



**BAHIR DAR UNIVERSITY**  
**COLLEGE OF AGRICULTURE AND ENVIRONMENTAL SCIENCES**  
**GRADUATE PROGRAM**  
**ESTRUS SYNCHRONIZATION AND ARTIFICIAL INSEMINATION**  
**TECHNOLOGIES IN ABERGELLE GOAT AT STATION AND ON-FARM**  
**CONDITIONS OF WAGHEMIRA ZONE, ETHIOPIA**

**MSc. Thesis**

**By**

**Bekahegn Wondim Alene**

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**June 2019**

**Bahir Dar, Ethiopia**



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**Submitted to the Graduate Program in the Partial Fulfillment of the**  
**Requirements for the Degree of Master in Animal Genetics and Breeding**

**June 2019**

**Bahir Dar, Ethiopia**

## DECLARATION

This was certified that the thesis entitled “**Estrus Synchronization and Artificial Insemination technologies in Abergelle Goat at Station and On-farm conditions of Wagemira zone, Ethiopia**” submitted in partial fulfillment of the requirement for the award of the degree of Master of science in “Animal Genetics and Breeding” to the graduate program of College of Agriculture and Environmental Science, Bahir Dar University by Mr. Bekahgn Wondim (ID: BDU1018742PR) is an authentic work carried out by him under our guidance. The matter embodied in this project work has not been submitted earlier for the award of any degree or diploma to the best of our knowledge and belief.

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## THESIS APPROVAL SHEET

As member of the Board of Examiners of the Master of Sciences (M.Sc.) thesis open defense examination, we have read and evaluated this thesis prepared by **Mr. Bekahegn Wondim Alene** entitled “**Estrus Synchronization and Artificial Insemination technologies in Abergelle Goat at Station and On-farm conditions of Wagemira zone, Ethiopia**”. We hereby certify that; the thesis is accepted for fulfilling the requirements for the award of the degree of Master of Sciences (M.Sc.) **in Animal Genetics and Breeding**.

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## LIST OF ACRONYMS / ABBREVIATIONS

<b>Abbreviations</b>	<b>Full content</b>
AI	Artificial Insemination
AFK	Age at first Kidding
ART	Assisted Reproductive technology
AV	Artificial Vagina
BCS	Body condition scores
CBBP	Community-based breeding program
CL	Corpus Luteum
CSA	Central Statistics Agency
eCG	Equine chronic gonadatropins
FAO	Food and Agriculture Organization
FAP	Fluorogestone acetate
FSH	Follicle Stimulating Hormone
GDP	Gross Domestic Product
GnRH	Gonadotrophine Releasing Horne
hCG	Human chronic gonadatropins
ICARDA	International center for agricultural research in dry areas
ILRI	International Livestock Research Institute
I U	International Unit
KI	Kidding Interval
LH	Luteinizing Hormone
LI	Lambing Interval
LIVES	Livestock and irrigation value chains for Ethiopian smallholders
LS	Litter size
MAP	Medroxyprogesterone acetate
MGA	Melengestrol Acetate
PGF2 $\alpha$	Prostaglandin F Two Alpha
PMSG	Pregnant Mare Serum Gonadotrophine
TR	Twining Rate

## ABSTRACT

### **Estrus Synchronization and Artificial Insemination technologies in Abergelle Goat at Station and On-farm conditions of Wagemira zone, Ethiopia**

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*Reproductive bio-technological tools are important to suit kidding time with better forage availability and accelerate improved genetics in the selective breeding programs. The study was conducted from 2018 - 2019 to investigate the effect of different estrus synchronization protocols on estrus response and conception rate of Abergelle goats with fixed time artificial insemination (AI). The protocols evaluated were: 1) Pregnant mare serum gonadatropins (PMSG+Enzaprost<sup>®</sup>), 2) single, and 3) double injection of prostaglandin (PGF2 $\alpha$ ). Semen characteristics of Abergelle bucks under station and CBBP management conditions were also investigated. The perception of farmers on the reproductive technologies was assessed using a semi-structured questionnaire. A total of 335 (station = 92 and CBBP site = 243) Abergelle does were used for the experiment. For the on-station experiment, 25 does were distributed for each of the treatment groups and 17does were allocated as a control group. For the on-farm experiment, 73 does for PMSG+Enzaprost<sup>®</sup>, 58 does for PGF2 $\alpha$  single injection, 72 does for PGF2 $\alpha$  double injection and 40 does for control groups were allocated. For the AI and semen analysis experiment, five bucks from the station and 18 bucks from the CBBP villages were used. At the station, does were allowed to graze for about 8hrs daily followed by supplementation of mixture of 300-350g cowpea hay and wheat bran per day for about one month before the commencement of the experiment. Similarly, bucks were kept by separate attendant and allowed for grazing for about 4 hrs dally followed by supplementation of the above-specified ration for about one and half month prior to the actual experiment. For the on-farm experiment, does used in the trail were separated from farmers flock and kept by separate attendants, allowed to graze as usual practice by farmers followed by supplementation of similar ration as the on- station per day for about one month before the actual experimental time. Bucks were kept also by separate*

attendant allowed to graze as usual practice of farmers followed by supplementation of the above-specified ration for about one and a half month prior to the actual experiment period. In both, station and CBBP villages, for the PMSG treatment groups, progesterone impregnated vaginal sponge was inserted at day 1 and stayed for 11 days inside, and 48 hrs before sponge removal, 1ml (600IU) PMSG hormone followed by 1ml Enzaprost<sup>®</sup> were administered as a separate injection. For single injection PGF2 $\alpha$  treatment groups, 1ml of Enzaprost<sup>®</sup> was administered at day 1. For the PGF2 $\alpha$  double injection groups, 1ml of Enzaprost<sup>®</sup> were administered at day 1 and the second 1ml injection of Enzaprost<sup>®</sup> at day 11. All the treatment groups were followed for estrus detection with four hrs interval for about 48 hrs, and AI was performed for those coming to heat. The control groups were left as normal practice and allowed to mate as usual. The study revealed that overall estrus response of 87.6%, 61.4% and 53% were investigated from PMSG+ Enzaprost<sup>®</sup>, PGF2 $\alpha$  (single injection) and PGF2 $\alpha$  (double injection), respectively. The average time for the onset of estrus and estrus duration were 14.4 $\pm$ 0.47 hrs and 45.5 $\pm$ 0.75 hrs, respectively. The study also revealed that mid parity does (3<sup>rd</sup> and 4<sup>th</sup>) and does with body condition score of 2.5 and 3 were found to be significantly responsive for estrus ( $p < 0.05$ ). Relatively higher conception rate ( $p < 0.05$ ) was resulted from PGF2 $\alpha$  double injection (78.8%) followed by PGF2 $\alpha$  single injection (64.7%). A significantly higher ( $p < 0.05$ ) proportion of litter size (1.6 $\pm$ 0.22) was resulted from PMSG+Enzaprost<sup>®</sup>. Most of the semen characteristics in the study does not show any significant difference at ( $p > 0.05$ ) against management condition and ejaculate frequency. It was observed that PMSG+ Enzaprost<sup>®</sup> was best responsive ( $p < 0.001$ ) for induction of estrus, and double injection of PGF2 $\alpha$  had a shorter ( $p < 0.001$ ) time for the onset of estrus with highest conception rate ( $p < 0.05$ ). It was concluded that according to the type of hormonal treatment, time of AI should be set by considering induction and duration of estrus to achieve a better conception rate.

**Keywords:** Abergelle goat, AI, Conception rate, Estrus response, Synchronization, PGF2, PMSG

## **Chapter 1: INTRODUCTION**

### **1.1 Background and Justification**

The goat, which was the first animal to be domesticated by humankind, is mainly raised in Asian (~500 million heads), African (~290 million heads), Europe (~21 million heads) and North America (~3 million heads) (Amills, 2014) continents. Goats, known as the “poor man's cow”, are the most prolific of all domesticated ruminants under tropical and subtropical conditions which are able to breed year-round and have an important livelihood contribution for the resource-poor farmers (Mamabolo and Webb, 2005).

The goat population of Ethiopia is estimated to be more than 29 million (CSA, 2015) which has increased by 30% in the last 12 years (Solomon Abegaz *et al.*, 2014). Goats are raised at a large proportion (58%) found in the lowlands of the country raised in large flocks by pastoralists while, 42% of the total are found in the highlands where there is a strong complementary relationship between small ruminant keeping and cropping (ESGPIP, 2008; Markos Tibbo, 2006). Goats comprise 5.32% of the total tropical livestock units of Ethiopia; contribute an estimated 12 to 14% of annual meat products, 10.5% of milk production, 47% of the agricultural GDP (Behnke, 2010), 30% of all domestic meat consumption (Zelalem Alemayehu & Fletcher, 1991; Belay Derbe and Mengistie Tay, 2013) and 6% of all animals exported (Solomon *et al.*, 2014). Together with sheep, goats contribute about 90% of the live animal/meat and 92% of the total skin export trade value (FAO, 2004). However, in the country goat production and productivity is characterized by minimum profit resulting from under exploitation of indigenous genetic resource, poor or seasonal fluctuations of feed resources, periodic droughts, extensive dry seasons and severe feed shortages resulting in undernourishment and low productivity. The average carcass weight produced from a yearling goat in Ethiopia is only 8.5 kg (FAO 2004).

Abergelle goat breed which is under rift valley roud family has an estimated population of over 300,000, found along the Tekeze River in Southern Tigray (Tembien and Inderta), Waghimra, Raya Azebo, and North Gondar (Simien) and kept by the Agew and Tigray ethnic groups (FARM AFRICA, 1996). Goat together with other livestock species

contributes about 70% of the income sources of smallholder farmers at the mid and lowland areas of Waghemira (Bekahgn Wondim *et al.*, 2017). According to the result from established Community Based Breeding Programs (CBBP) works on the breed, the average flock size was 27 heads per household (Alubel Alemu, 2015). The breed is characterized by its lower body weight, lower production potential, longer kidding interval, lower kidding rate and lower litter size with better meat quality and temperament in comparison to other indigenous goat breeds (Alubel Alemu, 2015). As a result of this, holistic and an attractive approach of genetic improvement program has been designed, implemented and promising results have been achieved, that is community-based breeding program (CBBP).

Reproduction is critical for the attainment of profitability in any livestock enterprise, including goat rearing. Various assisted reproductive technologies have been applied to accelerate the genetic gain and enhance the reproductive performance of various livestock species. In goat rearing, estrus synchronization followed by Artificial Insemination (AI) is currently the most practical technology for optimizing reproductive efficiency (Tsuma *et al.*, 2015). AI accelerates the rate of genetic gain within a herd/flock, maximizes the number of offspring from a desirable sire, enables genetic exchange over wide geographical areas, and also allows the use of genetic material from incapacitated sires or those no longer alive if their semen had been preserved. Estrus synchronization combined with artificial insemination (AI) is used regularly in cattle and has been useful for genetic improvement and breeding management. These technologies would also be useful for goat keepers interested in using AI to increase the genetic merit of offspring and accelerating improved genetics (William Knox, 2013). Thus the application of reproductive biotechnology tools will be vital to increase kidding rate, better dissemination of improved genetics and to best suit the overall breeding management activities.

## **1.2 Statement of the problem**

For accelerating the genetic gain in the selective breeding programs, requires higher intensity of selection. In the contrary, because of the presence of larger flock size and very short mating season in the study area, there is a need for keeping higher number of serving sires for addressing the whole females so that intensity of selection must be relaxed. Thus, use of reproductive biotechnology tools together with the selection program will enable for faster genetic progress. Feed resource availability and overall management

harassments are other important concerns. The area is characterized as dry area so that feed resource is very deficient up to 6 months of from January up to June (Bekahgn Wondim *et al.*, 2018) so that adjusting appropriate birth season should be come other important step for improving survival and birth weight of kids (Appendix fig 6). Therefore this MSc thesis was initiated to evaluate the efficiency of estrus synchronization and Artificial insemination technologies in Abergelle goat at station and CBBP village conditions.

### **1.3 Objectives**

#### 1.3.1 General objective

The general objective of this research was to evaluate the performances of Estrus synchronization and Artificial insemination technologies for the case of Abergelle Goat breed

#### 1.3.2 Specific Objectives

- Evaluate estrus response and conception rate of Abergelle Does for different estrus synchronization protocols
- Evaluate the efficiency of Artificial Insemination (AI) following Estrus Synchronization in Abergelle Does at Station and On-farm conditions
- Evaluate Semen quality of Selected Abergelle Bucks from the Station and On-farm conditions
- Compile Keeper's Perception on Estrus Synchronization and Artificial Insemination tools at the model CBBP villages

## Chapter 2: LITERATURE REVIEW

### 2.1 Reproductive characteristics and levels of performances for major reproductive traits of Ethiopian Goats

Reproductive trait takes a paramount share for leading a successful animal production program as reproductive failure is the first sign of decreased productivity in livestock enterprise (Mukasa Mugerwa *et al.*, 2002). In the conventional breeding program, sometimes reproductive traits are difficult to measure and are strongly influenced by management decisions for the reason that it can affect selection intensity in terms of conception rate, litter size, young mortality and interval between parturitions and consequently the rate of genetic improvement in the remaining traits under selection (Notter, 2000). In most of the Ethiopian farming areas, the traditional free-ranging management system of goat production allows for year round breeding, which creates a good opportunity for bucks to service does any time, which is different from controlled system under on station and intensive systems of production. However, uncontrolled breeding is complicated by diseases transmission and inbreeding when the bucks are small in number as the poor reproductive performances of Ethiopian sheep and goats can be associated with genetic factors, poor management, seasonal fluctuations in feed resources and diseases (Mukasa-Mugerwa *et al.*, 2002).

#### 2.1.1 Age at first kidding (AFK)

There is a big variation among production system and breeds for age at first kidding ranging from (12-24 months) which could be due to genetic and environmental differences for Ethiopian and tropical goats at large (Getahun Legese, 2008). The mating scheme, number and availability of breeding males in the flock determine the speed for first kidding and genetic improvement program in the flock (Wilson and Durkin 1988). Hence, the average age for first kidding for most of indigenous African goats is 17 months (Wilson, 1989). The potential of Keffa and Adilo goats to have their first kids at the age of 12.5 months have been documented in a study conducted under traditional system in southern part of Ethiopia (Getahun Legese, 2008). A research by (Assen and Aklilu 2012) reported that 15 months for AFK for local goats found in central Tigray and Dale district. AFK of about 20.1 months have been reported for local goats in pastoral and agro-pastoral areas of Southern Ethiopia (Adugna Tolera and Aster Abebe, 2007). For station managed

Arsi-Bale goats, 28 months for AFK reported was relatively higher (Dadi *et al.*, 2008) compared with other indigenous goats in Ethiopia. In general, most of the reports explored so far with this review show that the earliest AFK was recorded in the traditional production systems which indicating that the existing uncontrolled breeding practice in this system is in favor of early kidding of indigenous goats than the controlled breeding practices in the improved system for the reason that free access of breeding bucks year round together with the flock is tending the does to conceive very early. Data on average age at first kidding (AFK) for some indigenous goats in Ethiopia are summarized in table 2.1.

#### 2.1.2 Kidding interval (KI)

It refers to the duration between two consecutive kidding or it is the number of days between successive parturitions. KI is an important reproductive trait which measures the reproductive efficiency of the animals. A study conducted by (Getahun Legese,2008) and (Girma Abebe,2008) has revealed that at under normal breeding circumstance there is a possibility of indigenous Ethiopian goats to have three kidding in two years time which might influenced by various factors including previous litter type, parity and lambing season (Dibissa, 2000). The KI for most Ethiopian indigenous goats are between the estimates for Small East African goats. This has been confirmed by (Tatek *et al.*, 2004), (Adugna Tolera and Aster Abebe, 2007) and (Deribe Gemiyu, 2009) who reported 8 months of KI for some indigenous goat breeds in Ethiopia. Average kidding interval of about 11 and 14 months for Abergelle and Begait goats respectively were reported by (Berhane and Eik 2006). (KI) of  $5.47 \pm 0.96$  and  $6.6 \pm 1.37$  was reported for Woyto Guji and Central Highland goat breeds respectively (Netsanet Zergaw *et al.*, 2016). In the traditional production system, relatively higher intervals (9-12 months) have been reported by (Markos Tibo, 2000) where as 12 months under traditional management systems were reported by (Samuel, 2005; Dereje Tadesse *et al.*, 2016). KI of about 10 months was reported for Arsi-Bale goats kept at on-station management condition (Dadi *et al.*, 2008). Data on average age at first kidding (AFK) for some indigenous goats in Ethiopia are summarized in Table 2.1.

Table 2. 1 Age at first kidding and kidding interval (month) of indigenous goats of Ethiopia

Goat types	KI	AFK	Sources
Arsi-Bale	8.1	12.1	Tatek Woldu <i>et al.</i> , 2004
Adilo	-	12.9	Getahun Legese, 2008
Goats in Metema	8.4	13.6	Tesfaye, 2009
Goat in Alaba	9.1	11.9	Deribe, 2009
Arsi-Bale	10.5	28.5	Dadi <i>et al.</i> , 2008
Central highland	11.5	-	Samuel, 2005
Abergelle and Bagait	12	-	Berhane and Eik, 2006

*AFK = age at first kidding; KI = kidding interval*

### 2.1.3 Litter size (prolificacy)

Prolificacy is economically important reproductive term resulted from a combination of ovulation rate and embryo survival and its efficiency is measured in the number of lambs or kids born per parturition. Litter size for most indigenous goats of Ethiopia are reported to be between 1.07 and 1.5 and varied up on breed, management conditions and physiological stage/age of does as the peak prolificacy generally achieved between 4 and 8 years of age (Notter *et al.*, 2000). However, in few cases, twinning rate of 41 to 50% has been reported for local goats in Awassa Zuria (Markos Tibo, 2000) and in western lowlands of Ethiopia (FARM-Africa 1996). With same study above, 1.21 litter size have been recorded in the central Ethiopian highlands which accounted about 17% and correspondingly the mean annual kidding rate of central highland goats obtained was  $1.46 \pm 0.06$  (Mengistie Taye and Belay Derbe, 2013), 1.47 for local goats in Alaba, Southern Ethiopia (Deribe Gemiyu, 2009). Previous studies generally show that litter size and twinning rate are the most variable traits reported for indigenous goats in Ethiopia indicating the presence of huge opportunity to improve these traits through selection and improved management focusing on breeds having better potential for the traits. It is also

found that litter size and twinning rate of Somali and Afar goats are smaller than the values reported for most of the indigenous goats in Ethiopia even under traditional systems.

## **2.2 Reproductive Performance of Abergelle goat**

Reproductive characteristics are important characteristics of the breed which directly affect its productivity (Getahun Legesse, 2008). The level of reproductive performance is dependent on the interaction of genetic and environmental factors (Duygu, 2010). Age at first kidding, kidding interval and litter size are the most important characteristics which determines the productivity of the flock. Age at first kidding (14.9) months and kidding interval of (8.28) months were reported for Abergelle goat (Belay Derbe, 2008). Litter size of 1.06 was also reported by (Shumuye Belay *et al.*, 2014) for the same breed.

## **2.3 Estrus cycle and physiological actions in Goats**

Once puberty is attained, the doe comes on heat (estrus) at regular intervals thereafter. Estrus is associated with the desire to be bred. This behavior (estrus) is repeated at regular intervals, unless interrupted by pregnancy or disease, and this repetition is referred to as the estrus cycle. Therefore, the estrus cycle is defined as the number of days between two consecutive periods of estrus (Tsuma *et al.*, 2015). The estrous cycle is a series of hormonal cascades that change the morphology of the female reproductive system to prepare for pregnancy (Audra, 2014). According to the author, Goats exhibit a 21 days estrous cycle with estrus duration of 24-48 h, and ovulation occurring, on average, 24 h after the onset of estrus (Rahman *et al.*, 2008). Transitional periods from anestrous to active cyclicity generally occur during the mid-summer months as days begin to shorten (Abecia *et al.*, 2011). According to (Attwood, 2007), like ewes, does are seasonally polyestrous as their reproductive cycle responds to changes in day length. Sexual activity is usually greatest during autumn and winter. Estrus activity of doe is greater in the tropics than in temperate climates. The average length of estrous cycle in the doe is 21 days, but can vary from 18-22 days depending on the breed differences, stage of breeding season and environmental stress (Jainudeen *et al.*, 2000).

The abnormally short cycles that are observed in the doe early in the breeding season may be associated with premature regression of the corpus luteum. The estrus lasts for 24-48 h in the doe and the duration of estrus can be influenced by breed, age, season and presence

of the buck (Jainudeen et al., 2000). Estrus has shorter duration at the end of breeding season and in the first breeding season of young does. The complete estrous cycle in doe is divided into 4 well marked phases, namely proestrus, estrus, metestrus and diestrus (Rahman, 2008). In pre pubertal and aged does anoestrus (stage of sexual quiescence characterized by lack of estrus behavior) normally occurs. This may be due to the complete suppression of ovarian activity or silent ovulatory cycles without behavioral signs of estrus. In all domestic animals including the doe, anoestrus also may occur as a pathological condition (Pineda, 2003). The duration of each event associated with the estrus cycle and pregnancy in the doe is illustrated in Fig 2.1.

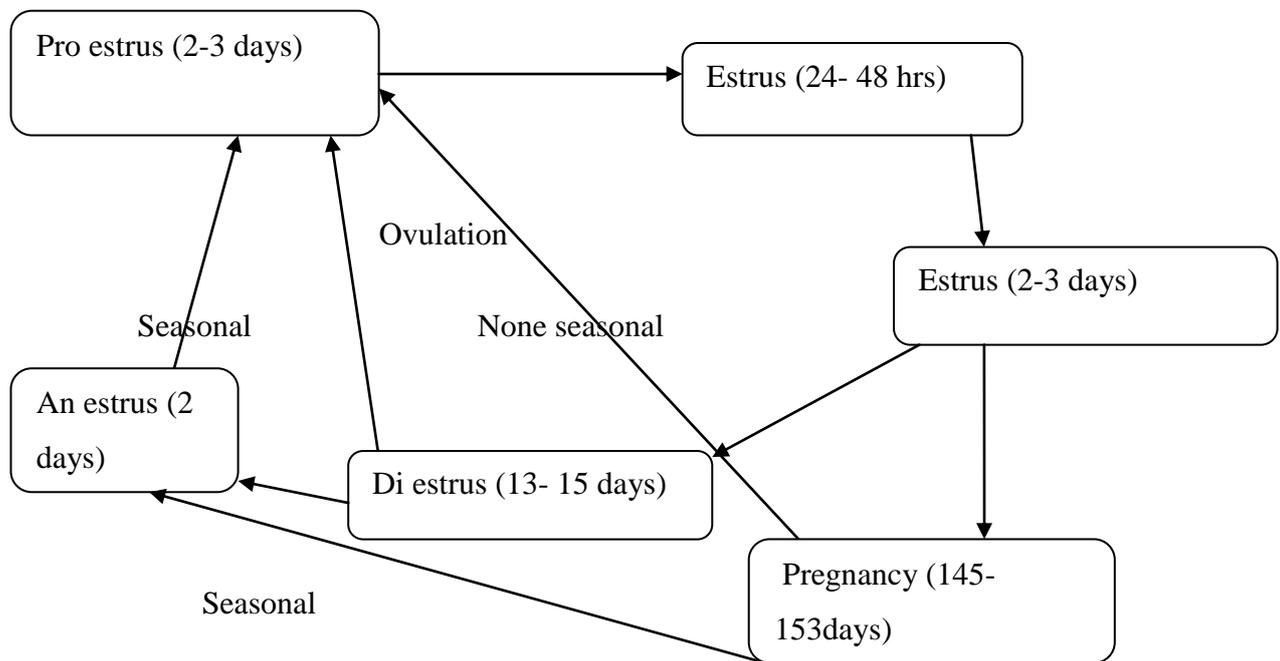


Figure 2.1 Estrus events and estrus cycle in goats (Rahman *et al.*, 2008)

Healthy doe will come into estrus every 20 to 21 days, except when pregnant (Fig 2.2). To determine the estrus cycle length of each breeding doe, the producer should maintain breeding records. Good records are important for informing a breeding program, as they can help the breeder predict when a doe is likely to come on heat, and thus enable more careful observation for heat signs at that time. Additionally, good records will help to determine if a doe has a problem, if she comes on heat too early, delays, or fails to come on heat at the expected time and she has not been previously bred.

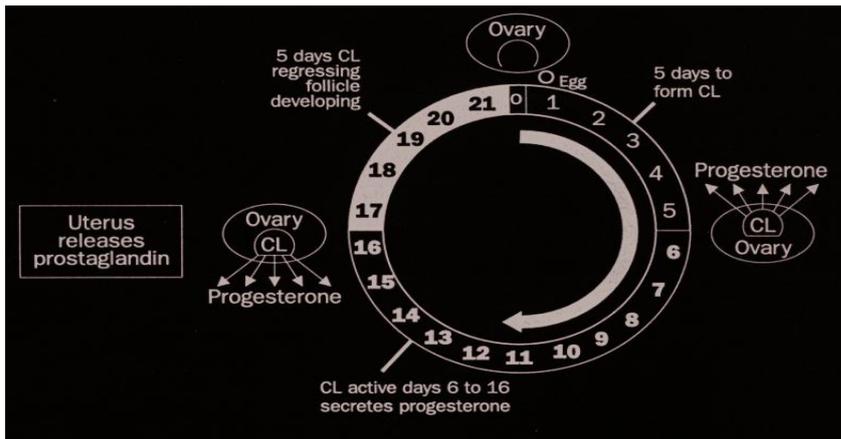


Figure 2.2 Schematic representation of estrus cycle in does (Tsuma *et al.*, 2015)

## 2.4 Hormonal changes and follicular waves in does

According to (Ginther, 1994), the growth of ovarian follicles in the ovary of doe is characterized by the presence of 4 or more waves of follicle growth in the same cycle and it is in the final wave where the dominant follicle ovulates. The author also revealed that, the behavior of strong follicular dominance is less apparent in the ovary of the doe. Each follicular wave is preceded by an increase in FSH secretion (Medan *et al.*, 2005). Unlike other farm animals, the subsequent follicular wave begins even though the dominant follicle of the previous wave is still in its peak of development (Rahman, 2008). (Menchaca and Rubianes, 2002) investigated that progesterone treatment affects follicular dynamics in dairy does. Ovulation in the doe is spontaneous and most goat breed ovulates between 24-36 h after onset of estrus, the Nubian goat ovulates later, which is possibly due to a longer estrous cycle in this breed (Jainudeen *et al.*, 2000). According to (Pineda, 2003), the average ovulation rate in the doe is 1-3 oocytes but can vary from 1-5 depending upon the breeds and management conditions

## 2.5. Hormonal treatments for controlling the estrous cycle in goats

The reproductive physiology of goats can be controlled by several methods developed in recent decades. Some of these involve administration of hormones that modify the physiological chain of events involved in the sexual cycle. Progesterone or its synthetic analogues are becoming important hormones affecting the luteal phase of the cycle, simulating the action of natural progesterone produced by the corpus luteum after ovulation, which is responsible for controlling LH secretion from the pituitary (Abecia *et*

*al.*, 2012). Use of prostaglandins is an alternative method for controlling reproduction by regressing the corpus luteum and inducing a subsequent follicular phase with ovulation. Naturally; hypothalamus, pituitary, ovary, and uterus are known glands, tissues or organs which control the estrous cycle in does. Each of these components of the reproductive system secretes chemical compounds called hormones, which regulate their own function or the function of other components. Many hormones are involved in control of the estrous cycle, and their release into the bloodstream can be measured experimentally (Michael and Thomas, 2005; Soren *et al.*, 2012; Alemat Gidena, 2017). Thus, use of hormones to induce estrus has allowed mass artificial insemination in goats in particular and small ruminants in general. At commercial level, synchronization of estrus allows for the control of lambing and kidding, with subsequent production of same batch young animals for slaughter. More efficient use of labor, animal facilities, multiple ovulation and embryo transfer programmes' are also other possible package advantages of estrus synchronization and artificial insemination. The major hormones, which are most commonly administered to animals to synchronize estrus, are shown in Table 2.3.

Table 2. 2 Sources and Functions of the Major Reproductive Hormones and there commercial products

Hormones	Source	Function	Product name/commercial
GnRH	Hypothalamus	Releases FSH and LH	Cystorelin®, Factrel®
FSH	Anterior pituitary	Stimulation	Folltropin®
Estrogen	Ovarian follicle	Stimulation	Not used in current system
LH	Anterior pituitary	Stimulation	Not used in current system
Progesterone	Corpus luteum	maintain pregnancy	MGA®, intravaginal
Prostaglandin	Uterus	Regress Corpusluteum	Lutalyse ®, Estrumate®

(Adapted from Alemat Gidena, 2017)

#### 2.4.1 Prostaglandins (PGF2 $\alpha$ ) and its synthetic analogues

Prostaglandins are natural chemicals in the body with hormone-like qualities which has important implications in reproductive physiology of animals which is naturally occurring during the normal estrous cycle of a non-pregnant animal. PGF2 $\alpha$  is released from the uterus 16 to 18 days after the animal was in heat. This release of PGF2 $\alpha$  functions to destroy the corpus luteum. The release of PGF2 $\alpha$  from the uterus is the activating

mechanism that results in the animal returning to estrus every 21 days. In small ruminants, prostaglandin F2 $\alpha$  is the primary luteolytic agent (McCracken et al., 1970). Prostaglandins are mainly administered intramuscularly and subcutaneously, although the intravulvo-submucosa route has been investigated with varying success (Omontese *et al.*,2013). Prostaglandins have the major advantage of being administered by intramuscular injection besides the reduction in hormonal residues, since it is rapidly and almost completely metabolized in the lungs (Gonzalez and Veiga, 2005). Several synthetic analogues have been used to induce rapid regression of the corpus luteum. The mechanism of actions of (PGF2 $\alpha$ ) in the does reproductive tract is in such a way that; an injection of synthetic PGF2 $\alpha$  will imitate the natural PGF release to cause CL regression and then Synchronized regression of the CL will synchronize a decline in progesterone and result in the final growth of the dominant follicle to produce estradiol and behavioral heat (Michael and Thomas, 2005). Although natural PGF2 $\alpha$  like Lutalyse<sup>®</sup> and Carboprost<sup>®</sup>, causes normal luteolysis through gradual degenerative changes, synthetic analogues of PGF2 $\alpha$  like cloprostenol sodium, marketed as Fenprostamol<sup>®</sup> , Estrumate<sup>®</sup> and estroPlan<sup>®</sup> and Enzaprost<sup>®</sup> are usually have a more rapid and dramatic effect on progesterone synthesis in the lutein cells (Colak *et al.*,2008) and (Ansari *et al.*,2010). Dinoprost tromethamine treatment synchronised estrus by decreasing average time to first estrus by approximately 2 weeks (Dudhatra *et al.*,2012).

Physiological state of dose, level of the prostaglandin, the interval between administration of the prostaglandin, the responsiveness of the corpus luteum to the prostaglandin/stage of the estrus cycle (Lassal *et al.*, 2004), season and the inclusion of gonadatropins as co-treatment (Omontese *et al.*, 2013) are the factors reported to affect estrus response and subsequent fertility following administration. For normal cyclic females, estrus occurs within 2 to 6 days after they are given intramuscular injections of prostaglandin F2 $\alpha$  (Lutalyse<sup>®</sup>) or one of its analogues (ProstaMate<sup>®</sup>, Estrumate<sup>®</sup>, estroPLAN<sup>®</sup>, In-Synch<sup>®</sup>) (Islam, 2011). Prostaglandins should be administered from day 3 of the estrus cycle when the corpus luteum of the goat is responsive to PGF2 $\alpha$  (Rubianes and Menchaca, 2003). It is worthy of note that administration of PGF2 $\alpha$  will cause abortion at any trimester of pregnancy in goats (Aksu, 2003) Prostaglandins may be used throughout the entire year in tropical breeds, although (Kawu, 2000) reported that the highest estrus response of Savannah Brown does to prostaglandin treatment occur during the cold-dry season. As investigated by (Wheaton et al., 1993; Omontese *et al.*, 2016) the

dose of prostaglandin is less significant compared to the administration protocol where as double injection consistently resulted in higher estrus response rates than single injection. Thus, in order for PGF<sub>2</sub>α to be effective, females must be cyclic and in a responsive phase of the estrous cycle.

#### 2.4.2 Progestagen

Progestagen is a class of steroid hormones that bind and activate the progesterone receptor (PR) (Michelle *et al.*, 2011). According to (Whitley and Jackson, 2004), various forms of Progestogen and different methods of administration have been used in cycling does, as well as in seasonally an estrus does, to induce or synchronize estrus. Progestogen administration is common, especially in seasonally estrus animals and has been used with or without accompanying treatments such as gonadatropins or prostaglandin analogs. Progesterone is the major and most important Progestagen in the body for maintaining pregnancy and blocks” estrus and ovulation during the diestrus phase of the estrous cycle. Most importantly, providing a progestin will induce some pre-pubertal heifers and anestrous cows to begin cycling and to have a normal-length estrous cycle following the first ovulation (Michelle *et al.*, 2011). Apart from their role to synchronize estrus during the breeding season (Gordon, 1997) progesterone and Progestagen are widely used to induce estrus during the non-breeding season (Evans, 2004). The characteristics of the ovulatory follicle are also dependent on the ability of the Progestagen treatments to control gonadatropins secretion as it has been investigated in sheep (Flynn *et al.*, 2000). An inverse relationship between LH pulse frequency and progesterone concentrations is well documented in sheep (Evans, 2004).

#### 2.4.3 Gonadatropins -Releasing Hormone (GnRH)

Gonadotropin-releasing hormone (GnRH) is the primary hypothalamic signal peptide that governs reproductive function which regulates the synthesis, glycosylation, and secretion of LH and FSH, which in turn stimulate gonadal steroidogenesis and gametogenesis (Yohei *et al.*, 2003). Estrus cycle is controlled by GnRH (Gonadotrophic Releasing Hormone) released by the hypothalamus. Just before the onset of estrus, the pituitary gland, under the control of the hypothalamus in the brain, releases an increasing amount of Luteinizing Hormone (LH) and Follicular Stimulating Hormone (FSH). Growth of follicles is regulated by pituitary hormones (FSH, LH. LH) stimulates the final maturation

of the follicle containing the eggs (oocytes) and stimulates the follicle to produce the hormone estrogen. Estrogen brings the doe into behavioral estrus or 'heat.' The rising concentration of estrogen stimulates a surge in LH that stops further secretion of estrogen by the follicle. Once the egg has been released, LH transforms the follicle into a CL. After ovulation, under the influence of the gonadotropin hormone LH the cells inside the ruptured follicle begin to grow and undergo changes (luteinization) to form the CL. The progesterone is called the hormone of pregnancy and produced from CL. Its task is to prepare the uterus for accepting the fertilized egg, and to maintain pregnancy by preventing recurrence of the estrus cycle during gestation. The CL is destroyed if fertilization does not occur. Prostaglandin F<sub>2α</sub> (PGF<sub>2α</sub>) hormone produced by the inner wall of the uterus destroys CL. The reduction in progesterone concentration is consequently followed by a rise in gonadotropin release (FSH and LH), development of another follicle(s) and egg(s), and recurrence of heat. This cycle will continue throughout the reproductive life of the ewes and will only be interrupted by pregnancy, disease, under or overfeeding (Tsuma *et al.*, 2015). The traditional view was that eutherian mammals express only a single form of GnRH, known as mammalian GnRH (GnRH I: pGlu-His-Trp-Ser-Tyr-Gly-Leu-Arg-Pro-Gly-NH<sub>2</sub>). Most occurring trade names Cystorelin®, Fentanyl®, Factrel® and Ova Cyst®, are a naturally occurring hormone that causes the release of other hormones (FSH, LH). One of these hormones affects follicle development on the ovary; and the other causes ovulation.

Research indicates that when GnRH is given with prostaglandin to estrous cyclic and non-cyclic females, the patterns of follicular development are altered, inducing ovulation. This treatment may induce estrus in 10 to 30 percent of anestrus females.

## **2.6 Estrus synchronization**

Estrus synchronization is the manipulation of the estrous cycle of does so that females can be bred with normal fertility during a short, pre defined interval. The value of estrus synchronization is vital in does as the duration of both estrous cycle and estrus is variable and estrus detection cannot be accomplished safely without a buck (Jainudeen *et al.*, 2000). Approaches towards synchronizing estrus in livestock have to focus on either the manipulation of the luteal or the follicular phase of the estrous cycle. In the doe, the window of opportunity is generally greater during the luteal phase, which is of longer duration and more responsive to manipulation. Different approaches have been concerned

with either extending the luteal phase by supplying exogenous progesterone or with shortening this phase through regression of the corpus luteum. Successful techniques must not only establish synchrony, but also provide a reasonable level of fertility in the synchronized cycle. Estrus synchronization, especially under intensive production systems, can utilize protocols that enable AI to be done at a specific predetermined time following a synchronization treatment irrespective of whether the ewes are seen on heat or not (Tsuma *et al.*, 2015). Estrus synchronization allow for clustered lambing/kidding thus enable more efficient flock management and market access. Many young lambs born around the same time can be more intensively taken care of for limited periods, as opposed to all year round. lambing can be synchronized to occur at specific times to coincide with market needs for example when there is feed availability for the dam and young for milk production and growth, or market demand for sheep. Different methods are available to synchronize estrus in ewes (Tsuma *et al.*, 2015).

### **2.6.1 Hormonal Protocols for Estrus Synchronization in Goats**

Most commonly, two classes of hormones are available and widely used protocol for estrous synchronization in small ruminant which are progesterone and prostaglandins based protocols with based on long treatment, 12 to 14 day for sheep and 11 to 17 day for goats, with progestogen/progesterone (P4) (Alexandre and Bianca, 2018).The P4 is usually given as vaginal pessaries containing synthetic analogs (medroxyprogesterone acetate - MAP or fluorogestone acetate - FAP) or an intravaginal device impregnated with natural progesterone. Subcutaneous implants or intramuscular injections may also be used to simulate the action of endogenous progesterone produced by the corpus luteum after ovulation (Uribe, 2002).

Gonadotrophins are used together with P4, at the end of the protocol, in order to aid estrus synchronization and ovulation. The most commonly used gonadotropins are the Equine Chorionic Gonadotropin (eCG), which is essential for the induction of ovarian activity in small ruminants in anestrus, and the Follicle-Stimulating Hormone (FSH) (Moreira *et al.*, 2014; Alexandre and Bianca, 2018). The ECG doses range from 250 to 1000 IU, while the FSH doses range from 10 to 20 mg, depending on age, reproductive season, body condition, species, and breed. After treatment with P4 and gonadotropin, the animals tend to manifest estrus in approximately 48 hours and ovulation occurs in approximately 60 hours (Siliva *et al.*, 2010).

### 2.6.1.1 Prostaglandin (PGF<sub>2</sub>α)

Prostaglandins are a group of physiologically active lipid compounds constitute a group of 20 carbon unsaturated fatty acids which are naturally occurring compounds that are produced by most cells in the body and have a variety of biological actions. Prostaglandin released from the uterus, plays an important role in regulating reproductive cycles in domestic species through the control of luteal activity in non pregnant animals and the initiation of delivery in pregnant animals (Ricciotti *et al.*, 2011). PGF<sub>2</sub>α is a naturally occurring luteolytic hormone that has also been utilized to induce estrus and abortion in ruminants through CL regression. In the absence of an embryo, uterine concentration of PGF<sub>2</sub>α increase during the late luteal phase (Funston *et al.*, 2005). PGF<sub>2</sub>α is transported to the CL via a counter-current mechanism. Luteal cells are known to have PGF<sub>2</sub>α receptors on the plasma membrane and direct inhibitory effects of PGF<sub>2</sub>α on luteal progesterone secretion and PGF<sub>2</sub>α is known to reduce luteal blood flow due to vasoconstrictor activity (Niswender *et al.*, 2000). When pregnancy does not occur, the CL must regress to allow follicular growth and ovulation and the reproductive cycle begins again. For the purposes of estrous synchronization, injection of PGF<sub>2</sub>α is only effective in cycling mammals (Funston *et al.*, 2005) and is only applicable when there is a responsive CL on the ovary. Lutalyse™ (PGF<sub>2</sub>a) and Estrumate® (cloprostenol) are the two commonly used prostaglandins. Prostaglandin F<sub>2</sub>α, by its luteolytic activity, can synchronize estrus, but only when an active CL exists at the time of application. Prostaglandin causes regression of the CL, resulting in removal of the negative block progesterone has on FSH and LH release. Prostaglandin can be administered to the females in one-shot and two-shot options. One-shot option is applied in the way that single injection of prostaglandin is given to cyclic females, and then these females are bred as they express estrus with disadvantage of one-third of the females will not respond to the injection, but the advantages are the lower cost of one injection and that females are only handled once other than for breeding (Islam, 2011). Another one shot option requires detection of estrus before any prostaglandin treatment is administered (Fig2.3). The producer detects estrus for 5 days and breeds each cow as she exhibits estrus. The cows that have not exhibited estrus by the fifth day are given an injection of prostaglandin, which should induce them to come into estrus in about 3 to 5 days (Michael and Thomas, 2005; Alemat Gidena, 2017).

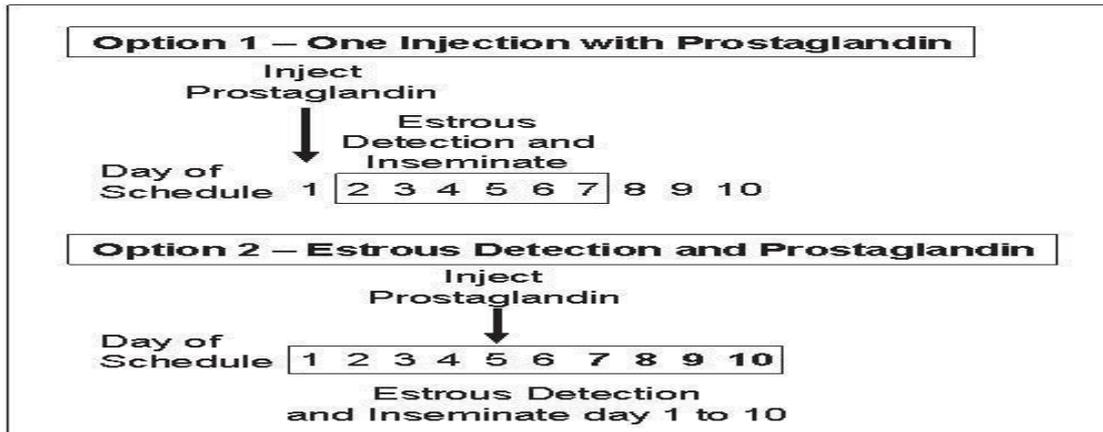


Figure 2.3 One shot prostaglandin (Alemat Gidena, 2017)

The two – shot program (Fig 2.4) for synchronization with prostaglandin are designed to increase the proportion of females with responsive CL for the deterioration with  $PGF_{2\alpha}$ . The program has two options like the one-shot program; the first option is the two injections of prostaglandin 14 days apart. The non-cycling animals will not generally respond to prostaglandin products. The advantage of this option is that more animals should come into estrus at any given time than with the one-shot options with a disadvantage of it involves the cost and labor of administering two injections of prostaglandin to all animals (Michael and Thomas 2005) and (Pacala, *et al.*, 2009). The second two-shot prostaglandin injection option is applied with double shot double insemination procedure; Give first injection and breed all females exhibiting estrus and then give the second injection to only females that were not breed. This option lowers expense and handling, but results in two synchronized groups instead of one and a longer breeding period. Timed insemination instead of estrous detection may be used, but conception rates are generally lower than with estrous detection (Michael and Thomas, 2005)

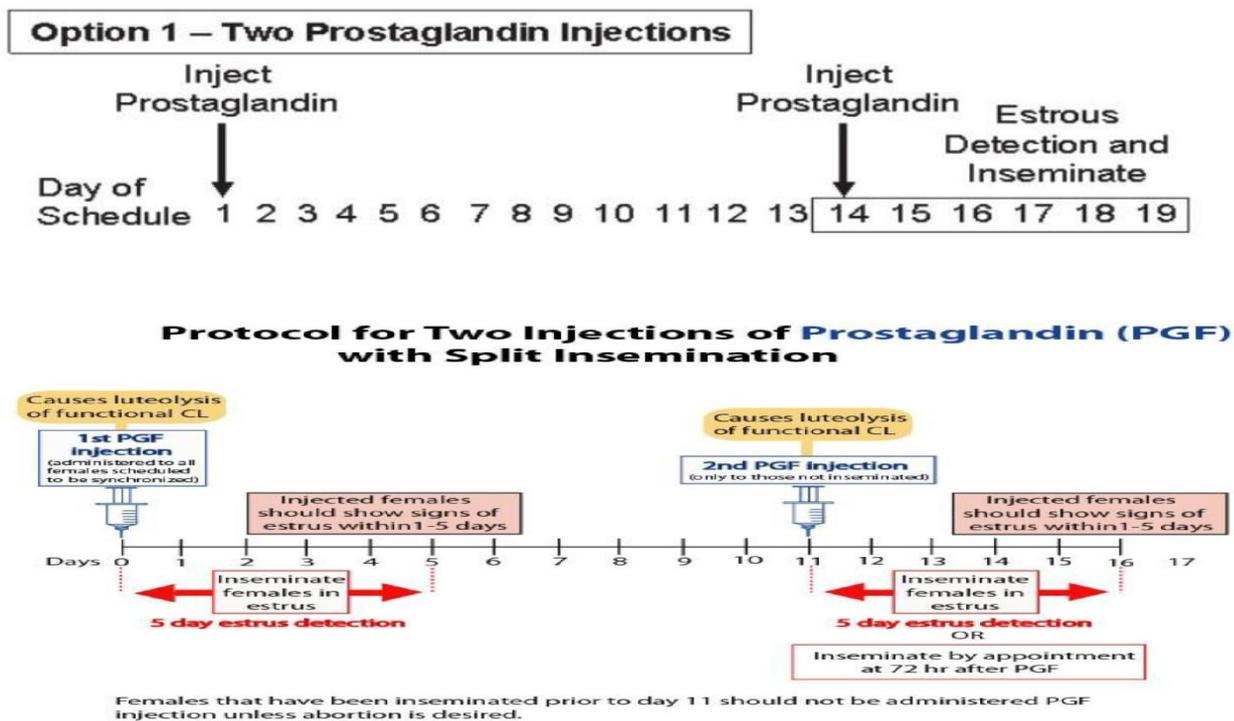


Figure 2.4 Two Shot Prostaglandin (Alemat, 2017)

### 2.6.1.2 Progesterone

Progesterone or its synthetic analogue, equine chorionic gonadotrophin (eCG) are generally used in the reproductive season of goats (Dogan *et al.*, 2005), outside the natural breeding season (Souza *et al.*, 2011; Dogan *et al.*, 2016) and in the transition period (Dogan *et al.*, 2004). Progestogen (synthetic analogs of progesterone) can be provided by feeding (MGA), implants under the skin (Synchro-Mate B®), sponges (or pessaries) or plastic delivery devices inserted into the vagina /CIDR (Schoenian, 2012). Intra-vaginal sponges have been used as the traditional method for synchronizing estrus in animals. Two types of sponges are Chronogest (FGA) and Veramix® (MAP) widely used with pregnant mare serum gonadotropins (PMSG), injected at the time of sponge removal or 48 hours prior to sponge removal. Intra-vaginal sponges have high retention rates (>90%) and females usually show estrus 24 to 48 hours after removal. Pregnant mare serum gonadotropin (PMSG) is a glycoprotein hormone synthesized in the domestic horse placenta. Commercial preparations are isolated from the serum of pregnant mares. It is a unique member of the gonadotropins family which contains high levels of FSH and LH activity. Its ability to stimulate ovulation is determined in a bioassay, and the results are

expressed as international units (IU). PMSG that has been purified to homogeneity has approximately 16,000 IU/mg protein. PMSG Hormone is a well know used hormone together with progestogen to increase ovulation and to induce ovulation in livestock prior to artificial insemination. FSH and LH secretion from the pituitary gland blocks by progesterone and therefore follicles do not develop and grow on the ovary, consequently blocking the estrous cycle. It is administered for a number of days and then discontinued. Once removed, the block it had on FSH and LH secretion is removed. The decreased progesterone level results in the release of FSH and LH, follicular development and maturation in the ovary, and oestrus can be expected in 24 to 36 hours of removal (Tsuma *et al.*, 2015).

#### 2.6.1.3 Gonadatropins releasing hormone and Prostaglandin (GnRH-PGF2 $\alpha$ based synchronization protocol

A combination of GnRH and Prostaglandin are good for cyclic females and this combination may induce estrus in cows experiencing postpartum anoestrus. The use of GnRH provides a means to standardize the pattern of follicular growth in a majority of animals (Doleiel *et al.*, 2002). GnRH is typically given the first day of a synchronization system to program emergence and growth of the subsequent wave of follicles through inducing ovulation of dominant follicles in the ovary. Through removal of the existing dominant follicle, the emergence of a new wave of follicles approximately two days later is achieved in most females (Michael and Thomas, 2005, Islam, 2011; Aemat, 2017). The initial GnRH injection (day 1; GnRH) is used to program follicle growth in cyclic females and to induce ovulation (to provide progestin pre-exposure) in anestrous females. The PGF2 $\alpha$  (PG; day7) induces deterioration of CL that are present to cause a decline in progesterone. The second GnRH given on day 9-10 encourages ovulation of dominant follicles that have been pre- programmed by the first GnRH treatment. The four systems for synchronization of estrus with GnRH-PG combinations are ovsynch, Co synch, select synch and hybrid synch (Islam, 2011).

### **Synchronization of Ovulation (Ov-synch)**

The program “ov-synch” stands for synchronization of ovulation which is a fixed-time A.I. synchronization protocol that has been developed, tested and used extensively in lactating dairy cattle (Pursley *et al.*, 1997). The program starts with an injection of GnRH on day 1, an injection of prostaglandin on day 7, a second injection of GnRH on day 9 and then timed insemination on day 10 (Richardson *et al.*, 2002). This program has advantages of close-fitting synchronization of estrus; most females respond to the program and it encourages estrus in non-cycling cows that are at least 30 days postpartum with some recognized disadvantages of the relative expense and close management of females to be handled three times before breeding. Short-term calf removal (48 hours) following the prostaglandin injection may improve the response in postpartum cows (Richardson *et al.*, 2002).

### **Co-synch**

The co-synch program starts with an injection of GnRH on day 1 following with an injection of prostaglandin on day 7 and then a second injection of GnRH with breeding on day 9. This program’s advantages are tight synchronization of estrus, most females respond to the program and it encourages estrus in non-cycling cows that are at least 30 days postpartum with the disadvantage of cost for hormones and tight follow-up and twice handling of cows before breeding. With this program, Cows will show improved estrus response when 48 hour calf removal is utilized after the prostaglandin injection (Perry, 2012).

### **Select-synch**

An injection of GnRH on day 1, injecting females with prostaglandin on day 7 and then estrous detection and AI at day 8 collaborate with the idea of the Select-Synch program. The advantage of this program is lower cost and reduced handling compared with Ovsynch and CO-Synch programs with the primary disadvantage of the time required for estrous detection (Richardson *et al.*, 2002).

## **2.7 Factors affecting the success of estrus synchronization in Does**

Season, nutrition, health state, AI (Holtz, 2005) and type of hormone administered (Gordon, 1997) are known to contribute to variability in efficiency of estrus. Previous super-ovulation protocol in our laboratory was consisted of 70 mg FSH (Ovagen; ICPbio Limited, New Zealand) and 1000 IU hCG (Ovidrel; Laboratories Serono, Switzerland) per doe (Rahman *et al.*, 2007b). However, even with lower doses of hormones similar OR rate at 36 h and significantly higher OR rates at 60 and 72 h time intervals were obtained. Currently, we further reduced the dose rate of hCG (Ovidrel) to 250 IU for 60 h time interval and OR rates increased to double (Rahman, 2008). It is not clear whether this increment of OR rates was the effect of time interval or hormonal dose rates or combined effects of time interval and hormonal dose rates. However, decreasing hormonal dose rates alone might not be responsible for increasing OR rates as both higher and lower dose rates of hormones at 36 h time interval provided similar OR rates in the previous studies in our laboratory.

## **2.8 Estrous Detection**

Sexual receptivity in the female is expressed in a short period of time known as estrus. Estrus is the period during which the female will stand to be bred by the male (Audra, 2014). Females undergo behavioral changes during the estrous period that indicate sexual excitement and receptivity to the male. Goats experience an estrous period lasting an average of 36 h with a range from 19 to 48 h (Abecia *et al.*, 2011). In goats, estrus can be induced with the strategic exposure of anestrus does to intact males. This response is dependent on the depth of seasonal anestrus and associated with a first ovulation in two to three days after the introduction of the buck. The response to the male effect is influenced by the sexual aggressiveness of the buck, the intensity of the stimulation and the body condition of the does. Immediate contact results in a greater response than fence-line contact or intermittent contact. The pheromones responsible to induce estrus are present in buck hair, but not in urine, and are not associated with buck odor during the breeding season. Females in all species exhibit an increase in physical activity as the estrous period approaches, which is generally observed as increased locomotion, increased vocalization, urination, tail flagging and aggressive behavior towards other females (Abecia *et al.*, 2011). The use of males with marking harnesses equipped with marking

chalk can be used to determine when females stand to be bred by the male (Fierro *et al.*, 2011).

In the criteria of estrous response, fertility rate, kidding rate, fecundity rate and prolificacy rate, using CIDR device for synchronization of Iranian downy goat resulted for 100%, 84.2%, 84.2, 89.7% and 106.3% efficiency rate respectively which were comparable with reports of other researchers who reported 94.4% after using FGA and eCG (Motlomelo *et al.*, 2002). Estrus response of Iranian downy goats with CIDR device and eCG protocol was higher than the estrus response of 84% observed in Sahel goats (Omontese *et al.*, 2012) and 66.7% in Saanen dairy goats (Kajaysri and Thammakarn, 2012). According to the above indicated report, all of the treatments (except PGF without eCG) were effective in synchronizing estrus in Iranian indigenous downy goats furthermore, the use of CIDR, sponge or implant treatments alone or with eCG to be equally efficient in synchronizing estrus in goats (Majid and Mazaher, 2017). This shows that even though CIDR contains a less potent hormone (i.e. progesterone), its efficiency was comparable to that of progestagens. ) which was in agreement with (Bukar *et al.*, 2012) who found that the use of eCG concurrent with second injection of prostaglandin F2 $\alpha$  significantly increased estrus response. Some authors reported higher estrus response when progesterone or progestagens were used in association with gonadotrophins (Orontes *et al.*, 2012).

Although eCG enables the follicles to reach final maturation stage, it presents some limitations like, high cost and formation of antibodies following repeated synchronization treatments in a fraction of the goat population. Moreover, results reported by (Holtz *et al.*, 2008) revealed that timing of the fixed-time insemination was disorganized by the prolonged follicular phase of such females. The author also proved that, onset and density of estrus are important as they could affect the efficacy of fixed-time artificial insemination program. In progesterone-based groups a higher proportion of does were detected in estrus up to 36 hours following eCG.

## **2.9 Semen collection, processing and artificial insemmination in Goats**

### 2.9.1 Semen collection

Semen is liquid or semi gelatinous cellular suspension containing the male gametes or spermatozoa and secretions from the accessory organs of the male reproductive tract ( Koray and Ali, 2016). In reproduction, the onset of the production of spermatozoa represents only the final step in a series of complex changes that govern their number and properties. The commonest methods used for collection of buck semen are the artificial vagina (AV) and the electro ejaculator (EE). The most commonly used and easiest procedure is the use of the AV. The AV method is painless, quicker and does not stress the animal at all. The AV resembles a car radiator hose and is about six inches in length. It has an inner rubber liner (containing water at a temperature of 100°F) placed between the liner and the hose (Tsuma *et al.*, 2015). The warmer water simulates the vagina of a doe. A latex rubber collection cone is placed in the AV and a graduated collection tube is placed on the end of the cone (Islam *et al.*, 2008).

### 2.9.2 Semen quality evaluation

For increasing the use of AI in the goat, semen evaluation of the buck has assumed considerable importance. Consideration of optimal level for concentration, volume and motility will be important for harvesting fertile semen. After collection and evaluation, the semen that meets the required standards is extended to increase its volume so that it can give many breeding doses. Semen extension involves use of semen diluents or extenders that will nourish the spermatozoa and provide a good environment for their survival (Koray and Ali, 2016). Volume is an important criterion in semen evaluation and the average buck semen volume to be between 0.5 and 1.0 ml (Smith, 1978). The quality of the semen may decrease as the total volume of the ejaculate increases (Huat, 1976). But, generally larger volumes mean more sperm. Older bucks generally have a larger volume of ejaculate than younger bucks. Therefore, age should be considered with respect to this parameter of semen evaluation. It is best to make the motility estimate as soon after the semen is collected as possible. Since good semen handling dictates that the collection tube should be at 37°C at the time of collection, all of the things that come in contact with the semen should be at 37°C. One item that is often neglected is the temperature of the microscope stage. Although the semen does not come in direct contact with the stage,

within a few seconds of placing the slide on the microscope, the slide temperature will be that of the microscope. To avoid this problem, a device known as a stage warmer can be used to keep the slide at 37°C while it is being examined (Smith, 1978).

### 2.9.3 Artificial insemination in Goats

Artificial insemination is a technique whereby semen collected from the buck is deposited by an AI technician in the right place in the reproductive tract of the doe at the appropriate time (Tsuma *et al.*, 2015). AI technology is currently the most practical technology for optimizing reproductive efficiency is well known for its use in animal breeding programme to accelerate improved genetics and to better disseminate genetically and economically important traits to the wider production population. Artificial insemination enables farmers to introduce superior germplasm, accelerates the rate of genetic improvement within a herd, maximizes the number of offspring from a desirable sire, and enables genetic exchange over wide geographical areas (Naqvi, *et al.*, 2000). Semen collection, semen processing, storage and the actual insemination are the sub steps under AI (Tsuma *et al.*, 2015).

## Chapter 3: MATERIALS AND METHODS

### 3.1. Description of the Study Area

The study was conducted both at on-station and on-farm experimental conditions. The on-station experiment was conducted at Aybra main research station under Sekota dry land agriculture research center while the on-farm experiment were conducted at Workadivno and Bilaqu CBBP villages where the community based goat genetic improvement program was implemented.

Table 3. 1 Description of the study sites

Study Sites	Latitude	Longitude	Altitude (Masl)	Distance from Addis and Sekota(km)	Rain fall(mm)	Temperature (°c)
Aybra	12°43'65"	39°02'027"	1978	737 and 17	300-700	20.33 °c
Workadivno	12°58'12"	39°04'25"	1389	770 and 50	300-700	23- 37°c
Bilaqu	12°48'41"	38°43'43"	1343	785 and 65	255	22 °c

Source: <http://gismap.ciat.cgiar.org/MarkSimGCM/>, Alubel Alemu, 2015

The rain fall distribution was very erratic with short rainy season from June - August.

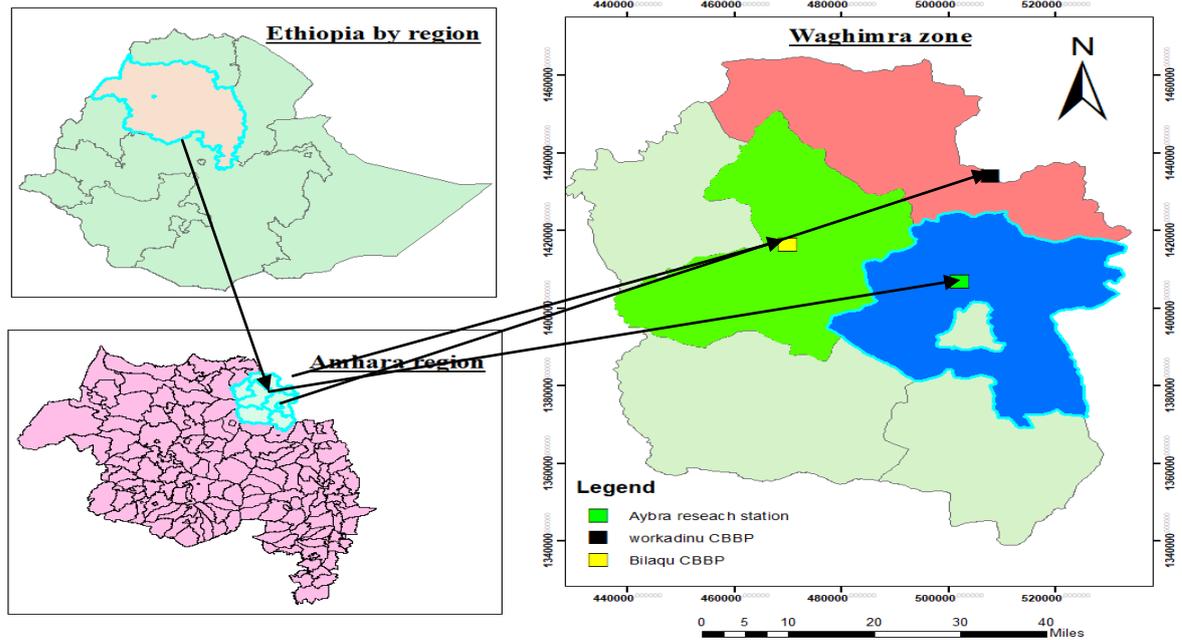


Figure3.1 Map of the study area

### 3.2. Experimental Animals and Management

#### Experiment 1 – On-station experiment

A total of 92 Abergelle does of age ranging from 2 - 6 years, with at least first parity, non-pregnant, normal health condition, non-suckling and with body condition score from 2.5 – 3.5 (scale 1= emaciated–5= very fat) and scored by palpation of lumbar region (Koyuncu and Altınçekiç, 2013) were used for the experiment. Five Abergelle bucks of age ranging from 3 – 6 years, with normal health condition and fit for different morphological criterions were also used for the experiment. Does were allowed to graze/browse daily for 8 hrs at the natural pasture with dominantly abundant acacia and other browse species around the research site. A mixture of 300-350g cowpea hay and commercial concentrate/wheat bran per day were supplemented for each animal. Doe's were supplemented with the above ration for about one month before the actual experimental time while bucks were graze/browse and exercise around the research station for about 4 hrs per day in separate from Does to be kept by anther attendant with supplementation of the above specified rations of which commercial concentrate/wheat bran were purchased from Sekota feed processing plant and available at the research station as per the schedule where as the cow pea hay were obtained from the available source at the station and fed for about one and half month prior to the actual experiment as spermatogenesis takes

about 47 days. Prior to the experiment, animals were quarantined and vaccinated against anthrax which was common to the area prior to the experiment period. Previous history of Does for their exposure for hormonal estrus synchronization was checked for systematic distribution of previously synchronized does across each treatment groups. Unfortunately, there were no Does exposed for hormonal treatments earlier.

### **3.3 Experiment procedures**

The experiment was carried out during the main breeding seasons of the breed (mid June-mid July). At each experimental conditions (both at the station and CBBP villages), Estrus synchronization, semen quality analysis and AI activities were conducted.

#### **3.3.1 Estrus Synchronization at Station condition**

Experimental animals (92) were divided into four treatment groups of 25 animals for treatment one up to treatment three and 17 animals for control groups as follows.

**Treatment 1:** Single injection of prostaglandin

**Treatment 2:** Double injection of prostaglandine 11 days apart

**Treatment 3:** PMSG (pregnant mare serum gonadatropins) with Enzaprost

**Treatment 4:** With no hormonal treatment (Control)

#### **Treatment one: Single injection of prostaglandin**

First, pregnancy was checked very seriously using ultrasound scanner before hormone administration since the prostaglandin analogs causes' immediate abortion for pregnant does. Non-pregnant doe's were intramuscularly administered with 1ml Enzaprost® (one of the analogs for prostaglandin). Estrus detection was conducted six times a day with four hours interval for 48 hrs. Does coming to heat were identified using proven bucks fitted with an apron and by observing physical and behavioral change of the doe (bleating, vigorous tail movement, mucus discharge and restlessness). All does showing estrus were inseminated with fresh semen collected from Abergelle breeding bucks. A 0.25 ml straw containing approximately 150 million sperm cells with 60% and above mass motility rate were used for insemination of the does. AI has performed once by professional inseminator.

### **Treatment two: Double injection of prostaglandine 11 days apart**

Before hormone administration, same process with the above was employed for checking pregnant animals. Does were intramuscularly administered at day 1 with 1ml Enzaprost® followed by another injection of 1 ml Enzaprost® at day 11. Estrus detection was conducted six times a day with four hours interval for 48 hrs. Does coming to heat were identified using proven bucks fitted with an apron and by observing physical and behavioral change of the doe (bleating, vigorous tail movement, mucus discharge and restlessness). All does showing estrus were inseminated with fresh semen collected from Abergelle bucks. A 0.25 ml straw containing approximately 150 million sperm cells with 60% and above mass motility rate was used for insemination of the does. AI has performed once by professional inseminator.

### **Treatment three: PMSG (pregnant mare serum gonadatropins) with Enzaprost**

After ultrasound detection for pregnancy, doe's were treated with progesterone impregnated vaginal sponges (Syncro-part ®; CEVA laboratories, Libourne, France) for 11 days. The equipments to be used to insert the sponge into the vagina, the sponge, and the vagina had disinfected and cleaned from any dirt/contamination following the standard procedures. Sponge insertion and other activities had been conducted very carefully to avoid injury. Lubricants were used for lubrication purpose. Animals were injected intramuscularly with 1ml gonadatropins 48 hrs prior to the removal of the sponges. Estrus detection was conducted six times a day with four hours interval for 48 hrs. Does coming to heat were identified using proven bucks fitted with an apron and by observing physical and behavioral change of the doe (bleating, vigorous tail movement, mucus discharge and restlessness); similar approaches with the above. All does showing estrus had inseminated with fresh semen collected from Abergelle breeding bucks with in 48 hrs after the removal of vaginal sponges. A 0.25 ml straw containing approximately 150 million sperm cells with 60% and above mass motility rate was used for insemination of the does. Artificial insemination had performed once by professional inseminator.

### **Treatment four: Control**

For the comparison of the effect of each of the treatment groups, some does were assigned (without any hormonal treatment) to cycle and induce estrus under normal physiological conditions. About 17 Does were selected with similar criterion as above; allow having similar management conditions (feeding, health and separate herding) which were similar with other treatment groups. Since it was difficult to detect heat with no hormonal treatment, estrus data was not taken rather data were recorded after conception.

### **3.4 Experiment 2 – On-Farm**

A total of 243 Abergelle does of age range from 2- 6 years, at least at first parity, non pregnant, normal health condition, non suckling and with body condition score from 2.5 – 3.5 were selected from participant farmers flocks from the two CBBP villages. Non-pregnant does were identified using ultrasound scanner. Similarly, 18 Abergelle bucks of age ranging from 3 to 6 years with best breeding value profile estimated earlier during selection and by considering other parameters (body conformation, color, horn size and orientation and testicle size) with equal proportion from the two CBBP villages were assigned for the experiment.

Except the number of experimental animals used, the remaining procedures for each treatment groups were similar with the on-station experiment. Treatment one was done at Workadivno village while treatments two and three were done at Bilaqu village which was based on the access for the required number of experimental animals. Treatment four (control) was used at each of the villages. Treatment groups were arranged as presented below.

**Treatment 1:** Single injection of prostaglandin (58 Does)

**Treatment 2:** Double injection of prostaglandine 11 days apart (72 Does)

**Treatment 3:** PMSG (pregnant mare serum gonadotrophin) with Enzaprost (73 Does)

**Control** (40 Does of 20 Does from each village)

The experimental animals were isolated from farmer's flock and kept by separate attendants in a group according to the treatment groups and allowed for grazing/browsing at their usual pasture area followed by supplementation of mixture of 300-350g cowpea hay and commercial concentrate/wheat bran per day for each animal for about one month prior to the actual experiment while the Bucks were flushed with the above mentioned rations for about one and half months prior to the actual experiment. Prior to the experiment, Animals had quarantine and vaccinated against anthrax which was common at the area during that period. The experimental animals were returned back to farmers after 10 days of insemination date.

### 3.5. Semen Collection, Processing and Artificial Insemination

Semen was collected from 5 bucks from the station and 18 bucks from the CBBP villages and trained to serve with artificial vagina following a standard procedure (Tusma *et al.*, 2015). Bucks were trained to serve with artificial vagina 10 days prior to the actual experiment date. To stimulate bucks for collection, 2-3 Does were injected with 2 ml 17 $\beta$ -estradiol (Jane, 2007) before 16 hr of semen collection. About 10 successful semen samples/ejaculates were collected at the station and about 33 collections were obtained from the CBBP villages. The collected semen was immediately evaluated for volume, appearance (color, contamination), motility, and concentration. Semen was diluted with commercial extender called "Ovixcell" which is a specific extender for sheep and goat semen commercialized by IMV Technologies in France for the reason that to increase its volume and improve viability. Before dilution the extender was kept under - 4 $^{\circ}$ c and the dose of the extender was determined based on total number of spermatozoa per ejaculate, number of spermatozoa per insemination, total volume per ejaculate and concentration. Thus the total required volume of extender to be added was calculated using the following formula:

$$= \left( \frac{\left( \frac{\text{Tot.No.of spermatozoa}}{\text{No.spermatozoa per insemination (150 mil.)}} \right) \times \text{straw vol. (0.25 ml)} - \text{Total volume}}{\text{Total volume}} \right)$$

The standard number of spermatozoa per insemination is expected to be from 150- 250 x10 $^6$  (Vera *et al.*, 2012). For this experiment 150x10 $^6$  spermatozoa per insemination were

used since it the above specified extender was used. Total number of straws needed was also calculated as total number of spermatozoa per ejaculate divided by the standard number of spermatozoa per insemination ( $150 \times 10^6$ ). The total straw volume to be used for one insemination was 0.25 ml. During the semen processing phase, total number of motile spermatozoa per ejaculate was calculated as volume multiplied by concentration and the Semen had collected up to 3<sup>rd</sup> ejaculate per buck and evaluated for the above parameters. Mass motility was checked with rank from 1- 5 (1= poor, 5= excellent) by observing with microscope. Does were inseminated with fresh diluted semen within fixed time of 48 hrs of hormonal administration by opening the vagina gently using speculum (Paulenz et al., 2005). During the whole process of hormonal application and insemination, a skilled veterinarian was used.

### **3.6. Farmers Perception Survey**

A questionnaire survey was conducted on a total of 46 goat keepers which was purposely selected from the two CBBP villages (29 from experiment and 17 from control groups) with criteria of having goats for Estrus synchronization and artificial insemination experiment. Individual questionnaire was made using semi- structured questionnaire on the issues of the overall outlooks and feedbacks of reproductive technologies, goat rearing experience, trend of reproductive characteristics of their goat, the mating method they follow and season, reproductive management of goats, buck usage and sharing modality extra, and overall outlooks on estrus synchronization and AI technologies used so far.

### **3.7. Data collected**

- Date of hormone administered
- Date of first estrus sign
- Duration of estrus
- Semen quality records (Volume, Motility and Concentration)
- Date of insemination
- Failure to estrus
- Conception records (Pd)
- Kidd survival rate
- Farmer's perception and feedback records

- Estrus response, Conception and kidding rate were calculated by using the following formula:
- Conception rate =  $\frac{\text{Does concieved}}{\text{Does inseminated}} \times 100$
- Kidding rate =  $\frac{\text{Kids born}}{\text{Does concieved/pregnant}} \times 100$
- Estrous response =  $\frac{\text{Does show oestrus}}{\text{does administerd hormone}} \times 100$

### 3.8. Statistical analysis

Data collected from the experiment and survey was coded and entered into Microsoft excel of 2007 software program for further analysis. Preliminary data analysis like normality test was employed before conducting the main analysis. Data on estrus response, conception rate and kidding rate were analyzed using the Gen mode procedure of Statistical Analysis System SAS (9.0), List square means for dependent variable (onset and duration of estrus, litter size, birth weight) and semen quality parameters were computed by the GLM procedure of the Statistical Analysis System SAS (9.0). The model used for analyzing onset of estrus and estrus duration was:

$$Y_{ijklm} = \mu + Mt_i + Pr_j + Pa_k + Ag_l + BC_m + e_{ijklm}$$

Where:  $Y_{ijlm}$  = the observed time for onset and duration of estrus

$\mu$  = overall mean

$Mt_i$  = is the effect of  $i^{\text{th}}$  mgt condition

$Pr_j$  = is the effect of  $j^{\text{th}}$  protocol

$Pa_k$  = is the effect of  $k^{\text{th}}$  parity

$Ag_l$  = is the effect of  $l^{\text{th}}$  Age

$BC_m$  = is the effect of the  $m^{\text{th}}$  body condition score

$e_{ijklm}$  = is random residual error

The model used for the analysis of litter size and birth weight was:

$$Y_{ijk} = \mu + Mt_i + Pr_j + Pa_k$$

Where:  $Y_{ijk}$  = the observed litter size and birth weight

$\mu$  = overall mean

$Mt_i$  = is the effect of  $i^{\text{th}}$  mgt condition

$Pr_j$  = is the effect of  $j^{\text{th}}$  protocol

$Pa_k$  = is the effect of  $k^{\text{th}}$  parity

$e_{ijk}$  = is random residual error

The model used for semen volume, motility and concentration was:

$$Y_{ijk} = \mu + Mt_i + Fe_j + Bcs_k + Ag_l$$

Where:  $Y_{ijkl}$  = the observed semen volume, motility and concentration

$\mu$  = overall mean

$Mt_i$  = is the effect of  $i^{\text{th}}$  mgt condition

$Fe_j$  = is the effect of  $j^{\text{th}}$  frequency of ejaculate

$Bcs_k$  = is the effect of  $k^{\text{th}}$  body condition score

$Ag_l$  = is the effect of  $l^{\text{th}}$  age

$e_{ijk}$  = is random residual error

Each of the means were separated with Tukey karmers test and significant differences was test at  $\alpha = 0.05.$ , Index technique was used to rank the different parameters of goat breeding and management. SPSS version 16 was used to compare keeper's perception with Likert scale for the reproductive technologies data analysis.

## Chapter 4: RESULT AND DISCUSSIONS

### 4.1 Estrus response

The result of the current study revealed that estrous response among the three treatment groups were varied with noticeable difference for onset of estrus (Table 4.1). Estrus response for the Does injected with PMSG+Enzaprost<sup>®</sup> was higher (92%) at on-station (Table 4.2) and (87.5%) at on-farm condition (Table 4.3) respectively with same protocol followed by single injection of PGF2 $\alpha$  (72%) at on-station and (56.8%) at on-farm management conditions. The higher result for estrus response in case of PMSG+Enzaprost<sup>®</sup> might be associated with the reason that the inclusion of gonadatropins as co-treatment improves estrus response (Omontese *et al.*, 2013) and the protocol associating sponges with injection of PMSG and prostaglandin is a universal protocol which is valid for anoestrus goats and cycling goats when these are in the same flock so that goats which are anoestrus will be induced to ovulate with PMSG and goats already cycling with a functional corpus luteum will be synchronized with prostaglandin (Abecia *et al.*, 2012). So that since the breed is experiencing seasonal pattern of breeding and the season of hormone application were at mid June which was the transition time to their breeding, most of the Does might be brought to normal cycling state with PMSG so that the prostaglandin can easily induce estrus even though some physiological disturbances like formation of puss and vaginal ulcer were observed from the does treated with vaginal sponges as ewes treated intra vaginally with sponges showed histological and cytological alterations in the vaginal wall as assessed by hemorrhage and per vascular infiltrate and an increased number of epithelial cells, neutrophils, macrophages and erythrocytes (Manes *et al.*, 2015). Estrus result of PMSG+Enzaprost<sup>®</sup> was exceeded than 75% response from PGF-eCG protocol in Iranian downy goats reported by (Majid and Mazaher, 2017) though the difference might be associated with the reason that hormone change from PMSG to eCG between the two studies, breed difference and agro ecology. However, it was comparable with the result 94.4% reported by (Bukar *et al.*, 2012) in boar does with same protocol but lower than 100 % obtained by (Dogan *et al.*, 2004) in Saanen does treated with 750 IU PMSG and 97.4% in Somali goats of Ethiopia treated with intra vaginal sponges (MAP and FGA).

Relatively lower estrus response was revealed from the Does subjected with double injection of PGF2 $\alpha$  (36%) at on-station (Table 4.2) and (57.5%) at on-farm (Table 4.3) management conditions which was lower than the result 95% found in indigenous Greek goats (Amarantidis *et al.*, 2004), the 97% in Nadooshani goats (Bitaraf *et al.*, 2007) and 81.25% in Ethiopian local goats (Alemat Gidena, 2017). However it was higher than 30% estrus response reported by (Majid and Mazaher, 2017) in Iranian downy goats and lower than 73.7% reported by (Rekik *et al.*, 2014) with similar protocol. Whereas the estrus response from single injection of PGF2 $\alpha$  in the current study (72%) at on- station (Table 4.2) and (56.8%) at on-farm (Table 4.3) management condition was lower than other findings (93%) in Rambouillet and Dorper synthetic ewes (Audra, 2014) using CIDR+ PGF protocol.

In the present study, it was very likely that the overall time for the onset of estrus for double injection of PGF2 $\alpha$  was displayed very early ( $9.5\pm 0.58h$ ) which was significantly earlier ( $p < 0.001$ ) with relatively longer estrus duration ( $42.1\pm 1.56h$ ) compared with the other protocols (Table 4.1). Similarly, double injection of PGF2 $\alpha$  resulted significantly lower ( $p < 0.05$ ) time for the onset of estrus ( $10.4\pm 0.65h$ ) at the on-station experiment (Table 4.2) and significantly lower ( $p < 0.001$ ) time for the onset of estrus ( $10.7\pm 0.74h$ ) for the on-farm management condition (Table 4.3) was revealed for the same protocol. However, no significant divergences at ( $p > 0.05$ ) among the three treatment groups were revealed for the duration of estrus at both of the management conditions.

The study also revealed that early to mid parity does (1<sup>st</sup>, 2<sup>nd</sup> and 3<sup>rd</sup>, ) with body condition score from 2.5 up to 3 were found to be more responsive for estrus and had better time for the duration of estrus which were in agreement with the finding reported that body condition score of ewes had a significant effect ( $P < 0.05$ ) on kilogram lambs born per ewes, birth weight of lambs and FSH concentration of ewes in Sanjabi ewes (Muhammad *et al.*, 2013). The author also reported that Ewes with body condition score of 3 had normal estrus while ewes with lower body condition score had shorter estrous period. Positive relationship between body condition score and plasma leptin and FSH concentrations in Iranian fat tail ewes at mating time (Towhidi *et al.*, 2007) was also another finding which strengthened the current result.

Table 4. 1 The effect of hormone, management condition, parity, Age and Bcs on estrus response (%), Onset of estrus (h) (LSM±SE) and estrus duration (h) (LSM±SE) of Abergelle Does.

Variables	N	Estrus response (%)	Onset of estrus(h) (LSM±SE)	Estrus duration(h) (LSM±SE)
Over all	278	188 (67.6)	14.4±0.47	45.5±0.45
Cv			36	21
Treatments				
Management condition		NS	***	Ns
On-station	75	50 (66.6)	11.9±0.62 <sup>b</sup>	39.5±1.44
On-farm	203	137 (67.4)	15.5±0.47 <sup>a</sup>	42.3±0.87
Protocol		***	***	Ns
PMSG+Enzaprost <sup>®</sup>	97	85 (87.6) <sup>a</sup>	12.8±0.66 <sup>b</sup>	39.0±1.09
PGF2 $\alpha$ (Single)	83	51 (61.4) <sup>b</sup>	18.8±0.85 <sup>a</sup>	41.6±1.39
PGF2 $\alpha$ (double)	98	52 (53) <sup>b</sup>	9.5±0.58 <sup>c</sup>	42.1±1.56
Parity		NS	Ns	**
1 <sup>st</sup>	21	14 (66.6)	12.9±0.82	41.8±2.84 <sup>a</sup>
2 <sup>nd</sup>	56	33 (58.9)	13.8±1.19	43.5±1.79 <sup>a</sup>
3 <sup>rd</sup>	111	82 (73.8)	13.3±0.75	46.0±0.75 <sup>a</sup>
4 <sup>th</sup>	61	41 (67.2)	12.7±0.99	38.7±1.72 <sup>b</sup>
5 <sup>th</sup>	19	12 (63.1)	14.8±2.11	36.7±2.53 <sup>b</sup>
6 <sup>th</sup>	8	6 (75)	14.6±0.84	38.7±3.52 <sup>b</sup>
Age/dentition		Ns	Ns	Ns
1p	4	2 (50)	10.6±4.00	34.3±2.00
2p	47	28 (60.8)	14.4±1.28	41.8±2.08
3p	60	42 (71.4)	15.2±1.08	43.8±1.67
4p	151	107 (71)	14.6±0.58	43.7±0.93
Bcs		Ns	**	Ns
2.5	136	90 (65.9)	12.8±0.75 <sup>a</sup>	40.6±1.03
3	84	60 (72)	12.0±0.57 <sup>b</sup>	40.2±1.35
3.5	42	29 (69)	16.3±1.20 <sup>a</sup>	41.9±1.89

Where: PMSG=Pregnant mare serum gonadatropins, h= hours PGF2 $\alpha$  =Prostaglandin f2  $\alpha$ , N=total population, Bcs= body condition score, LSM= least square means and SE= standard error, '\*'=p<0.05, '\*\*'=p<0.01 '\*\*\*' =p<0.001

Table 4. 2 The effect of hormone on estrus response (%), Onset of estrus (h) (LSM±SE) and estrus duration (h) (LSM±SE) on Abergelle Does at station condition

Hormone	N	Estrus response (%)	Onset of estrus (h) (LSM±SE)	Duration of estrus (LSM±SE)
Overall	75	50 (66.6)	12.1±0.47	43.2±1.44
Cv			26.7	24.1
Protocol		***	*	Ns
PMSG+Enzaprost®	25	23 (92) <sup>a</sup>	11.8±0.73 <sup>a</sup>	41.7±2.26
PGF2α (Single)	25	18 (72) <sup>b</sup>	13.5±0.80 <sup>a</sup>	43.7±2.45
PGF2α (double)	25	9 (36) <sup>c</sup>	10.4±0.65 <sup>b</sup>	45.6±2.99

Where: PMSG=Pregnant mare serum gonadatropins hormone, h= hours PGF2α = Prostaglandin f2 α, N= total population, LSM= least square means and SE= standard error, '\*'=p<0.05

Table 4. 3 The effect of hormone on estrus response (%), Onset of estrus (h) (LSM±SE) and estrus duration (h) (LSM±SE) on Abergelle Does at on-farm condition

Hormone	N	Estrus response (%)	Onset of estrus(h) (LSM±SE)	Duration of estrus(LSM±SE)
Overall	203	138 (67.9)	15.1±0.60	46.0±0.86
Cv			38	21
Protocol		***	***	Ns
PMSG+Enzaprost®	72	63 (87.5) <sup>a</sup>	14.4±0.84 <sup>b</sup>	44.5±1.23
PGF2α (Single)	58	33 (56.8) <sup>b</sup>	21.8±0.90 <sup>a</sup>	47.8±1.67
PGF2α (double)	73	42 (57.5) <sup>b</sup>	10.7±0.74 <sup>c</sup>	46.9±1.70

Where: PMSG=Pregnant mare serum gonadatropins, PGF2α =Prostaglandin f2 α, N= total population, LSM= least square means and SE= standard error, '\*'=p<0.05, '\*\*' =p<0.01, '\*\*\*'=p<0.001

### Onset of estrus and estrus duration

The result of the current study showed that at both of the management conditions ( Station and On-farm), most of the Does were started to show estrus within 8 up to 16 h of hormone injection. For instance for the on station experiment, 91.3% of the PMSG group, 83% of the single injection of PGF2α group and 100% of the double injection of PGF2α group experienced estrus with in 8 up to 16 h of hormonal treatment ( Fig 4.1). Similarly,

61.9% of the PMSG group, 27.2% of the single injection of PGF2 $\alpha$  group and 66.6% of the double injection of PGF2 $\alpha$  group show estrus within 8 up to 16 h of hormonal treatment at the on farm management condition (Fig 4.1). For the double injection of PGF2 $\alpha$ , 36% of the Does were in estrus from 36h up to 48h and 48% of the Does were stayed in estrus for about 52h up to 60h from hormone treatment (Fig 4.2). overall estrus duration ( $45.5\pm 0.45$ h) was shorter which might be associated with the reason for lower conception rate of Does since the insemination was done after 48h fixed time so that in case of Abergelle Does, the approach may not best suit with field application of 48h timed artificial insemination. The result for onset of estrus in this study was comparable with the report ( $11.0 \pm 2.84$  h) (Ayele Abebe *et al.*, 2018) for Menz ewes synchronized with GnRH and much earlier than ( $37.1 \pm 2.64$ h) in case of single injection of PGF2 $\alpha$  with same report where as the duration of estrus for the current study was slightly longer than  $34.97\pm 3.74$ h obtained from Ethiopian local goat treated with double injection of PGF2 $\alpha$  (Alemat Gidena, 2017).

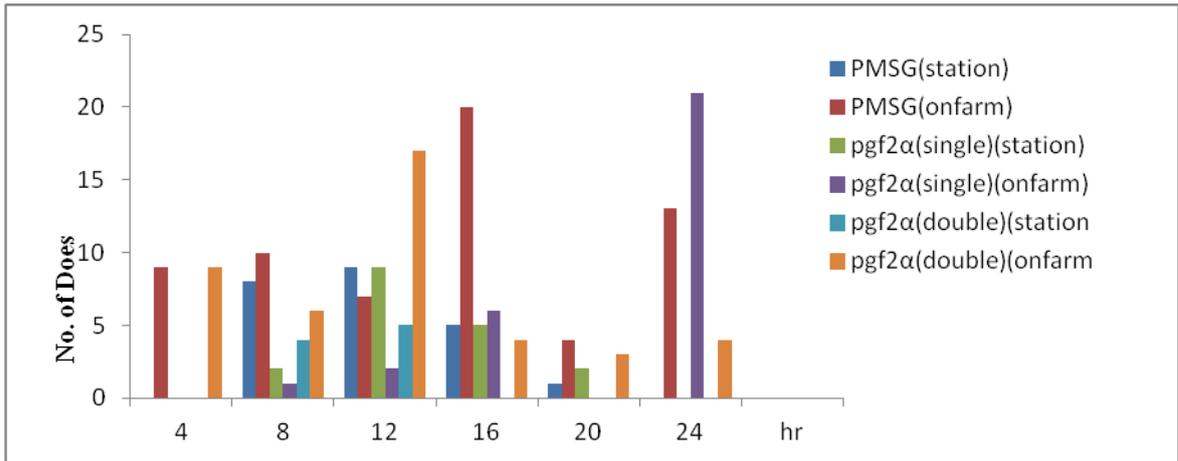


Figure 4.1. Time for the onset of estrus at on- station and on-farm management conditions

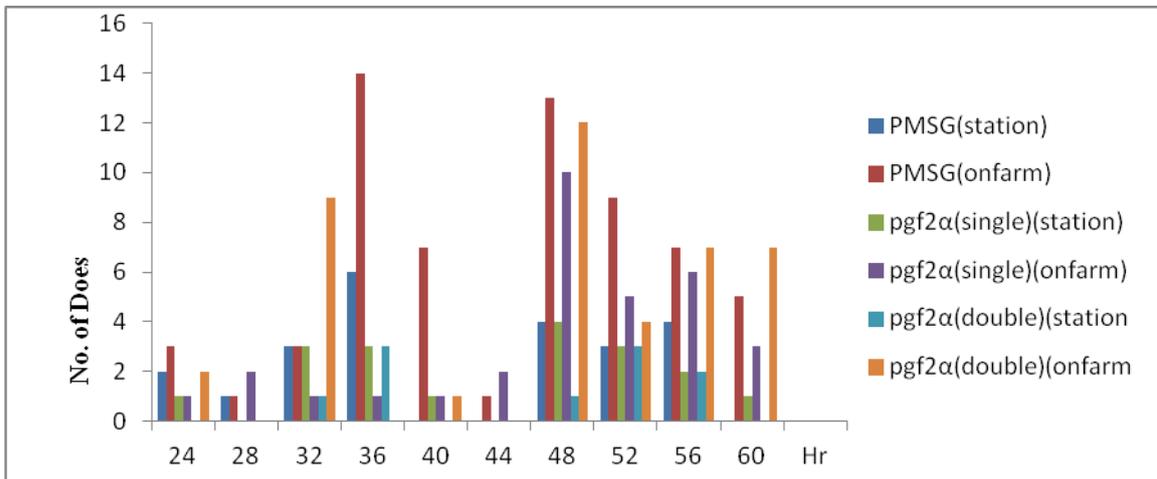


Figure 4.2 Time for estrus duration at on- station and on-farm management condition

#### 4.2 Conception rate, kidding rate, litter size and birth weight

The overall conception rate (65.4%) at both of the management conditions (Station and On-farm) were relatively higher than the control (52.6%) (Table4.4). Better conception rate (78.8%) from double injection of PGF2α followed by single injection of PGF2α (64.7%) (Table 4.4) was might be with the reason that its appropriate insemination time since most of the Does were in estrus from 48h up to 60h from hormonal treatment. In comparison with other results, the current result was higher than (29.4%) using natural mating and (44%) using AI on Rawa goats (Abid Mehmood ,2011) and (61.54%) in Ethiopian local goats (Alemat Gidena,2017) and comparable with the report (83.3%) from PGF groups in Iranian downy goats (Majid and Mazaher, 2017). Relatively lower result

was investigated by the current study for kidding rate which might be associated with due to the reason that prolonged dry season during the experiment period leading for serious early abortion. In the current study, PMSG+ Enzaprost® showed significantly higher ( $p < 0.05$ ) proportion of litter size ( $1.6 \pm 0.22$ ) compared with other protocols and the control group which was in agreement with the rationale of supper ovulatory response of equine Chorionic Gonadotrophin (eCG) or PMSG and FSH (Rahman *et al.*, 2008).

Table 4. 4 The effect of hormone, management condition and parity on conception rate (%), kidding rate (%), litter size (LSM±SE) and birth weight (LSM±SE) on Abergelle does

Variables	N	Conception (%)	Kidding Rate (%)	Litter size (LSM±SE)	Birth weight (LSM±SE)
Over all	188	123 (65.4)	110(89.4)	1.23±0.04	2.27±0.03
Cv				36	14
Treatments					
Management condition		NS	Ns	Ns	***
On-station	50	32 (64)	28 (87.5)	1.17±0.07	2.02±0.07 <sup>b</sup>
On-farm	138	91 (65.9)	82 (90.1)	1.19±0.05	2.34±0.03 <sup>a</sup>
Protocols		*	*	***	**
PMSG+Enzaprost®	85	49 (57.6) <sup>b</sup>	45 (91.8) <sup>a</sup>	1.65±0.1 <sup>a</sup>	2.0±0.07 <sup>b</sup>
PGF2α (Single)	51	33 (64.7) <sup>b</sup>	30 (90.9) <sup>a</sup>	1.0±0.0 <sup>b</sup>	2.3±0.06 <sup>a</sup>
PGF2α (double)	52	41 (78.8) <sup>a</sup>	35 (85.3) <sup>b</sup>	1.0±0.02 <sup>b</sup>	2.2±0.05 <sup>a</sup>
Control	57	30 (52.6) <sup>b</sup>	30 (100) <sup>a</sup>	1.0±0.04 <sup>b</sup>	2.1±0.04 <sup>a</sup>
Parity		Ns	Ns	Ns	Ns
1 <sup>st</sup>	14	6 (42.8)	3 (50)	1.17±0.0	2.3±0.19
2 <sup>nd</sup>	33	21 (63.6)	20 (95.2)	1.13±0.07	2.1±0.07
3 <sup>rd</sup>	82	62 (75.6)	55 (88.7)	1.17±0.06	2.2±0.05
4 <sup>th</sup>	41	24 (58.5)	21 (87.5)	1.26±0.1	2.2±0.06
5 <sup>th</sup>	12	5 (41.6)	5 (100)	1.37±0.24	1.9±0.10
6 <sup>th</sup>	6	4 (66.6)	4 (100)	1.0±0.0	2.1±0.06

Where: PMSG=Pregnant mare serum gonadatropins, PGF2α =Prostaglandin f2α, N= total population, LSM= least square means and SE= standard error, '\*'=p<0.05, '\*\*' =p<0.01 and '\*\*\*' =p<0.001

Table 4. 5 The effect of hormone on conception rate (%), kidding rate (%), litter size (LSM±SE) and birth weight (LSM±SE) on Abergelle Does at on station condition

Hormone	N	Conception Rate (%)	Kidding rate (%)	Litter size (LSM±SE)	Birth weight (LSM±SE)
Overall	50	32 (64)	28 (87.5)	1.1±0.08	2.0±0.08
Cv				32	20
Protocol		Ns	Ns	*	Ns
PMSG+ Enzaprost®	23	11 (47.8)	10 (90.9) <sup>b</sup>	1.6±0.22 <sup>a</sup>	1.7±0.16 <sup>b</sup>
PGF2α (Single)	18	14 (77.7)	12 (80.0) <sup>c</sup>	1.0±0.0 <sup>b</sup>	2.3±0.11 <sup>a</sup>
PGF2α (Double)	10	5 (50.0)	6 (100.0) <sup>a</sup>	1.0±0.0 <sup>b</sup>	2.1±0.16 <sup>b</sup>
Control	17	8 (47.0)	7 (87.5) <sup>c</sup>	1.0±0.0 <sup>b</sup>	2.0±0.09 <sup>b</sup>

Table 4. 6 The effect of hormone on conception rate (%), kidding rate (%), litter size (LSM±SE) and birth weight (LSM±SE) on Abergelle Does at on farm condition

Hormone	N	Conception rate (%)	Kidding rate (%)	Litter size (LSM±SE)	Birth weight (LSM±SE)
Overall	138	91 (65.9)	82 (90.1)	1.26±0.05	2.32±0.03
Cv				35	15
Protocol		**	*	***	*
PMSG + Enzaprost®	63	38 (60.3) <sup>b</sup>	36 (94.7) <sup>b</sup>	1.6±0.11 <sup>a</sup>	2.1±0.07 <sup>b</sup>
PGF2α (Single)	33	18 (54.5) <sup>b</sup>	18 (100.0) <sup>a</sup>	1.0±0.0 <sup>b</sup>	2.4±0.07 <sup>b</sup>
PGF2α (Double)	42	35 (83.3) <sup>a</sup>	29 (82.8) <sup>c</sup>	1.0±0.03 <sup>b</sup>	2.5±0.05 <sup>a</sup>
Control	40	21 (52.5) <sup>b</sup>	21 (100.0) <sup>a</sup>	1.0±0.06 <sup>b</sup>	2.2±0.05 <sup>b</sup>

Where: PMSG=Pregnant mare serum gonadatropins, PGF2α =Prostaglandin f2α, N= total population, LSM= least square means and SE= standard error, '\*'=p<0.05, '\*\*' =p<0.01 and '\*\*\*' =p<0.001

### 4.3 Semen Evaluation

In the current study, the overall average semen volume ( $0.64 \pm 0.03$  ml) obtained (Table 4.7),  $0.62 \pm 0.07$  ml (Table 4.9) and  $0.66 \pm 0.04$  ml (Table 4.10) from station and on-farm management condition, respectively does not show any significant difference at ( $P > 0.05$ ) with the factors of management conditions, ejaculate frequency, age and body condition of bucks except small variation observed among different bucks. The reason might be due to the reason that consideration of bucks with almost similar body condition, body size, similar feed supplementation and other management conditions during the experiment period as semen production of animal largely depends on several factors such as age, maturity, nutritional status, general health, endocrine balance and normality of sex organs (Peters, 2002). The findings of the present study were slightly higher than the result of (Singh *et al.*, 1985) obtained average ejaculate volume of 0.46 cc in black bengal buck, an average ejaculate volume of  $0.27 \pm 0.12$  ml, reported by (Khan, 1999) and an average semen volume ranging from  $0.43 \pm 0.03$  to  $0.45 \pm 0.22$  in black bengal bucks reported by (Islam *et al.*, 2008). The higher result revealed in the current study might be justifiable with the reason that semen collection was done during active breeding season of the breed as semen volume of buck was recorded to be larger in amount during the breeding seasons because the accessory glands are more active during that time (Karagiannidis *et al.*, 2000; Islam *et al.*, 2008) However, (Marx *et al.*, 1975) reported average ejaculate volume of 1.27 ml, (Koray and Ali, 2016) reported an average semen volume per ejaculate of  $1.0 \pm 0.2$  ml in norduz goats, which was much greater than the results of the present study. These differences could be due to the individual variation of bucks (breed) and seasonal variations, as seasonal variations affect sexual glands (Islam *et al.*, 2008). Furthermore, the values obtained in this study were more or less similar with other investigators,  $0.5 \pm 0.3$  ml reported by (Siddiqua *et al.*, 2016).

The present study also revealed that ejaculate frequency had no significant impact on the semen characteristics of Abergelle bucks which was more or less in line with (Shamsuddin *et al.*, 2000) found that repeated ejaculation may not change sperm motility and percentage of

abnormal spermatozoa but semen volume, sperm concentration and number of total sperm per ejaculate decline.

Body condition score had show significant effect at ( $p < 0.05$ ) for the mass motility of sperm at the on station experiment (Table 4.9) and the motility obtained in the current study (70% to 80%), supports the findings of (Khan, 1999) obtained motility of 73.87%, (Islam *et al.*, 2008) who reported 76 up to 78% mass motility for black bengal bucks, the findings of (Bhuskat *et al.*, 2000), Shamsuddin *et al.*, 2000) also supports the results of the present study with a small deviation of the values. However, relatively higher result (85.00%) was reported by (Koh, 1976) which might be due to animal variation as the former study include crossbred bucks together with others for the experimental and other factors such as seasonal variations, nutrition, management, frequency of semen collection, collection interval, temperature and pressure of AV and physical condition of the animal might affect motility (Nazir, 1988).

Comparably, highest result for semen concentration ( $3.1 \pm 0.2 \times 10^9$ ) and ( $3.1 \pm 0.1 \times 10^9$ ) was revealed for the on- station and on- farm bucks respectively which were exceeded than reports ( $2.853 \pm 0.12 \times 10^9$ ), ( $2.708 \pm 0.83 \times 10^9$ ), ( $2.549 \pm 0.87 \times 10^9$ ) and ( $2.434 \pm 0.81 \times 10^9$ ) for black Bengal bucks (Islam *et al.*, 2008). The findings of the study also collaborates with the results of (Singh *et al.*, 1985) obtained sperm concentration of 2.619 to  $2.910 \times 10^9$ /ml. and (Khan, 1999) obtained sperm concentration of  $3.777 \times 10^9$ /ml. Insignificant effect for frequency of ejaculate on semen concentration was revealed in the current study though different scholars investigated as frequency of ejaculation was important for sperm concentration; four to eight daily ejaculations have been found to significantly decrease semen volume, sperm motility, sperm concentration and number of spermatozoa per ejaculation in ram (Thwaites, 1995) and higher concentration of sperm was obtained when semen was collected once daily (Sharma *et al.*, 1969).

Table 4. 7 The effect of management condition and ejaculate frequency on semen characteristics of Abergelle bucks

Variables	N	Volume(ml) (LSM±SE)	Motility (%)	Concentration (LSM±SE)
Over all	43	0.64±0.03	3.8 (76)	3.14±0.11
Cv		41	16	25
Management condition		Ns	Ns	Ns
On-station	10	0.62±0.07	3.5 (70)	3.1±0.2
On-farm	33	0.66±0.04	4.0 (80)	3.1±0.1
Frequency of ejaculate		Ns	Ns	Ns
1 <sup>st</sup>	23	0.64±0.06	3.5 (70)	3.2±0.19
2 <sup>nd</sup>	15	0.59±0.05	3.8 (76)	2.9±0.15
3 <sup>rd</sup>	5	0.68±0.11	3.9 (78)	3.2±0.26

Table 4. 8 The effect of management condition on semen characteristics of Abergelle bucks

Management condition	N	Volume(ml) (LSM±SE)	Motility (%)	Concentration (LSM±SE)
Over all	43	0.64±0.03	3.8 (76)	3.1±0.11
CV		41	16	25
Management condition		Ns	Ns	Ns
On-station	10	0.65±0.04	3.9 (78)	3.1±0.14
On-farm	33	0.61±0.07	3.4 (68)	3.1±0.2

Table 4. 9 The effect of age, body condition and ejaculate frequency on Volume (ml) (LSM±SE), Motility (%) and Concentration (LSM±SE) of Semen collected from Abergelle Bucks kept at station condition

Variables	N	Volume (ml) (LSM±SE)	Motility (%)	Concentration (LSM±SE)
Over all	10	0.61±0.07	3.4 (68)	3.17±0.2
CV		34	24	17
Body condition score		NS	*	NS
3	4	0.83±0.04	4.4 (88) <sup>a</sup>	3.2±0.4
3.5	6	0.67±0.12	2.9 (58) <sup>b</sup>	3.1±0.22
Age/ dentition		NS	Ns	Ns
3p	4	0.78±0.06	3.3 (66)	3.3±0.4
4p	6	0.72±0.11	4.0 (80)	3.0±0.22
Frequency of ejaculate		NS	Ns	NS
1 <sup>st</sup>	5	0.56±0.09	3.1 (62)	3.4±0.39
2 <sup>nd</sup>	4	0.6±0.1	3.8 (76)	2.9±0.11
3 <sup>rd</sup>	1	1.1±0.0	4.1 (82)	3.2±0.0

Table 4. 10 The effect of age, body condition and ejaculate frequency on Volume (ml) (LSM±SE), Motility (%) and Concentration (LSM±SE) of Semen collected from Abergelle Bucks kept at on farm condition

Variables	N	Volume (ml) (LSM±SE)	Motility (%)	Concentration (LSM±SE)
Over all	33	0.65±0.04	3.9 (78)	3.1±0.14
CV		41	14	27
Body condition score		NS	NS	NS
3	9	0.69±0.1	4.2 (84)	3.2±0.19
3.5	24	0.68±0.05	3.9 (78)	3.0±0.18
Age/ dentition		NS	NS	NS
3 p	8	0.78±0.07	4.2 (84)	2.9±0.18
4 p	25	0.59±0.05	3.9 (78)	3.2±0.18
Frequency of ejaculate		NS	NS	NS
1 <sup>st</sup>	18	0.75±0.16	3.9 (78)	3.1±0.22
2 <sup>nd</sup>	11	0.64±0.13	4 (80)	2.9±0.2
3 <sup>rd</sup>	4	0.67±0.07	4.3 (86)	3.2±0.3

#### 4.4 Socio economic characteristics and Goat breeding management practices of respondents

##### 4.4.1 Sex, Age and Educational status of respondents

The survey result revealed that relatively better proportion of literacy rate at the study area (44.5%) was found (Table 4.11) which was considered as an opportunity for easily transferring the livestock technologies at the area. Relatively better literacy rate at the study site might be an indicator for voluntary participation of these farmers in the community based goat genetic improvement program for the last five years and even there active involvement and courage for the goat estrus synchronization and AI programs. The result reported by (Asaminew Tassew and Eyasu Seifu, 2009) also confirmed the negative contribution of illiteracy on easy of dairy technology transfer in and around Bahir Dar.

Table 4. 11 Sex, Age and Educational status of respondents

Variables	Participant		Non participants	
	N	Percentage	N	Percentage
Sex				
Male	27	93.1	16	94.1
Female	2	6.8	1	5.9
Age				
< 24 years	1	3.4	2	11.7
24-45 years	14	48.2	7	41.1
46-64 years	8	27.5	4	23.5
>64 years	4	13.7	4	23.5
Educational level				
Illiterate	16	55.1	10	58.8
Read and write	8	27.5	4	23.5
Primary school	2	6.8	3	17.6
≥Secondary school	1	3.4	0	0.0
Religious leader	2	6.8	0	0.0

#### 4.4.2 Income source and livestock holding of the respondents

The higher proportion of income source from livestock rearing (58.6%) for the participant and (52.9%) for the non participant farmers (Fig 4.3) was comparable with the result (53%) reported by (Alubel Alemu, 2015) and slightly differed from (70%) contribution reported by (Bekahegn Wondim *et al.*, 2018) at the same study site. Sample size could be an important reason for the difference.

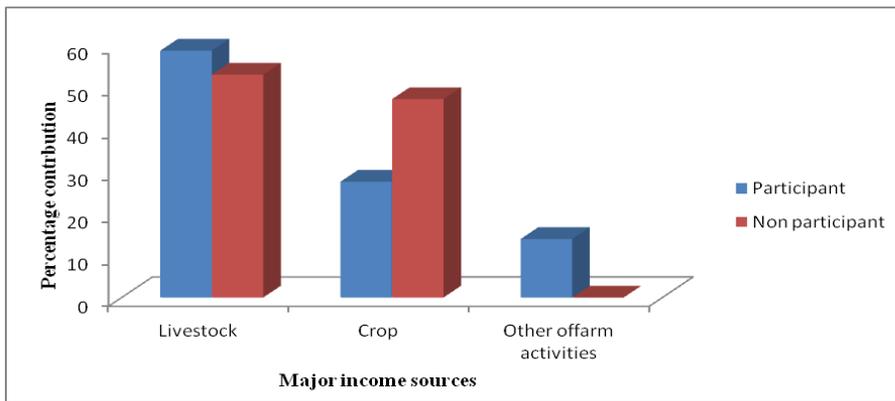


Figure4.3 Major Income sources with their percentage contribution at the study area

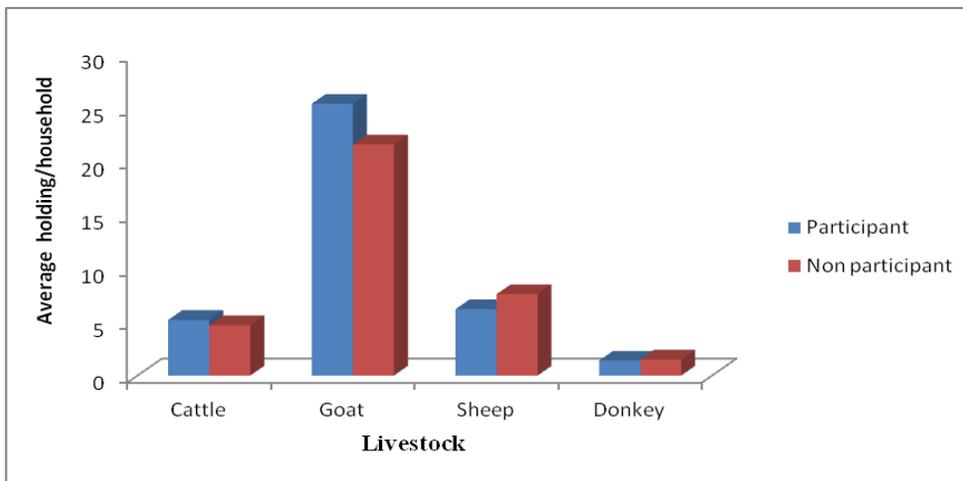


Figure 4.4 Average livestock holding per household at the study area

#### 4.4.3 Goat breeding objective and reasons of keeping goat

As indicated with the indices for each reasons of goat production (Table 4.12), cash income was ranked first by most of the respondents which might be due to the reason that small ruminants are the liquid and immediate source of income for the small holder farmers. The result of the current study is in line with different studies conducted in Ethiopia concerning with goat production objectives. For instance (Alubel Alemu, 2015) on Abergelle and central highland goat, (Mahilet Dawit, 2012) on Hararghe Highland goat and (Ahmed Seid, 2013) on Ethiopian indigenous goats in Horro Guduru Wollega zone and (Solomon Abegaz, 2013) on Abergelle and Western Lowland goat breeds, reported that cash income was the first rank among different goat production objectives.

Table 4. 12 Preference indexes for different goat production and management parameters

	<b>Rank</b>					Index
	1 <sup>st</sup>	2nd	3rd	4 <sup>th</sup>	5th	
<b>Reason for keeping goats</b>						
Income source/cash	22	20	14	4	0	0.35
Meat and milk	8	11	9	25	0	0.23
Manure	0	0	3	11	23	0.08
Socio cultural value	0	1	10	5	16	0.09
Saving	16	14	10	1	7	0.25
<b>Criteria for choice of breeding stock</b>						
Body size	18	18	8	2	0	0.25
Color	8	14	18	6	0	0.21
Horn orientation	0	0	3	15	28	0.09
Pedigree profile	12	16	11	0	7	0.22
Disease resistance	16	12	12	5	1	0.23

#### 4.4.4 Season of birth and birth type at the study area

From the current result (78.5%) of the does were gave birth from October up to January (Fig 4.5) which indicates that Abergelle goat breed is almost seasonal breeder. This seasonality of

breeding might be associated with feed resource availability as tropical breeds were not follow seasonal pattern of breeding with normal breeding situation (Philippe *et al.*, 2004). So that the information is vital for breeding management like exchange of bucks during the peak mating seasons from May to July, avoid inbreeding problem and culling of unwanted bucks especially for the community based genetic improvement program.

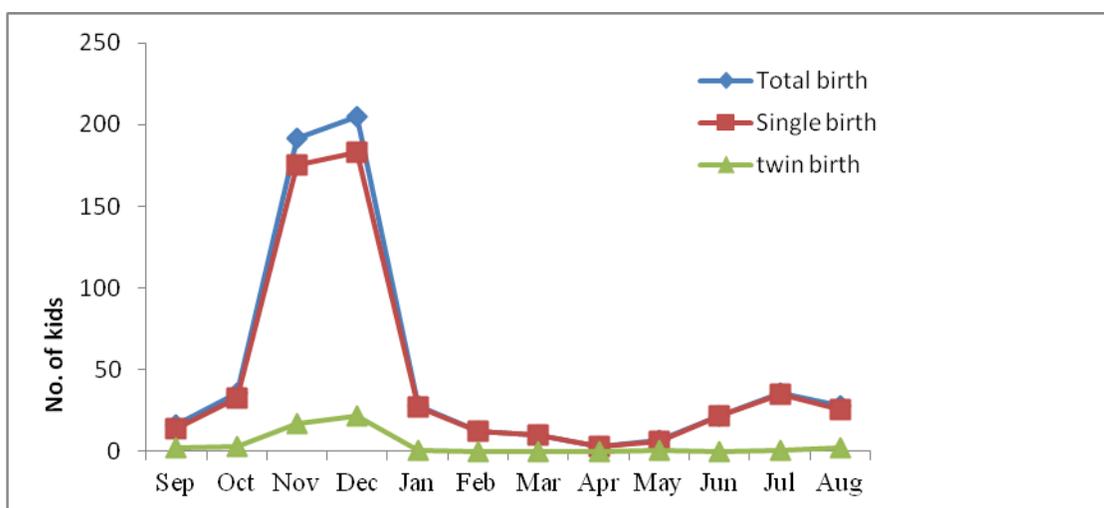


Figure 4.5 Season of birth and birth type of Abergelle Does

#### 4.4.5 Breeding management practices

The higher proportion (89%) of goat keepers used controlled mating system and 84.7% of the farmers had culling practice (Table 4.13). The reason for easily adoption of controlled mating and culling practice was due to the farmers involvement in the community based breeding program which enable them to use the selected buck only in accordance with the designed buck usage modality implemented in the CBBP. The higher intensity of culling for male kids (45.6%) and yearling buck (30.4) were for the reason that within the CBBP, selection has been practiced for sires only so that during selection time more number of unselected male kids and yearling bucks can be culled out.

Table 4. 13 Different goat breeding and management practices of respondents

Management practices	N	Percent
Type of mating used		
Un controlled	0	0
Hand mating	5	11
Group mating	41	89
AI	0	0
Culling practice		
Yes	39	84.7
No	7	15.3
Age category under culling		
Kid	5	11
Weaned male	21	45.6
Weaned female	3	6.5
Yearling bucks	14	30.4
Does	3	6.5
Flock size trend for the last three years		
Increasing	7	15.2
Decreasing	26	56.5
Stable	11	23.9
Unknown	2	4.3

#### 4.4.6 Farmers perception on estrus synchronization and AI technologies

Estrus synchronization for goats, use of single injection of PGF2 alpha and double injection of PGF2 alpha protocols were preferred by goat keepers with almost very good rank. The reason for better preference of these protocols by goat keepers might be associated with less or no stress detected during hormone application process and better conception rate recorded. However, PMSG protocol for synchronization of goats and artificial insemination technologies were less preferred by goat keepers with good rank. Sort of stress detected from the Does during sponge insertion and removal might be the possible reason that goat keepers were less preferred the protocol. Moreover, the reason for low rank given for AI might be due to the reason that farmers understand as all of the inseminated Does must be conceive though the conception rate from AI was higher than the control group as presented in table 4.14.

Table 4. 14 Likert score of respondents for different reproductive technologies

<b>Variables</b>	<b>Some score</b>	<b>Likert score</b>
Estrus synchronization	101	3.5
PGF2alpha (single)	47	3.4
PGF2alpha (double)	59	3.9
PMSG	45	3.0
AI	76	2.7

## **Chapter 5: CONCLUSION AND RECOMMENDATIONS**

### **5.1 Conclusion**

The result of the present study confirmed that in situations where there are enough feed supplementation programs, estrus follow up, pregnancy diagnosis prior to administration of hormones and overall improved animal husbandry practices, the value of estrus synchronization following artificial insemination is an efficient technology in improving the reproductive performances of goats. With this regard, Abergelle does were responsive (67.6 %) for both PGF2 $\alpha$  and PMSG based protocols during the main breeding season. From the protocols compared, PMSG +Enzaprost<sup>®</sup> were best (87%) for induction of estrus. It could be concluded from the result that higher proportion of PMSG treated does had shorter estrus duration (36 up to 48h) as compared to does treated with PGF2 $\alpha$ . Whereas most of PGF2 treated does had longer estrus duration (48 -60 hrs). Taking this into account, fixed time AI at 48 hrs resulted a better conception rate in PGF2 (78.8% for double and 64.7% for single injection) as compared to PMSG (57.6%) and control group (52.6%). Fixed effects of parity of doe and body condition score were important factors affecting estrus response of Abergelle does. Furthermore, estrus was pronounced in all the protocols that estrus signs of restlessness, mounting to each other, frequent bleating, swollen and red vulva, mucus discharge from the vulva, seeking for bucks and standing to be mounted were easily and reliably identified.

All of the hormonal protocols showed a better conception rate (65.4%) in comparison with the control group (52.6%). Despite the lower conception rate, twinning rate was better in the case of PMSG+Enzaprost<sup>®</sup> treated does as compared to other protocols and the control.

Semen parameters/characteristics of Abergelle bucks were found to be under normal range with no variation observed by management condition and ejaculate frequency. However, fixed effects of age and body condition had an effect on semen mass motility. There was no variation observed between the on-station and on-farm management conditions for all the parameters evaluated in this research which calls for improving the feeding and other management condition at the station. From different criterions evaluated, farmers had showed their preference for estrus synchronization by using prostaglandin injections for their goats.

## 5.2 Recommendations

Based on the above results, the following recommendations were forwarded.

PMSG+Enzaprost<sup>®</sup> protocol was found to be best in terms of induction of estrus, however the insemination time should be from 36h up to 48h for better conception efficiency

For ease of application, farmers preference and better conception rate, PGF2 $\alpha$  (double injection) was found to be best protocol followed by PGF2 $\alpha$  (single injection) for abergelle does during the main breeding season together with strengthened pregnancy diagnosis activity before hormonal administration as PGF2 $\alpha$  can lead for serious abortion

Prior to a synchronization program, appropriate animal selection for instance early to mid parity does (1<sup>st</sup>, 2<sup>nd</sup> and 3<sup>rd</sup>) with body condition score of 2.5 and 3 can best respond to the hormonal treatments for Abergelle goat.

For areas where the breeding objective of keepers targets for multiple births in goats, using PMSG based protocol <sup>for</sup> synchronization will be important since this study resulted with maximum twinning and triplet birth with this protocol which was apart from the prostaglandin and the control groups.

The implications of differences in age, body condition score and scrotal circumferences among the bucks in the fertility results after AI is yet unknown and needs to be further studied

Finally, strengthened further study on the investigation of different estrus synchronization protocols for supper ovulatory response in goats together with cost benefit/feasibility studies and serum analysis for further precision of estrus detection concepts should become an important further research area for Ethiopian local goats.

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## **APPENDICES**

Table 1. ANOVA for the effect of management condition, hormone, bcs, parity and age on estrus duration and onset of estrus for Abergelle Does

Dependant variables	Source of variations	DF	Mean Square	Pr > F
Onset of estrus	Dentition	3	14.398877	0.6647
	Parity	5	9.557148	0.8821
	Bcs	2	177.131697	0.0020
	Hormone	2	883.274251	<0.0001
	Management condition	1	331.127166	<0.0006
Duration of estrus	Dentition	3	61.786739	0.5736
	Parity	5	355.599057	0.0026
	Bcs	2	24.720584	0.7662
	Hormone	2	153.498334	0.1940
	Management condition	1	216.444514	0.1284

Table 2. ANOVA for the effect of hormone on the onset of estrus and estrus duration for Abergelle does managed at on station and on farm conditions

Management condition	Dependant variables	Source of variations	DF	Mean Square	Pr > F
On-station	Onset of estrus	Hormone	2	34.29815288	0.0473
	Duration of estrus	Hormone	2	56.3407786	0.5997
On-farm	Onset of estrus	Hormone	2	1152.168643	<0.0001
	Duration of estrus	Hormone	2	139.2183698	0.2600

Table 3 . ANOVA for the effect of hormone, management condition and parity on litter size and birth weight on Abergelle Does

Dependant variables	Source of variation	Df	Mean square	Pr > F
Litter size	Management condition	1	0.1441834	0.7810
	Hormone	3	3.76664408	<0.0001
	Parity	5	0.12229643	0.6559
Birth weight	Management condition	1	2.20198034	<0.0001
	Hormone	3	0.74783422	0.0011
	Parity	5	0.11620869	0.4973

Table 4 . ANOVA for the effect of hormone on litter size and birth weight on Abergelle Does at on- station and on-farm condition

Management condition	Dependant variables	Source of variations	DF	Mean Square	Pr > F
On-station	Litter size	Hormone	3	0.83636364	0.0040
	Birth weight	Hormone	3	0.48707648	0.685
On-farm	Litter size	Hormone	3	3.39109350	<0.0001
	Birth weight	Hormone	3	0.54455590	0.0070

Table 5 . ANOVA for the effect of management condition and ejaculate frequency on semen characteristics of abergelle bucks

Dependant variables	Source of variations	DF	Mean Square	Pr > F
Volume	Management condition	1	0.01211392	0.6847
	Ejaculate frequency	2	0.2407475	0.7190
Motility	Management condition	1	1.62911244	0.0524
	Ejaculate frequency	2	0.59237835	0.2455
Concentration	Management condition	1	0.02350162	0.8489
	Ejaculate frequency	2	0.37019052	0.5649

Table 6. ANOVA for the effect of management condition on semen characteristics of Abergelle bucks

Dependant variables	Source of variations	DF	Mean Square	Pr > F
Volume	Management condition	1	0.01522833	0.6434
Motility	Management condition	1	1.56875546	0.0590
Concentration	Management condition	1	0.01069643	0.8966

Table 7 . ANOVA for the effect of age, Bcs and ejaculate frequency on volume, motility and concentration of Abergelle bucks at station and on- farm management condition

Dependant variables	Source of variations	DF	Mean Square	Pr > F
Volume (station)	Age	1	0.00534759	0.7451
	Bcs	1	0.03898396	0.3961
	Ejaculate frequency	2	0.11715944	0.1693
Motility (station)	Age	1	0.088644719	0.1715
	Bcs	1	3.44326537	0.0255
	Ejaculate frequency	2	0.67608843	0.2378
Concentration(station)	Age	1	0.13854599	0.6479
	Bcs	1	0.01527326	0.8783
	Ejaculate frequency	2	0.27095170	0.6552
Volume (on-farm)	Age	1	0.21269351	0.1017
	Bcs	1	0.00029512	0.9502
	Ejaculate frequency	2	0.03917188	0.5959
Motility (on-farm)	Age	1	0.43103234	0.2475
	Bcs	1	0.56978048	0.1853
	Ejaculate frequency	2	0.24363659	0.4644
Concentration (on-farm)	Age	1	0.37847995	0.4840
	Bcs	1	0.24770089	0.5706
	Ejaculate frequency	2	0.18296776	0.7857

Table 8 . Chi-square value and “p” value of factors for estrus response

<b>Dependant variable</b>	<b>Source of variation</b>	<b>Df</b>	<b>Chi sq</b>	<b>Pr &gt; ChiSq</b>
<b>Estrus response</b>	Management condition	1	0.02	0.8777
	Hormone	2	35.97	<0.0001
	Parity	5	5.52	0.3557
	Dentition/age	3	2.47	0.4813
	Bcs	2	0.95	0.6206

Table 9 . Chi-square value and “p” value of protocols on estrus response at on-station and on-farm condition

<b>Management condition</b>	<b>Dependant variables</b>	<b>Source of variation</b>	<b>Df</b>	<b>Chi sq</b>	<b>Pr &gt; ChiSq</b>
On-station	Estrus response	Protocol	2	16.79	0.0002
On-farm	Estrus response	Protocol	2	21.48	<0.0001

Table 10 . Chi-square value and “p” value of factors for conception rate and kidding rate

<b>Dependant variables</b>	<b>Source of variation</b>	<b>Df</b>	<b>Chi sq</b>	<b>Pr &gt; ChiSq</b>
Conception rate	Management condition	1	1.17	0.2803
	Protocol	3	11.32	0.0101
	Parity	5	5.78	0.3279
Kidding rate	Management condition	1	1.02	0.3135
	Protocol	3	11.2	0.0107
	Parity	5	3.34	0.6476

Table 11 . Chi-square value and “p” value of factors for conception and kidding rate at on-station and on-farm

<b>Management condition</b>	<b>Dependant variables</b>	<b>Source of variation</b>	<b>Df</b>	<b>Chi sq</b>	<b>Pr &gt; ChiSq</b>
On-station	Conception rate	Protocol	3	5.04	0.1687
	Kidding rate	Protocol	3	2.38	0.4974
On-station	Conception rate	Protocol	3	11.74	0.0083
	Kidding rate	Protocol	3	9.46	0.0237

**LIST OF FIGURES IN THE APPENDIX**



Figure1. Picture for goats used for synchronization at station and during sponge insertion



Figure2.pictures during artificial insemination



Figure 3. Pictures during ultrasound scanning and semen evaluation

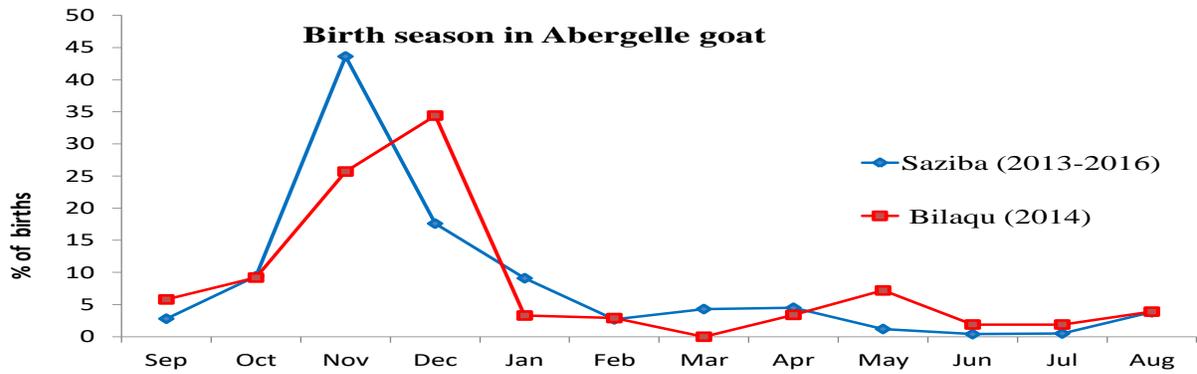


Figure4. pictures during semen collection and processing



Figure5. pictures of synchronized does and born kids at station

**Cont...**



Picture 6: Birth season of Abergelle Goat breed

# **Questionnaire survey for goat reproduction and management**

## **Chapter one: General household information (tick one or more)**

### **1) Interviewee name**

---

Age-----

### **2) Position in the household**

- 1) Household head
- 2) Brother
- 3) Sister
- 4) Son
- 5) Daughter
- 6) Other-----

### **3) Household head**

**Sex**

- 1) Male
- 2) Female

**Age (years)**

---

< 24 years

24-45 years

46-64 years

>64 years

---

5) 61-70

6) > 70

### **4) Educational status**

- 1) Illiterate
- 2) Read and write
- 3) < 6 grade
- 4) 7- 10 grade
- 5) 11-12 grade
- 6) Diploma and above
- 7) Other specify

### **5) Source of income (1= least, 4 =major)**

- 1) Crops-----
- 2) Livestock and products-----
- 3) Home industries-----
- 4) Salary/wedge-----
- 5) Others specify-----

### **6) Livestock kept (put numbers)**

Cattle-----

Sheep-----

Goat-----

Donkey-----

Chicken-----

**Chapter two: Goat productions and management**

**7) Purpose of keeping goats (tick one or more)**

Purpose	1st	2nd	3rd	4th	5th	6th	7th	8th	9th
Meat									
Milk									
Family asset									
Manure									
Skin									
Cash income									
Investment									
Cultural value									
Other									

**Chapter three: Goat breeding and reproduction**

**1) Common breed name (Goat) -----**

**8) Holding by age and sex**

Does	
Weaned	
Bucks	
kids	

**9) Primary reason for keeping goats**

- 1) Income source
- 2) meat and milk
- 3) Family asset
- 4) Socio cultural
- 5) Other specify

**10) criteria for choice of buck(s) for breeding**

- 1) Conformation
- 2) Color
- 3) Horn orientation
- 4) Market Performance
- 5) Pedigree profile
- 6) Other specify

**11) Type of mating used**

- 1) Uncontrolled
- 2) Hand mating
- 3) Group mating
- 4) AI

5) Other specify

12) **Have you a culling practice in your flock?**

- 1) Yes
- 2) No

13) **If yes**

<b>Culling reason</b>	<b>Kids</b>	<b>Weaned male</b>	<b>Weaned female</b>	<b>≥ 1 year bucks</b>	<b>Do es</b>	<b>Others</b>
Conformation						
Color						
Health						
Mating performance						

8) **Source and breed(s) of buck(s) used in the herd**

- 1) Own buck
- 2) Bought
- 3) Donated/selected
- 4) Shared
- 5) Communal buck
- 6) AI
- 7) Other specify

10) **Flock size trend for the last decade**

- 1) Increasing
- 2) Decreasing
- 3) Stable
- 4) Unknown

11) **If increasing what will be the possible reason**-----

-----

12) **If decreasing what will be the possible reason**-----

-----

13) **At which month does your goat give birth for the last five years?**

<b>1</b>	<b>Month of birth</b>	<b>No.s</b>
2		
3		
4		
5		
6		
7		
8		
9		
10		

14) **Is there a change in birth season across years to now?**

- 1) Yes
- 2) No

15) If yes what will be the possible reason for the change?

-----  
-----

16) How do you rate fertility rate of your flock for the last decade

- 1) Increasing
- 2) Decreasing
- 3) Stable
- 4) Not known

17) If increasing, what will be the possible reason

-----  
-----  
-----

18) If Decreasing, what will be the possible reason

-----  
-----  
-----

19) Fertility profile of your flock for the last decade?

Litter size	No. of Does
Single	
Twine	
Triple	

20) How do you express the trend of twining and above birth to now?

- 1) Increasing
- 2) Decreasing
- 3) Stable
- 4) Unknown

21) If increasing, what will be the possible reason

-----  
-----  
-----

22) If Decreasing, what will be the possible reason

-----  
-----  
-----

23) Do you know the use of hormones? which hormone?

-----

24) Have you ever used hormones for synchronizing your goats?

- 1) Yes
- 2) No

25) If yes, how do you express the result?

- 1) Excellent
- 2) V. good
- 3) Good
- 4) Unsatisfactory
- 5) No idea

26) Which improvement was achieved by using hormones in your flock?

-----  
-----  
-----  
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**27) Which hormone was responsible for the above improvement in your flock?**

- 1) PMSG (give score)
- 2) Single injection of Prostaglandin (give score)
- 3) Double injection of Prostaglandin (give score)

**28) Have you ever used AI. for your goats?**

- 1) Yes
- 2) No

**29) If yes, how do you express the result?**

- 1) Excellent
- 2) V. good
- 3) Good
- 4) Unsatisfactory
- 5) No idea

**30) Which improvement was achieved by using AI in your flock?**

-----  
-----

**31) Have you got a problem to use Reproductive biotechnology tools for your goats?**

- 1) Yes
- 2) No

**32) If yes which problem?**

- 1) Access for the technologies
- 2) Skilled technician
- 3) In compatibility with our breeding practice 4) Others specif

## **BIOGRAPHICAL SKETCH**

The author of this thesis, Bekahgn Wondim Alene, was born in 1991 G.C in Dembecha district of west Gojjam Administrative Zone of Amhara Regional State. He had attended and completed Elementary, Secondary and High School education in Dembecha town. After joining Bahir Dar University in 2010, he had completed his first degree in Animal Production and Technology in 2012 G.C. As soon as graduation, he had been serving the district Agriculture Office of Dembecha for one year and four months and then employed by Sekota Dry Land Agriculture Research center, (SDARC) under Amhara Region Agricultural Research Institute (ARARI), as a junior researcher in goat research division. From the time of employment at ARARI onwards, he had served for more than three years till he joined the School of Graduate Studies of the Bahir Dar University in September 2017. After completing the course work, he carried out his MSc thesis research on “Estrus Synchronization and Fixed Time Artificial Insemination Technologies in Abergelle Goat at Station and Community-Based Breeding Village Conditions of Wagemira, Ethiopia” for partial fulfillment of the requirements for the degree of Master of Science in Agriculture majoring in Animal Genetics and Breeding.