 (Sheppard & Sheppard, 1937; Wright, 1937). Studies of its pharmacology did not keep pace with progress in the clinical field and it was not until 1950s that any notable contribution to an understanding of its clinical pharmacology was made. Brodie and his co-workers followed the concentration of the drug in the urine and various body tissues both in dogs and man (Brodie, 1952; Brodie et al., 1951;1953). They were found that the liver and plasma concentrations of thiopentone, which were high almost immediately after a single injection, soon fell rapidly. The muscle concentration, although high almost immediately after injection, continued to rise for some 20 minutes, then fell. The fall being fairly rapid during the first hour but becoming progressively slower in the next 2 or 3 hours. In contrast to this, the concentration in the body fat, which was negligible at first, increased rapidly during the first hour and then more slowly until the maximum was reached in 3-6 hours (Hall, Clarke and Trim, 2001).
Therefore, the concentration of the thiopentone sodium in fat was obviously increase to expense that in the plasma and all other tissues. Although the brain concentration of thiopentone was below that of the blood plasma, both showed similar changes and therefore the depth of narcosis can be regarded as being related to the plasma concentration of the drug (Hall, Clarke and Trim, 2001).

From these finding it is clear that the factors which govern the duration and depth of narcosis of thiopentone after injection are the amount of the drug injected, speed of injection, rate of distribution of the drug in the non-fatty tissues of the body and rate of uptake of thiopentone by the body fat. The speed of injection and the quantity injected are related (Hall and Clarke 1991).

For small animals 1.25% and 2.5% freshly prepared solutions of thiopentone are used, depending upon the animal’s size (Lumb and Jones, 1996). Whenever convenient, the more dilute solutions should be used, since overdosing is less likely and irritation is less in the event of accidental perivascular injection (Lumb and Jones, 1996).

Retention or the administration of carbon-dioxide has the effect of reducing the plasma pH. Alteration of the plasma pH has a complex effect on the distribution of thiopentone in the body. If the plasma pH is lowered by carbon dioxide, the undissociated fraction of thiopentone sodium is increased and since only this fraction is fat soluble, the decrease in pH result in an increase in the uptake of the drug by the fatty tissue. (Hall, 1966).
1.3.2. Pharmacodynamics and side effects of anaesthetic induction agents:

1.3.2.1. Ketamine:

Ketamine differs from most other anaesthetics in that it does not depress ventilatory responses to hypoxia thus, arterial oxygenation and functional residual capacity are usually well maintained during ketamine anaesthesia (Domino EF, et al., 1977; Shulman D, Beardsmore Cs, Aronson HB, Godfrey S, 1985; Mankikian B, et al., 1986).

The cardiovascular action of ketamine is characterized by indirect cardiovascular stimulation. Heart rate and arterial pressure increase as a result of direct stimulation of the CNS, leading to increased sympathetic outflow (Wong, D.H.W. and Jenkins, L., 1974).

Cardiac arrhythmias were uncommon in animals under the influence of ketamine anaesthesia (Hall and Clarke, 1992).

Ketamine induced increasing in heart rate and blood pressure can be ascribed to the sympathomimetic activity of ketamine (Clarke, Martin and Short, 1982).

In llamas receiving intramuscular injections of xylazine-ketamine combination, the heart rate was reported to be decrease but there were no significant changes in blood pressure (Dubois et. al., 2004).

Mild respiratory depression has been reported, in clinical practice and this is usually manifested by an increased respiratory rate, which does not
compensate for a decreased tidal volume (Hall, Clark and Trim, 2001). In sheep, the respiration was shallow and rapid (30-70 minutes) following ketamine administration (Adams, 2001).

In llamas, respiratory depression and hypoxaemia were seen with high doses of xylazine-ketamine combination (0.8mg/kg of xylazine and 8mg/kg of ketamine) during the first 10 minutes of lateral recumbancy (Dubois et. al., 2004).

Administration of ketamine causes a declination of body temperature in cats by an average of 1.6 C. (Beck, et al., 1971). Combination of xylazine-ketamine (at a dose rate of 1.1mg/kg of xylazine and 6.6mg/kg of ketamine) which was injected intravenously in horses caused elevation in body temperature during recovery from anaesthesia (Adams, 2001).

Administration of anaesthetic protocol xylazine-ketamine in sheep lambs resulted a significant decrease in the heart rate which occurred at 5, 15, 20 and 25 minutes post anaesthetic induction. Highly significant bradycardia was recorded 10 minutes after anaesthetic injection. No significant changes in heart rate were observed 30 minutes post anaesthetic induction and continued to be so until full recovery was attained (Thoria, E.H.R., 2005).

Ketamine often causes increased salivation and secretion of respiratory tract mucous, which can easily be controlled by administration of an anticholinergic (Lumb&Jones,1996). Laryngeal and pharyngeal reflexes usually are weakened during ketamine anaesthesia (Hall&Clarck,1991).

Nevertheless, swallowing reflexes may be somewhat obtunded because most species can be intubated with relatively more defficulty when anaesthetized
with ketamine (Wright M, 1982). Careful airway management and endotracheal intubation should always be performed to prevent aspiration (Lumb and Jones, 1996).

Sheep was reported to become ataxic and settled into sternal recumbancy following i.v. administration of ketamine at a dose rate of 2mg/kg but it did not settle into lateral recumbancy and appeared to remain alert. Moreover, there was no evidence of analgesia at this dosage (Waterman and Livingston, 1987).

Pregnant ewes have been successfully anaesthetized for up to 2 hours with intravenous ketamine (2mg/kg) followed by continuous infusion of ketamine (0.2%ketamine in 5% glucose) at a rate of 4ml/min (Taylor P, et al., 1972). Levinson et al (1973) reported that ketamine increased uterine blood flow and did not produce deleterious effects on fetal cardiovascular or acid-base status. When given intramuscularly or intravenously to pregnant goats, ketamine rapidly traverses the placental membrane and increasing in both fetal heart rate and blood pressure occur. When used for induction, ketamine reportedly increases basal uterine tone and the intensity of contractions in both pregnant and non pregnant women. Nevertheless, human fetal mortality is less with ketamine than with other anaesthetics (White PE, 1990).

Ketamine crosses the placenta easily and its concentration in the fetus is approximately the same as the mother (Andy, et al., 1994).

The swallowing reflex is often preserved in animals receiving dissociative anesthetics. This may help prevent aspiration pneumonia if the animal regurgitates. However, this is not 100% and fasting and intubation are still
recommended when using these anesthetics. The animal's eyes will usually remain open and the corneas should be protected with a layer of ophthalmic petrolatum or other suitable ointment (WWW.ahc.umn.edu/rar/anesthesia.html.95k, 2009).

In dogs all reflexes except the ocular reflexes were depressed after the use of atropine-xylazine-ketamine combination (Clarke et. al., 1982).

1.3.2.2. Thiopentone sodium:

Thiopental rapidly reaches the central nervous system and its effects become apparent within 15-30s after injection. Concentrations of the drug in the plasma and cerebrospinal fluid run parallel so the depth of narcosis can be assumed to be dependent on, and vary with, the blood level. However, this relationship is not a simple linear one, as acute tolerance to the drug develops (Hall, Clark and Trim, 2001).

A period of apnoea usually followed rapid intravenous injection of thiopentone sodium and this may be due to the central nervous system depression as a result of initial high plasma concentration (Mark et al, 1965; Ayman, A.E, 2001). Diminishing in alveolar concentration lead to carbon dioxide retention in blood, which in turn leads to lowering of the blood pH (Hall and Clarke, 1991). Thiopentone does not produce laryngeal and bronchial spasm in deep anaesthesia (Tyagi et al, 1964).

Administration of thiopentone sodium for induction of anaesthesia resulted in tachycardia at different times such as during induction period in goats (Deppe and Aspe, 1992) or even during endotracheal intubations in goats (Rawling and Kolata, 1983). Thiopentone sodium appears to have a direct
depressing effect on the myocardium and in certain circumstances may produce cardiac arrhythmias (Hall and Clarke, 1992).

In horses, administration of thiopentone sodium was reported to induce moderate tachycardia (Tavernor and Lees, 1970).

In contrast, thiopentone sodium was reported in buffaloes to cause mild decrease in the heart rate which became pronounced at the maximal depth of anaesthesia (Kumar and Sharma, 1986).

Induction of thiopentone sodium alone resulted in a significant tachycardia which persisted during the whole course of anaesthesia in dogs (Ayman, A.E, 2001).

Thiopentone sodium has a non significant change on body temperature in goats (Singh and Kumar, 1988). However the observation after the administration of thiopentone sodium was a decrease in rectal temperature in buffaloes (Kumar and Sharma, 1986).

Monitoring of body temperature with combination of xylazine-thiopentone sodium showed a significant decrease in rectal temperature from 35 to 75 minutes and also at 85 and 90 minutes post anaesthetic induction in sheep lambs and a non significant change in rectal temperature with combination of medetomidine-thiopentone sodium in sheep lambs (Thoria E.H.R, 2005).

There is considerable disagreement among research workers concerning the effects of thiopentone on the cardiovascular system. This may be due to the varying methods used for determinations of such things as cardiac- output, differences of premedication, depth of narcosis and the degree of carbon
dioxide retention, as well as the speed of drug injection (Hall, Clark and Trim, 2001).

It appears to be generally agreed, in that the rapid intravenous injection of the drug causes a fall in blood pressure even in normovolaemic animals and that this can be serious in hypovolaemic states. After the initial fall, the blood pressure returns to about the normal level but often with a persistent tachycardia (Hall, Clark and Trim, 2001).

There is an apparent increase in the sensitivity of the laryngeal and bronchial reflexes under light thiopentone anaesthesia (Hall and Clarke, 1991).

The incidence of hepatic damage is related to the dose administered and so hepatic dysfunction always follows the use of large doses. However, the presence of liver damage does not contraindicate the drug as long as only minimal doses are employed. Uraemia increases the duration of thiopental narcosis and the drug should be used with care. Foetal respiration seems particularly sensitive to the depressant effects of thiopental (Hall, Clark and Trim, 2001).

1.4. Monitoring:

Physical signs of depth of anaesthesia: The most reliable indicator of anaesthetic depth is spontaneous movement or chewing motion upon painfull stimulation, which indicate inadequate anaesthetic depth. Active regurgitation evident as peristaltic movement of oesophagus in the neck at times accompanied by swallowing movements is another reliable sign of light level of anaesthesia. Passive regurgitation characterized by a
continuous flow of rumen fluid indicate deep anaesthesia (Christina Dart, 2005).

During light anaesthesia the eyes may be positioned dorso-laterally and muscle tone in the eyelid and palpebræle reflex may be present. A centrally positioned eye with dilated pupil, relaxed eyelid and absent palpebræle reflex may indicate deep anaesthesia (Christina Dart, 2005).

1.5. Recovery from anaesthesia

The animal should be placed prone at the end of anaesthesia. Regurgitation during anaesthesia is not a problem when the endotracheal tube is present and the cuff inflated to produce an airtight seal or the animal was fasting for 24-36 hours. Recovery is usually quiet and, unlike horses, ruminants may be in no hurry to stand after anaesthesia. Full control of swallowing and gastrointestinal motility may not return for several hours after xylazine administration and feeding must be delayed. Hay or grass and water may be allowed 3 hours after anaesthesia with most other agents used in these species (Hall, Clark and Trim, 2001).
CHAPTER TWO

2. Materials and Methods

2.1. Materials

2.1.1. Experimental animals

Sixteen clinically healthy ewes in the early and later late stages of pregnancy weighting 29 to 40 kg were included in this study. The animals were housed in closed pens at Khartoum University Teaching Hospital throughout the course of the study. They were fed on alfalfa (Medicago sativa), corn (Zea mays) and supplemented with concentrates and a mineral block inside the pens with free access to water. General examination of the animals was conducted before and after experimentation and routinely throughout the course of the study.

2.1.2. Drugs

2.1.2.1. Selected Preanaesthetic medications

Two preanaesthetic medications were used in this study namely, Xylazine hydrochloride (Rompun 2%, Riemser, Germany. Alfasan, Woerden-Holland) and diazepam (Valium 0.5%, Shaphar, Shanghai. China). Their vials were kept at temperature below 25°C according to the manufacture recommendations and protected from light.
2.1.2.2. Anaesthetic induction

Two anaesthetic induction agents were used in this study, ketamine hydrochloride (ketamine HCL 5%, Ketamax-50, India. Rotexmedica, Germany) and thiopentone sodium (Thiosol 2.5%, Neon, India). Thiopentone sodium was provided in glass vials each containing 500 mg of thiopentone sodium powder. The drugs were kept at temperature below 25°C and protected from light according to the manufacture recommendations.

2.1.3. Injection set

Plastic disposable of different capacities syringes (1 ml, 5 ml and 20 ml) were used for intravenous administration of the carefully calculated doses of the preanaesthetic medications and the anaesthetic induction agents. 1 ml syringes (Becton Dickinson, USA) were used for intravenous injection of xylazine hydrochloride, 5 ml syringes (Ameco, Egypt) were used for intravenous injection of diazepam and 20 ml syringes (Shanchuan, China) were used for intravenous injection of ketamine hydrochloride and thiopentone sodium. Disposable needles size 22G x 5/8” were used for carrying out the injections in all animals.

2.1.4. Monitoring tools

An ordinary stethoscope (Kenzmedico Co. Ltd. Japan) was used for monitoring the heart rate. Respiratory rate was monitored by using the stethoscope and/or observing the movement of the abdominal muscles in the flank region. Rectal temperature was monitored using a digital thermometer (Hartman). A watch was used for calculating the heart rate, respiratory rate.
Also a watch was used for monitoring the durations of the different anaesthetic parameters from induction of anaesthesia until recovery was attained.

2.2. Methods

2.2.1. Anaesthetic protocols and methods of injection

In this study each of the two preanaesthetic and anaesthetic drugs were used at selected dose levels. Xylazine Hcl (2%) was used at a dose rate of 0.1 mg/kg body weight.

Diazepam (0.5%) was used at a dose rate of 0.4 mg/kg body weight.

Ketamine Hcl (5%) was used at a dose rate of 22 mg/kg body weight.

Thiopentone sodium (2.5%) was used at a dose of 7.5mg/kg body weight in the animals at early stage of pregnancy and 10 mg/kg body weight in the animals at late stage of pregnancy, thiopentone sodium solutions were freshly prepared by dissolving 500 mg of the powder in 20 ml pyrogen-free sterile water for injection.

All drugs in this study were administered by intravenous route. The selected anaesthetic protocols used in this study are as follows:-

1. Xylazine Hcl (2%) + Ketamine Hcl (5%) were injected in ewes at early stage of pregnancy.

2. Xylazine Hcl (2%) + Ketamine Hcl (5%) were injected in ewes at late stage of pregnancy.
3. Diazepam (0.5) + Thiopentone sodium (5%) were injected in ewes at early stage of pregnancy.

4. Diazepam (0.5) + Thiopentone sodium (5%) were injected in ewes at late stage of pregnancy.

2.2.2. Experimental work

Twelve empty ewes were brought from University of Khartoum Farm and were artificially inseminated after synchronization of oestrus by vaginal sponges containing flurogestone acetate and pregnant mare serum gonadotropin (PMSG). The vaginal sponge was introduced into the vagina and left for a days and at the 10th day the sponge was removed and the ewes immediately were injected intramuscularly with prostaglandin at a dose rate of 0.5 ml. Semen was collected at the day of insemination using the artificial vagina similar to that described by Evans and Maxwell (1987).

Semen samples were assessed for volume, colour and consistency, mass and individual motility, concentration, dead/live percentage and abnormal morphology.

Skimmed powdered milk was used as diluents for extending the fresh semen of rams for short-term storage. It was prepared a day before the collection of semen. 10 gms of skimmed milk powder (Zboi ureeno pro spot ebu v zemich mimo EU) and 0.5 gms of glucose (Merek Co. Germany) were dissolved in 100 ml of bidistilled water and stirred by glass rod and heated to 92-95 C° in a water bath for 10 minutes. Then cooled to room temperature and cooled for 24 hours at + 4C° on the day of semen collection, streptomycin (50 mg) and penicillin (50000 IU) were added (Elmubark, 2001).
The dilution was done with both diluents and semen in the water bath at 37°C by sucking the required of diluents into calibrated pipette and adding it slowly to the semen, so the final concentration was $370 \times 10^6$ active sperm/ml. After that the mixture was gently shaken and the percentage of progressively motile sperms were assessed again after dilution. The diluted semen was sucked into a 0.5 ml polyvinyl straws for insemination (Elmubark, 2001).

The treated ewes is inseminated with 0.5 ml of fresh diluted semen containing 250 million sperms, after 48-52 hours from the end of hormonal treatment without observing occurrence of oestrous (Fixed time insemination). The semen was deposited intra cervical as long as cervix allowed the passage of the insemination gun (Elmubark, 2001).

As the ewe was inseminated it was restrained with the hind quarters held up and forelimbs on the ground between the assistant legs as described by Gordon (1983).

After 40 days following insemination, a transabdominal ultrasonographic test for diagnosis of pregnancy was performed, nine ewes were detected pregnant and then eight ewes were chosen for this study.

The other eight ewes at early stage of pregnancy detected using transabdominal ultrasonography were brought from University of Khartoum Farm.

The sixteen early pregnant ewes in our study were randomly divided into two groups, eight animals in each group. The values of physiological parameters including respiratory rate, heart rate and rectal temperature were recorded immediately before injection of selected anaesthetic protocols to
represent base line values. Hair over the jugular vein was clipped and shaved using clippers and shaving blades and the skin was aseptically prepared by swabbing with a suitable antiseptic. The jugular vein was exposed and raised by applying a digital pressure at the base of the jugular groove and the needle was placed inside the vein. Correct placement of the needle was indicated by a free flow of blood through the needle. These procedures were repeated in each animal. A total of four experiments were conducted in this study.

2.2.2.1. Experiment (1)

A first group which consists of eight early pregnant ewes, was premedicated with xylazine Hcl at a dose of 0.1 mg/kg body weight given slowly intravenously followed after 15 minutes by a rapid intravenous injection of ketamine Hcl at a dose of 22 mg/kg body weight.

2.2.2.2. Experiment (2)

A second group which consists of eight early pregnant ewes, was premedicated with diazepam at a dose of 0.4 mg/kg body weight given slowly intravenously followed after 15 minutes by a rapid intravenous injection of thiopentone sodium at a dose of 7.5mg/kg body weight.

2.2.2.3. Experiment (3)

After day 100-130, the first group which consists of eight ewes previously used in experiment (1) were premedicated again with xylazine Hcl at a dose of 0.1 mg/kg body weight given slowly intravenously followed after 15 minutes by a rapid intravenous injection of ketamine Hcl at a dose of 22 mg/kg body weight.
2.2.2.4. Experiment (4)

After day 100-130, the second group which consists of group of eight ewes previously used in experiment (2) were premedicated again with diazepam at a dose of 0,4 mg/kg body weight given slowly intravenously followed after 15 minutes by a rapid intravenous injection of thiopentone sodium at a dose of 10 mg/kg body weight.

Throughout each experiment, all changes in treated ewes were observed and recorded to study the nature of the effects brought about by premedication and anaesthetic induction. The physiological parameters such as the respiratory rate, heart rate and rectal temperature were recorded as described by Kelly (1984). These parameters were taken at 5 minutes intervals from time of premedication, after induction and throughout the duration of anaesthesia until total recovery from anaesthesia was attained.

2.2.3. Assessment of reflexes:

2.2.3.1. Palpebral reflex:

The presence or absence of the palpebral reflex is was tested by tapping the skin at the medial canthus of the eye or by running the finger along the eye lashes. The reflex disappears in light to medium plane of surgical anesthesia (Batoul, 1990).

2.2.3.2. Jaw reflex:

The jaw reflex was assessed by opening the jaws and the reflex was considered to be regained when the jaws remain passively open even after releasing them free (Lumb and Jones, 1996).
2.2.3.3. Tongue reflex:

The tongue reflex was assessed by pulling the tongue outside the mouth. The reflex was considered to be regained when the animal became capable of drawing the tongue inside the buccal cavity (Kitzman et al., 1982).

2.2.3.4. Cough reflex:

The cough reflex was considered positive when animal responded by coughing as a result of external pressure on the trachea (Blood, Rodistits and Handerson, 1994).

2.2.3.5. Swallowing reflexes:

This reflex was tested by applying external gentle pressure on the larynx or at the base of the tongue. It is considered positive when swallowing is seen (Rawaling and Kolata, 1983).

2.2.3.6. Pedal (digital withdrawal) reflex:

This reflex was assessed by applying digital pressure on the skin web between the claws and is considered positive when the animal withdraws its limb as a response (Hellebrekers and Sap, 1997).

2.2.4. Definitions and monitoring of different anaesthetic parameters and phases:

2.2.4.1. Injection time

Injection time was recorded when completing the injection of each of the individual anaesthetic drugs.
2.2.4.2. Induction of anaesthesia

The administration of a general anaesthetic comprises the induction of a reversible state of unconsciousness with amnesia, as well as muscle relaxation and analgesia (Jonathan, M.B; Petru, G; Lesley, B. 2007).

2.2.4.3. Anaesthetic phase

It is the period during which the animal showed signs of unconsciousness, becomes slow and regular (Gray, 1960).

2.2.4.4. lateral recumbancy phase

It is the phase during which the animal opens its eyes and the major reflexes are regained but it is incapable of adopting sternal position (Lumb and Jones, 1984).

2.2.4.5. Sternal recumbancy phase

It is the stage at which the animal succeeds to adopt sternal recumbancy without falling (Lumb and Jones, 1984).

2.2.4.6. Standing phase

In this investigation it is taken as the period at which the animal can stand but it’s incapable of walking.

2.2.4.7. Recovery phase

In this investigation the animal is considered to have recovered from anaesthesia when it is able to walk without falling.
2.2.4.8. Total recovery time

It is the summation of the time of the different phases from induction of anaesthesia to recovery (Nuha, 2004).

2.2.5. A pilot study

Some clinical observations revealed that, most of the commonly used doses of some anaesthetic drugs as recommended by (Hall, Clark and Trim, 2001). Such as xylazine and ketamine were found to be less effective and less reliable for anaesthetic induction in Sudanese sheep. Such observations indicated that it was indespensible and in valuable to conduct a pilot study with aim of testing and determining the accurate, safe and reliable doses of the two selected anesthetic protocols namely xylazine-ketamine and diazepam-thiopentone sodium.

The pilot study was carried out using a total of sixteen non pregnant ewes randomly devided into two groups, eight animals in each group. Anaesthetic induction was carried out in each group using gradually increasing doses of one of the selected anaesthetic protocols.

On the basis of the results of the pilot study in the empty ewes, the dose of ketamine (10 mg/kg) was proved to be not effective with the conclusion that a dose of 22 mg/kg was the dose of choice in pregnant ewes. On the other hand the dose of thiopentone sodium in the non pregnant ewes (10 mg/kg) was found to be with very dressing effects on respiration in ewes at early stages of pregnancy with thw conclusion that a dose of 7.5 mg/kg was selected for use in ewes at early stage of pregnancy and a dose of 10 mg/kg for use in ewes at late stage of pregnancy.
The correct, safe and reliable doses of the two protocols selected for use in this investigation were shown in table 1 and 2.

Tabe (1): Doses for the selected anaesthetic premedication

<table>
<thead>
<tr>
<th>Anaesthetic premedication</th>
<th>Stage of pregnancy</th>
<th>Concentration (%)</th>
<th>Doses Mg/kg.b.w</th>
</tr>
</thead>
<tbody>
<tr>
<td>Xylazine Hcl</td>
<td>Early</td>
<td>2%</td>
<td>0.1 mg/kg.b.w</td>
</tr>
<tr>
<td>Xylazine Hcl</td>
<td>Late</td>
<td>2%</td>
<td>0.1 mg/kg.b.w</td>
</tr>
<tr>
<td>Diazepam</td>
<td>Early</td>
<td>0.5%</td>
<td>0.4 mg/kg.b.w</td>
</tr>
<tr>
<td>Diazepam</td>
<td>Late</td>
<td>0.5%</td>
<td>0.4 mg/kg.b.w</td>
</tr>
</tbody>
</table>

Tabe (2): Doses for the selected anaesthetic induction

<table>
<thead>
<tr>
<th>Anaesthetic induction</th>
<th>Stage of pregnancy</th>
<th>Concentration (%)</th>
<th>Doses Mg/kg.b.w</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ketamine Hcl</td>
<td>Early</td>
<td>5%</td>
<td>22 mg/kg.b.w</td>
</tr>
<tr>
<td>Ketamine Hcl</td>
<td>Late</td>
<td>5%</td>
<td>22 mg/kg.b.w</td>
</tr>
<tr>
<td>Thiopentone sodium</td>
<td>Early</td>
<td>2.5%</td>
<td>7.5 mg/kg.b.w</td>
</tr>
<tr>
<td>Thiopentone Sodium</td>
<td>Late</td>
<td>2.5%</td>
<td>10 mg/kg.b.w</td>
</tr>
</tbody>
</table>
2.2.6. Statistical Analysis:

Data were subjected to analysis of variance followed by fisher’s protected least significant difference test by using StatView (Abacus concepts Inc., Berkeley CA, USA). \( p \leq 0.05 \) was considered to be statistically significant.
CHAPTER THREE

3. Results

3.1. Effect of the anaesthetic protocol 1 (xylazine-ketamine HCL) on the heart rate in early and late stages of pregnancy in ewes:

As shown in Table (3), with this anaesthetic protocol the heart rate was significantly (p≤0.05) decrease at 5, 15 minutes after premedication with xylazine and restore to normal immediately after application of ketamine HCL in ewes at early stage of pregnancy (Figure 1). The heart rate was significantly (p≤0.05) decrease at 5, 10, 15 minutes after premedication with xylazine and significantly (p≤0.05) increase at 20 minutes after application of ketamine HCL and significantly (p≤0.05) decrease at 25, 30, 35 minutes after application of ketamine HCL in ewes at late stage of pregnancy (Figure 1).

3.2. Effect of anaesthetic protocol 2 (diazepam-thiopentone sodium) on the heart rate in early and late stages of pregnancy in ewes:

This anaesthetic protocol resulted in significant changes in heart rate as shown in Table (4). There was a significant (p≤0.05) decrease in the heart rate which occurred at 5 minutes after premedication with diazepam and also there was a significant (p≤0.05) decrease in the heart rate which occurred at 20, 25 minutes after application of thiopentone sodium in ewes at early stage of pregnancy (Figure 1). There was no significant difference in heart rate in ewes at late stage of pregnancy (Figure 1).
Effect of xylazine-Ketamine HCl (protocol 1) and Diazepam-Thiopentone Sodium (protocol 2) on Heart rate of ewes at early and late stages of pregnancy
3.3. Effect of anaesthetic protocol 1 (xylazine-ketamine HCL) on the respiratory rate in early and late stages of pregnancy in ewes:

There was no significant difference in the respiratory rate in pregnant ewes at early stage of pregnancy when subjected to anaesthetic protocol 1 (xylazine-ketamine Hcl) as shown in Table (5), however the respiratory rate was significantly ($p \leq 0.05$) increased at 5 minutes after premedication with xylazine in ewes at early stage of pregnancy (Figure 2). The respiratory rate was significantly ($p \leq 0.05$) increased up to 15 minutes after premedication with xylazine and restore to normal immediately after application of ketamine HCL in ewes at late stage of pregnancy (Figure 2).

3.4. Effect of anaesthetic protocol 2 (diazepam-thiopentone sodium) on the respiratory rate in early and late stages of pregnancy in ewes:

With this protocol there was no significant difference in the respiratory rate in pregnant ewes at early stage of pregnancy when subjected to this anaesthetic protocol as shown in Table (6), however the respiratory rate was significantly ($p \leq 0.05$) increased at 5, 15 minutes after premedication with diazepam and also significantly ($p \leq 0.05$) increased at 20 minutes after application of thiopentone sodium in ewes at late stage of pregnancy (Figure 2).
Effect of xylazine-Ketamine HCl (protocol 1) and Diazepam- Thiopentone Sodium (protocol 2) on respiratory rate of ewes at early and late stages of pregnancy.

Respiratory Rate (Breath/min)

Time (min)

- Protocol 1 Early
- Protocol 1 Late
- Protocol 2 Early
- Protocol 2 Late
3.5. Effect of anaesthetic protocol 1 (xylazine-ketamine HCL) on the rectal temperature in early and late stages of pregnancy in ewes:

As shown in Table (7), there was no significant difference in the rectal temperature in pregnant ewes at early and late stage of pregnancy when subjected to this anaesthetic protocol (Figure 3).

3.6. Effect of anaesthetic protocol 2 (diazepam-thiopentone sodium) on the rectal temperature in early and late stages of pregnancy in ewes:

Monitoring of body temperature as shown in Table (8), there was no significant difference in the rectal temperature in pregnant ewes at early and late stage of pregnancy when subjected to this anaesthetic protocol (Figure 3).
Effect of xylazine-Ketamine HCl (protocol 1) and Diazepam-Thiopentone Sodium (protocol 2) on body temperature of ewes at early and late stages of pregnancy
3.7. Durations of the different anaesthetic phases following the induction of anaesthesia with selected anaesthetic protocols:

3.7.1. Duration of anaesthesia:

As shown in Fig (4), there was no significant difference in the anaesthetic phase duration when pregnant ewes at early or late stages were subjected to both anaesthetic protocols (protocol 1: xylazine-ketamine HCL, protocol 2: diazepam- thiopentone sodium).
3.7.2. lateral Recumbancy:

As shown in Fig (5), there was no significant difference in the lateral recumbancy phase in ewes at early stage of pregnancy when subjected to either protocol 1 (xylazine-ketamine HCL) or protocol 2 (diazepam-thiopentone sodium). However, the lateral recumbancy phase was significantly larger in ewes at late stage of pregnancy under protocol 2 (diazepam-thiopentone sodium) anaesthetic treatment compared to those under protocol 1 (xylazine-ketamine HCL) anaesthetic treatment.
3.7.3. Sternal Recumbancy:

As shown in Fig (6), there was no significant difference in the sternal recumbancy phase in ewes at early stage of pregnancy when subjected to either protocol 1 (xylazine-ketamine HCL) or protocol 2 (diazepam-thiopentone sodium). However the sternal recumbancy phase was significantly longer in ewes at late stage of pregnancy under protocol 2 (diazepam-thiopentone sodium) anaesthetic treatment compared to those under protocol 1 (xylazine-ketamine HCL) anaesthetic treatment.
3.7.4. Standing Phase:

As shown in Fig (7), there was no significant difference in the standing phase in ewes at early stage of pregnancy when subjected to either protocol 1 (xylazine-ketamine HCL) or protocol 2 (diazepam-thiopentone sodium). However the standing phase was significantly reduced in ewes at late stage of pregnancy under protocol 2 (diazepam-thiopentone sodium) anaesthetic treatment compared to those under protocol 1 (xylazine-ketamine HCL) anaesthetic treatment.

Comparison Between the Duration of the Standing Phase Resulted after application of xylazine-Ketamine HCl (P1) and Diazepam-Thiopentone Sodium (P 2) on ewes at early and late stages of pregnancy
3.8. General description of sedation, anaesthesia and recovery:

3.8.1. Sedative effects of the selected premedications:

3.8.1.1. Xylazine:

The total intravenous administration of xylazine at a dose rate of 0.1 mg/kg resulted after 30 second post injection in a walking movement where the animal took several unsteady steps as the first sign of sedation then the animal started lying down 3-5 minutes after xylazine injection.

3.8.1.2. Diazepam:

Four minutes following the total intravenous administration of diazepam at a dose rate of 0.4 mg/k, all animals at early or late stages of pregnancy were found to adopt sternal recumbancy position.

3.8.2. Induction of Anaesthesia:

Monitoring of the induction of the two anaesthetic protocols resulted a period of apnea after application of thiopentone sodium.

3.8.3. Regurgitation:

No regurgitation was observed in all pregnant animals at early or late stages of pregnancy when subjected to either protocol 1 (xylazine-ketamine HCL) or protocol 2 (diazepam-thiopentone sodium).

3.8.4. Ruminal Tympany:

Slight ruminal tympany was observed in all animals at early or late stages of pregnancy when subjected to either protocol 1 (xylazine-ketamine HCL) or protocol 2 (diazepam-thiopentone sodium).
3.8.5. Respiratory Emergencies:

Apparent apnoea was observed in all animals at early and late stages of pregnancy when subjected to thiopentone sodium as induction agents. This problem was successfully solved by stimulation of respiration by manual resuscitation.

3.8.6. Recovery:

Recovery from anaesthesia was attained after the end of narcosis and lateral recumbancy phase when all reflexes was regained.
CHAPTER FOUR

4. Discussion

As in other species sedation and anaesthesia are quite often required in sheep for surgical or diagnostic procedures. Induction of general anaesthesia may be influenced by sheep’s anatomical and physiological characteristics. The chief difficulties experienced during general anaesthesia with this species include excessive salivation, tympany and regurgitation. The ideal method of general anaesthesia would provide a reliable, rapid and smooth of induction, adequate hypnosis and analgesia for the surgical intervention, a rapid and uncomplicated recovery and minimal suppression of vital organ function throughout the anaesthetic procedure in pregnant ewes at early and late stages of pregnancy.

In this study anaesthetic premedication was carried out using two selected pre-anaesthetics medications namely, xylazine and diazepam. Anaesthetic induction was brought about using two selected anaesthetic induction agents namely, ketamine Hcl and thiopentone sodium.

This investigation is carried out to test the reliability and safety of total intravenous anaesthesia in pregnant ewes at early and late stages of pregnancy using two selected anaesthetic protocols which were xylazine-ketamine and diazepam-thiopentone sodium.
The scope of this investigation also covered the study of the effects of the two selected anaesthetic protocols on selected physiological and anaesthetic parameter.

Monitoring of the heart rate at 5 minutes interval following induction of anaesthesia with the anaesthetic protocol 1 xylazine-ketamine HCL as shown in Table (3) resulted in a significant decrease up to 15 minutes after induction in ewes at early and late stage of pregnancy. These findings are supported by the results obtained by Nuha M.O., 2004; Ghurashi 1999. Who reported the occurrence of bradycardia following anaesthesia with xylazine-ketamine in goat kids. In many species i.m or i.v injection of xylazine induces hypotension and bradycardia. (Adams, 2001). Xylazine administered intravenously significantly decreases heart rate in the horse. (Brower et al., 1980). Lumb and Jones, 1996), (S.A.Green & J.C.Thurmon, 1988). Also reported an immediate bradycardia following anaesthetic premedication with xylazine in horses.

There was a significant decrease in the heart rate at 5, 15, 20 and 25 minutes post anaesthetic induction with the protocol xylazine-ketamine HCl in sheep lambs. (Thoria E.H.R., 2005). Administration of xylazine intravenously induced significant decreases in heart rate in the horse. (Brower et al., 1980). Muir, W.W.et al 1977 also reported the occurrence of immediate bradycardia following anaesthesia with xylazine-ketamine in horses. The results of this investigation are also in accordance with the findings of Waterman A.E. (1981) and Rings D.M. and Muir W.W. (1982). Who studied the cardiopulmonary effects of xylazine-ketamine in calves. Also the administration of xylazine-ketamine in llamas was reported decrease in the heart rate. (Dubois et. al., 2004).
The heart rate restore to normal immediately after injection of ketamine HCL at the dose rate of 22 mg/kg in ewes at early and late stage of pregnancy. These finding are supported by the results obtained by Wong D.H.W. and Jenkins L. (1974). Who reported the occurrence of an increase the heart rate and arterial blood pressure as a result of direct stimulation of the CNS, leading to increased sympathetic outflow. Ketamine was also reported to increase the heart rate and blood pressure and such effects were ascribed to the sympathomimetic activity of ketamine. (Clarke, Martin and Short, 1982).

Induction of anaesthesia with the anaesthetic protocol 2 diazepam-thiopentone sodium as shown in Table (4) resulted in no significant difference in heart rate after premedication with diazepam in ewes at early and late stage of pregnancy. These results are in contrast with the results of injection of diazepam at clinical doses which reported to cause minimal cardiac depression at 0.05 to 0.4mg/kg intravenous injection doses cause increase in heart rate. (Muir W.W, Sams R.A, Huffman R.H, Noonman J.S, 1982). After injection of thiopentone sodium, the heart rate significantly decreased at 20, 25 minutes and no significant difference in heart rate was recorded at 30 minutes in ewes at early stage of pregnancy. These results support the finding of Kumar and Sharma, (1986) in buffaloes. However the results contrasted the findings of Ayman A.E, (2001), who reported that thiopentone sodium alone in dogs, resulted in a significant tachycardia. Moderate tachycardia was also reported in horses after the injection of thiopentone sodium. (Tavernor and Lees, 1970).

Monitoring the respiratory rate at 5 minutes interval following induction of anaesthesia with the anaesthetic protocol 1 xylazine-ketamine HCL as
shown in Table (5), showed no significant difference in the respiratory rate in pregnant ewes at early stage of pregnancy. These results supported the findings of Nuha M.O. (2004), who reported that administration of the anaesthetic protocol 1 xylazine-ketamine in goat kids resulted in a non significant increase in respiratory rate. However the respiratory rate was significantly ($p \leq 0.05$) increased up to 15 minutes after premedication with xylazine. These finding are supported by the results obtained by Mitchell & Williams 1977; Aziz & Carlyle 1978; Ugglä & Lindquist 1983; Jansen et al. 1984; Doherty et al. 1986; Waterman et al. 1987; Hsu et al. 1989; Nolan et al. 1990; Papazoglou et al. 1994; Celly et al. 1997b; Bacon et al. 1998).

These increase in respiratory rate restore to normal immediately after injection of ketamine HCL in ewes at late stage of pregnancy. These results were in agreement with those reported by Hall Clark and Trim (2001). In sheep respiration was also reported to be shallow and rapid (30-70 minutes) following ketamine administration. (Adams, 2001).

Induction of anaesthesia with the anaesthetic protocol 2 diazepam-thiopentone sodium as shown in Table (6), resulted in no significant difference in the respiratory rate in pregnant ewes at early stage of pregnancy. However the respiratory rate was significantly ($p \leq 0.05$) increased up to 15 minutes after premedication with diazepam and restore to normal immediately after injection of thiopentone sodium in ewes at late stage of pregnancy. Although diazepam at clinical doses was reported to cause minimal respiratory depression in horses, at high intravenous doses caused a slight decrease in respiration. (Muir WW, Sams RA, Huffman RH, Noonan JS, 1982). A period of apnoea usually followed rapid intravenous injection of thiopentone sodium and this may be due to the central nervous
system depression as a result of initial high plasma concentration. (Mark et al., 1965; Ayman, A.E, 2001).

Monitoring the rectal temperature at 5 minutes interval following induction of anaesthesia with the anaesthetic protocol 1 xylazine-ketamine HCL as shown in Table (7), showed no significant difference in the rectal temperature in pregnant ewes at early and late stage of pregnancy. These results were in agreement with those reported by Kumar and Sharma (1986). Who reported no significant effect on rectal temperature in buffaloes. Also the administration of xylazine was reported to cause no significant changes in rectal temperature in sheep lambs. (Thoria, E.H.R., 2005).

Although these finding is contrast to the result of administration of xylazine decreased the rectal temperature in different animal species. (Robertson Carter et al 1990). Apparently, the effect of xylazine on the body temperature of cattle was variable and might depend of the size of the dose administered. (Adams, 2001).

Induction of anaesthesia with the anaesthetic protocol 2 diazepam-thiopentone sodium as shown in Table (8), there was no significant difference in the rectal temperature in pregnant ewes at early and late stage of pregnancy. These finding support the result of administration of thiopentone sodium in goat according to Singh and Kumar (1988). And also the administration of medetomidine-thiopentone sodium revealed non significant change in rectal temperature in sheep lambs. (Thoria E.H.R, 2005).
Although the observation after the administration of thiopentone sodium was decrease in rectal temperature in buffaloes. (Kumar and Sharma, 1986). Monitoring of body temperature with combination of xylazine-thiopentone sodium showed a significant decrease in rectal temperature from 35 to 75 minutes and also at 85 and 90 minutes post anaesthetic induction in sheep lambs. (Thoria E.H.R, 2005).

As shown in Table (9), there was no significant difference in the anaesthetic phase duration when pregnant ewes at early or late were subjected both anaesthetic protocols (protocol 1: xylazine-ketamine HCL, protocol 2: diazepam-thiopentone sodium). Although these results are not similar to the findings of Nuha M.O. (2004). Using xylazine-thiopentone sodium and xylazine-ketamine in goat kids and the results in anaesthetic phase with significantly longer duration with xylazine-thiopentone sodium than that obtained when using xylazine-ketamine. The results of this investigation contrasted those of Kumar et al., (1983) and Kumar and Sharma (1986), who studied the effects of premedication with xylazine on thiopentone sodium anaesthesia in buffaloes. Also Gurashi (1999), who reported a significant prolongation of anaesthesia phase when anaesthesia was induced in goat kids with xylazine-ketamine.

Table (9), reflected no significant difference in the lateral recumbancy phase in ewes at early stage of pregnancy when subjected to either protocol 1 (xylazine-ketamine HCL) or protocol 2 (diazepam-thiopentone sodium). However the duration of the lateral recumbancy phase was significantly longer in ewes at late stage of pregnancy under protocol 2 (diazepam-thiopentone sodium) compared to those under protocol 1 (xylazine-ketamine HCL. Although these results are not similar to the findings of Nuha M.O.
(2004). Using xylazine-thiopentone sodium and xylazine-ketamine in goat kids and the results in duration of lateral recumbancy phase with significantly longer duration with xylazine-ketamine when compared with xylazine-thiopentone sodium.

The duration of sternal recumbency phase in Table (9), was found to show no significant difference in ewes at early stage of pregnancy when subjected to either protocol 1 (xylazine-ketamine HCL) or protocol 2 (diazepam-thiopentone sodium). However the sternal recumbancy phase was significantly longer in ewes at late stage of pregnancy receiving protocol 2 (diazepam-thiopentone sodium) compared to those injected with protocol1 (xylazine-ketamine HCL).

As shown in Table (9), there was no significant difference in the standing phase in ewes at early stage of pregnancy when subjected to either protocol 1 (xylazine-ketamine HCL) or protocol 2 (diazepam-thiopentone sodium). However the standing phase was significantly shorter in ewes at late stage of pregnancy under protocol 2 (diazepam-thiopentone sodium) compared to those under protocol 1 (xylazine-ketamine HCL).

Total recovery time is now considered to be among the most important factors governing the selection of anaesthetic protocols for the various surgical operations. A lot of surgeons prefer the use of anaesthetic protocols that can ensure adequate duration of anaesthesia for surgery and as a same time guarantee a fast, smooth and uneventful recovery from anaesthesia.

Although regurgitation of ruminal contents was generally accepted to represent a serious hazard in ruminants during general anaesthesia (Hall and Clark, 1982), it was found that it did not take place in almost all animals.
used in this study. This finding is in agreement with that obtained by Ghurashi, (1999) and Nuha, (2004) who reported the absence of regurgitation. This could be attributed to the small size of animals (goat kids) or due to the positioning of animals during anaesthesia (right lateral recumbancy), so that no great pressure was exerted on the rumen to force the ruminal contents to be regurgitated. Lack of regurgitation in pregnant ewes might encourage a lot of researchers to go for the selection of pregnant ewes as excellent experimental animals particularity for use with the selected anaesthetic protocols used in this study.

Slight harmless ruminal tympany was observed after induction of anaesthesia with all anaesthetic protocols that were used in this study. This tympany might be due to the absence of eructation together with occurrence of salivation.

Prolonged apparent apnoea was observed following induction of anaesthesia with diazepam-thiopentone sodium and it may be due to the depressive effect on respiratory rate of thiopentone sodium as reported by Mark et al, 1965; Ayman, A.E, 2001.

Xylazine has been given to pregnant ewes at early and late stages of pregnancy and has resulted in delivery of normal lamb kids with no abortions. These finding support the results of the administration of xylazine to mares during all stages of pregnancy which resulted in delivery of normal foals with no abortions (Tronicke & Vocke, 1970). On the other hands xylazine was reported to cause increased myometrial tone and increased intra-uterine pressure in cows (LeBlanc et al.,1984). Xylazine-induced increased intra-uterine pressure is likely to be mediated by the $\alpha_2$
adrenoceptor stimulation as this effect was abolished by administration of yohimbine but not by prazosin (an $\alpha_1$ adrenoceptor antagonist) (Rodriguez-Martinez et al., 1987). Also it was reported that xylazine may precipitate early parturition or abortion in cattle when used in the last trimester of pregnancy (S.A. Greene & J.C. Thurmon, 1988).
CONCLUSIONS

From the results obtained in this study and the discussion, it can be concluded that:

1. The anaesthetic protocols used in this study proved to be with less detrimental effects on cardiopulmonary systems and pregnancy.

2. A pilot study proved to be very vital for making reasonable estimation and calculations of the appropriate doses of the selected anaesthetic protocols used in this investigation.

3. Relatively larger doses of the selected anaesthetic protocols were recommended for use in pregnant ewes.

4. Xylazine induced decrease in heart rate can be moderated by ketamine sympathomimetic action.

5. The results of this investigation proved that xylazine is an excellent premedication in pregnant ewes and for research in such areas.

6. Ketamine containing anaesthetic protocols proved to be less detrimental with regard to respiration and can therefore can be considered more safe in pregnant ewes compared to thiopentone sodium containing protocols which were associated with apnoea and variable degrees of respiratory depression.
7. All anaesthetic protocols used in this study proved to be safe and reliable in pregnant ewes as indicated by the fact that not a single death and abortion was reported.

8. The findings of this investigation supported by previous findings indicated that relatively larger doses of the anaesthetic protocols are recommended in sheep compared to goats with the conclusion that "a sheep is not a goat".
FUTURE PROSPECTS

The findings of this investigation are expected to encourage more research in the following areas:-

1. Evaluation of the selected anaesthetic protocols for Caecarean Section in ewes with the aim of proving their reliability and safety for both foetus and mother.
2. Testing the selected anaesthetic protocols on other species of animals.
3. Testing the reliability of the selected anaesthetic protocols for various surgical operation.
REFERENCES


Ayman, A.E. (2001). Some aspects of anaesthesia in dogs using thiopentone sodium with or without selected 2 adrenoceptor agonist
and its reversal with atipamizole. M.Sc. Thesis, Faculty of Veterinary Medicine, U of K.


**Brodie, B.B; et al. (1953).** Fate of pentobarbital in man and dogs, and method for its estimation in biological material. Journal of Pharmacology and Experimental Therapeutics 109:26-34.


Dubois, W. R; et al. (2004). A comparison of two i.m doses of xylazine-ketamine combination and tolazoline reversal in llamas.


Elmubark (2001). Fertility rate after oestrous synchronization and artificial insemination in desert sheep under extensive system in the sudan. M.V.Sc. Thesis, Faculty of Veterinary Medicine, U of K.


Ghurashi, M.A. (1999). Some aspects of short term thiopentone sodium anaesthesia with or without selected pre-anaesthetic medications in goat kids. Athesis of M.V.SC. Faculty of Veterinary Science U of K.


Greene, S.A; et al. (1986). Decreased antidiuretic hormone concentration is associated with xylazine-induced diuresis in mares. (Abstract) Veterinary Surgery, 15, 459.


Hartsfield, S.M. (1980). The effects of acetylpromazine, xylazine, and ketamine on urine production in cats. Presented at the American College of Veterinary Anaesthesiology Annual Scientific Meeting, St. Louis, M.O.


Kerr, D.D; et al. (1972a). Sedative and other effects of xylazine given intravenously of horses. American Journal of Veterinary Research 23;525-532.


Lee, I; et al. (2003). Antagonistic effects of intravenous or epidural atipamizole on xylazine induced dorsolumbar epidural analgesia in cattle. The Veterinary Journal. 166.164-197.A.


Tamara, L. Grubb; et al. (2002). Comparison of Lidocaine, xylazine and lidocaine-xylazine for caudal epidural analgesia in cattle. Veterinary Anaesthesia and Analgesia. 29:64.


WWW.ahc.umn.edu/rar/anesthesia.html.95k. Anesthesia, Analgesia and Sedation. Research Animal Resources, copyright 2009, University of Minnesota Board of Regents.
المستخلص:

- أجريت هذه الدراسة على 16 من النعاج الحوامل السليمة لبحث مدى ملاءمة وأمان بروتوكولات التخدير الآتية:

  1. بروتوكول 1 (زايلزين 2% + كيتامين 5%)، حقن في مرحلة الحمل الأولية.
  2. بروتوكول 1 (زايلزين 2% + كيتامين 5%)، حقن في مرحلة الحمل الأخيرة.
  3. بروتوكول 2 (ديازيبام 0.5% + ثيوبنتون صوديوم 2.5%)، حقن في مرحلة الحمل الأولية.
  4. بروتوكول 2 (ديازيبام 0.5% + ثيوبنتون صوديوم 2.5%)، حقن في مرحلة الحمل الأخيرة.

قسمت النعاج الحوامل عشوائياً إلى مجموعتين، ثمانية نعاجات في كل مجموعة.

- استخدم بروتوكول 1 (زايلزين+ كيتامين) في المجموعة الأولى من النعاج الحوامل مرحلة الحمل الأولية ومرحلة الحمل الأخيرة.
- استخدم بروتوكول 2 في المجموعة الثانية من النعاج الحوامل مرحلة الحمل الأولية ومرحلة الحمل الأخيرة.
- استخدم عقار الزايلزين بجرعة مقدارها 0.1 ملجم/كجم من وزن الحيوان وديازيبام بجرعة مقدارها 0.4 ملجم/كجم من وزن الحيوان كمهدئات وذلك بعد قياس الحوامل بعد 15 دقيقة من حقن العلاج التمهيدي.

بعد 15 دقيقة من حقن العلاج التمهيدي تم إحداث التخدير وريدياً بإستخدام إما عقار الكيتامين بجرعة مقدارها 22 ملجم/كجم من وزن الحيوان أو عقار الثيوبنتون صوديوم بجرعة مقدارها 7.5 ملجم/كجم من وزن الحيوان في حالة مرحلة الحمل الأوليّة وجرعة مقدارها 10 ملجم/كجم من وزن الحيوان في حالة مرحلة الحمل الأخيرة.

- أثناء فترة التخدير سجلت أهم القياسات الفسيولوجية ك معدل التنفس ومعدل ضربات القلب ودرجة حرارة الجسم بالإضافة إلى الأجهزة الأساسية للقياس. كذلك تم قياس بعض أطوار التخدير (طور التخدير، طور الاستلقاء الجانبي، طور الاستلقاء القصي، طور الوقوف).
وجد أن معدل ضربات القلب قد انخفض معنويًا في ربع الساعة التي تلت حقن عقار الزايلزين وعاد إلى الوضع الطبيعي مباشرة بعد حقن عقار الكيتامين وذلقت في مرحلة الحمل الأولى والأخرى. كما كان هناك ارتفاع معنوي في معدل ضربات القلب بعد حقن عقار الثيوبنتون صوديوم وذلك في النعاج الحوامل (مرحلة الحمل الأولى). ولا يوجد تأثير معنوي بعد حقن هذا العقار في النعاج الحوامل (مرحلة الحمل الأخيرة).

لم يكن هناك اختلاف معنوي في معدل التنفس في النعاج الحوامل (مرحلة الحمل الأولى) عند حقنها بالبروتوكول 1 أو البروتوكول 2 على الرغم من أن هناك زيادة في معدل التنفس في ربع الساعة التي تلت حقن عقار الزايلزين أو الديازيبام وعاد معدل التنفس إلى الوضع الطبيعي مباشرة بعد حقن عقار الكيتامين أو الثيوبنتون صوديوم وذلك في مرحلة الحمل الأولى والأخرى. ولاحظ انقطاع التنفس فقط عند استخدام البروتوكول 2.

لم تؤدي البروتوكولات التخديرية التي استعملت اختلاف معنوي في درجة حرارة الجسم في مرحلتي الحمل الأولى والأخيرة.

أدى البروتوكول التخدير 2 زيادة معنوية في طول طور الاستلقاء الجانبي والقصي وقصر في طور الوفوق مقارنة ببروتوكول التخدير 1 في مرحلة الحمل الأخيرة.

أثبتت كل بروتوكولات التخدير التي استخدمت في هذه الدراسة لإحداث تخدير وريدي عام سلامتها وملاءمتها في النعاج الحوامل ويستدل على ذلك بعدم حدوث حالات وفاة أو إجهاض.