

SYNCHRONIZATION OF THE ESTROUS CYCLE IN  
THE FEMALE OVINE USING NORGESTOMET

By

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THE FEMALE OVINE USING NORGESTOMET

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## Chapter I

### INTRODUCTION

Artificial insemination (AI) is a technology widely used in the beef, dairy, swine and poultry industries. Use of this technology has allowed for greater efficiency in genetic gain within each of these industries. In the sheep industry AI of ewes has been utilized, however, wide variability in conception and fertility rates have been problematic and hindered its acceptance in the sheep industry. Laparoscopic AI (LAI) (Killeen and Caffery, 1982) and transcervical AI (TAI) (Andersen et al., 1973; Fukui and Roberts, 1978) are two artificial insemination techniques that have been utilized with some success in sheep. Laparoscopic AI has been effective, however, cost and required surgical intervention for LAI has slowed reception for use by commercial sheep producers. Transcervical AI could potentially be the most convenient practice for commercial producers to use (Halbert et al., 1990), but ease of the procedure using the recommended equipment and inconsistencies in fertility rates must first be revised

before it will become widely practiced.

In an overview by Salamon and Maxwell (1995) causes related to reduced fertility rates in ewes with both of these AI practices were ram semen preservation, ram variation, ewe body condition, environment, breeding season, insemination method, depth of insemination, inseminator experience, timing of insemination and synchronization technique. The present study is focused on the effect of synchronization techniques utilizing progestagen implants on timing of onset of estrus and ovulation in ewes.

Two studies were conducted to determine the effectiveness of norgestomet as a synchronization method in ewes. The following review will discuss the factors involved in accomplishing successful estrous response and conception rate in ewes when treated with various techniques for synchronization.

## CHAPTER II

### REVIEW OF LITERATURE

#### Estrous Cycle

The length of the estrous cycle in the ewe is approximately 17 days (Cupps, 1991). The ewe's estrous cycle can be divided into two observable phases; a follicular phase lasting 2-3 days in which ovulatory follicles grow and produce ova and a luteal phase dominated by the presence of one or more corpora lutea (CL) on the ovary. When estrus occurs during the follicular phase, the ewe accepts mounting from rams. The duration of estrus expression in the ewe is between 24 and 36 hours. Expression of estrus is highly variable, being shorter in younger females than in older and longer in the mid-breeding season than at the beginning or the end of the breeding season (McKenzie and Terrill, 1937; Parsons and Hunter, 1967).

The ewe is usually considered to be a spontaneous ovulator, however, researchers have concluded that the ewe is a semi-induced ovulator. This was observed when the introduction of a ram to anovular ewes caused ovulation six

days later (Pearce and Oldham, 1984).

The relationship between the beginning and the end of estrus and the exact time of ovulation is not entirely consistent. McKenzie and Terrill (1937) ovariectomized ewes at varying times after estrus detection and found that most ewes ovulate towards the end of estrus, but at a variable time from the beginning of estrus. Because of this inconsistency it is difficult to accurately predict when the ewe will ovulate.

Measurement of endocrine function can be a good indicator of which phase or stage of the cycle the ewe is in during the estrous cycle and when ovulation will occur. Gonadotropin Releasing Hormone (GnRH) is the primary neural control of the estrous cycle in the ewe. GnRH is released from the hypothalamus in a pulsatile manner (Halasz and Pupp, 1965). By action on the anterior pituitary gland, this neuropeptide stimulates the release of the gonadotropins, luteinizing hormone (LH) and follicle stimulating hormone (FSH). In turn, the release of GnRH, LH and FSH are affected by endocrine feedback from the ovaries. Estradiol, progesterone and inhibin regulate gonadotropin release by actions on the hypothalamus and pituitary gland (Clarke, 1984). External stimuli, such as light, also alters the reproductive state by changing GnRH secretion.

The luteal phase of the estrous cycle is controlled by CL present on the ovary. These CL release progesterone into the blood inhibiting LH secretion via negative feedback on the hypothalamic-pituitary axis. Pulsatile release of LH is generated at a rate of once every 3-10 h during the luteal phase of the estrous cycle (Baird et al., 1976).

Prostaglandin  $F_{2\alpha}$  ( $PGF_{2\alpha}$ ) is released by the endometrium in response to an interaction between ovarian oxytocin and estradiol. Luteolytic factor,  $PGF_{2\alpha}$ , is transported via the venous and lymphatic drainage where it is transferred into the ovarian artery through a countercurrent mechanism. Exogenous prostaglandins are highly effective as luteolytic agents causing CL regression (Cupps, 1991). As the CL regress and concentrations of progesterone in plasma decline, pulses of LH increase to about 1 pulse per hour (Baird, 1978). There is a gradual increase in pulse frequency throughout the follicular phase until estrus (Baird, 1978). There is also a gradual increase in the secretion of estradiol from the ovarian graafian follicles (Baird et al., 1981; Karsch et al., 1979) which triggers two central events, estrus and the LH surge. Ovulation occurs about 24h following the LH surge (Cumming et al., 1973) ending the follicular phase and marking the beginning the luteal phase (Figure 1).

Follicle stimulating hormone levels remain at a basal

level throughout the luteal phase (Pant et al., 1977). During the follicular phase plasma FSH levels decline gradually. At the time of the pre-ovulatory LH surge there is a concomitant surge release of FSH followed by a second FSH rise which occurs 18-24h after the LH surge (Salamonsen et al., 1973). Cummins et al. (1983) have demonstrated that FSH secretion is regulated by follicular secretion of inhibin.

#### Follicular Dynamics

The ewe, unlike the cow and mare, is a polyovulator as multiple CL may be found on the ovary during the luteal phase. Because of this characteristic, multiple ovulations or increasing ovulation rate is key in increasing the prolificacy in the ewe.

Folliculogenesis in the sheep takes about 6 months to recruit an oocyte from its primordial population to a final population of ovulating follicles, in which most of the follicle growth phase is spent in the preantral stages (Turnbull et al., 1977; Cahill & Mauleon, 1980). Once the antrum is formed around the oocyte, only 15 days are required to reach pre-ovulatory size. Most evidence suggests that the pre-ovulatory follicle emerges from the pool of small antral follicles (< 4 mm diameter) some time after onset of luteal regression (Smeaton & Robertson,

1971). This implies that some of the antral follicles < 4 mm diameter can be mobilized for rapid development. McNatty et al. (1982) indicated that after induced luteolysis on day 9 or 10 of the estrous cycle, estrogenic follicles  $\geq$  5 mm were absent 3 to 6 hours later but were present at 10 hours. This supported the idea that pre-ovulatory follicles emerge from the pool of small antral follicles. Using daily transrectal ultrasonography, Schrick et al. (1993), observed that an average of 11 follicles reached  $\geq$  4 mm in diameter and five different follicles became the largest at various stages during an estrous cycle. This information indicates that follicular development is continuous throughout the estrous cycle, rather than cyclic as it has been observed in the cow (Pierson & Ginter, 1988). With these factors in mind, recruitment of ovulatory follicles may be possible at any stage of the estrous cycle in the ewe.

#### Seasonal Effects

Ewes have a definite period when they become both anovulatory and enter into anestrus where they are completely unresponsive to sexual advances of rams (Hafez, 1952). However, Cahill and Mauleon (1980) indicated that follicular activity on the ovary did not differ between, during and out of the breeding season in the ewe. These authors found no differences in follicular growth rates,



total numbers of follicles, number of follicles per class or mitotic activity. However, there were significantly more follicles with an antrum present during the breeding season and a lack of estrogenic or dominant follicles out of the breeding season.

The regulation of ovarian activity that controls the seasonal breeding of the ewe revolves around the control of the LH pulse generator (Karsch, 1984). The activity of the LH pulse generator is dependent upon a variety of signals arising from both external and internal sources. The main internal source that regulates the LH pulse generator is that of estradiol's inhibitory action on gonadotropin secretion at the hypothalamic-pituitary axis. The main external source that effects the seasonality of the ewe is photoperiod. The photoneuroendocrine pathway of Karsch et al. (1984a) proposes a model for which this pathway controls the pulse generator of LH (Figure 2). In the model, light cues are picked up by photoreceptors located in the eyes and relayed over a monosynaptic nerve tract to the suprachiasmatic nuclei of the hypothalamus. After receiving input from the circadian system, the photoperiodic message is transmitted to the pineal gland. The pineal converts this neural input into a hormonal signal which takes the form of a circadian rhythm of melatonin secretion. The duration of elevated melatonin secretion, which is directly proportional

to the length of night, is interpreted as either inductive or suppressive. Inductive melatonin signals, associated with a decrease in daylength, stimulate the pulse generator and render it resistant to the frequency slowing action of estradiol; suppressive melatonin signals, associated with an increase in daylength, inhibit the pulse generator and sensitize it to inhibition by estradiol. The melatonin pattern itself determines the reproductive response, rather than merely permitting daylength to be measured by some other neural timekeeping device. Therefore, the photo-sexual response is independent of daylength (Bittman and Karsch, 1984).

The effect of photoperiod (short days) combined with the male effect and possibly improved nutrition all render the hypothalamic-pituitary axis less sensitive to estradiol and allow it to increase the frequency of tonic release of LH (Karsch et al., 1984).

#### Effect of Nutrition

The composition and amount of feed eaten by sheep before and around ovulation can influence the number of follicles that ovulate. The concept of "flushing" ewes or giving them a "rising plane of nutrition" has been known and used for increasing twinning in sheep (from Cupps, 1991). There is also a clear relationship between live weight or

body condition and ovulation rate (Morley, 1978), but the availability of feed just a few days before ovulation can also improve ovulation rate in ewes as much as 30% (Oldham and Lindsay, 1984).

### Estrous Manipulation

Regulation of the estrous cycle or synchronization of the estrous cycle in sheep can be accomplished by either luteolysis or by sustaining the concentration of progesterone in the blood.

Luteolysis is accomplished by treating the ewe with either  $\text{PGF}_{2\alpha}$  or its analogues which causes regression of the CL, thus causing initiation of the follicular phase. Estrous synchronization of sheep during the breeding season has been accomplished through administration of two doses of  $\text{PGF}_{2\alpha}$  9 to 11 days apart (Hackett and Robertson, 1980; Hackett et al., 1981; Henderson et al., 1984; Beck et al. 1987).

Sustaining the concentrations of progesterone in the blood is accomplished either by feeding a progestagen orally in the form of Melengestrol Acetate (MGA) for 10 days (Jabbar et al., 1994) or by inserting sponge pessaries containing medroxy-progesterone acetate (MAP) or controlled internal drug release devices (CIDRs) containing progestagens into the vagina or by placing implants

containing norgestomet subcutaneously in the ear of the ewe for nine to twelve days (Boland et al., 1978; Quirke, 1979; Hackett et al., 1981; Spitzer and Carpenter, 1981; Acritopolous-Fourcroy et al. 1982; Henderson et al., 1984; Fitch et al., 1986; Maxwell and Barnes, 1986; Beck et al. 1987). All of these progestagen treatments have been practiced using them in combination with follicular stimulants such as Pregnant Mare Serum Gonadotropin (PMSG), and have resulted in adequate fertility rates (55 to 100%) and estrus response rates (87 to 100%). Spitzer and Carpenter (1981) found that 83% of ewes had onset of estrus by 72h after norgestomet implant removal. Likewise, Maxwell and Barnes (1986) found that using intravaginal sponges and CIDRs accomplished similar results as 96% of the ewes exhibited estrus by 72h after removal. In both studies, a few ewes initiated estrus by 24h and a majority of ewes (>50%) had initiated estrus by 48h. Studies by Findlater et al. (1991) suggest that using PMSG with progestagens tightens synchrony and advances ovulation by 20 hours. Studies by Quirke et al. (1979) and Walker et al. (1989) which viewed ovaries via laparoscopy found that ovulation commenced within 57h of progestagen removal and continued until 81h when PMSG was utilized in combination with progestagens. However, Walker et al. (1889) found that the time ovulation from progestagen removal is highly variable

within and between flocks.

Many studies have been performed to evaluate the efficacy of both (prostaglandins vs progestagens) estrous synchronization regimes. Henderson et al. (1984) and Beck et al. (1987) compared the two estrous synchronization techniques and found that estrous response (83-98%) and fertility rates (65-75%) were not different among the two methods. However, progestagen synchronization is more efficient when compared to prostaglandins as fewer ewes returned to service (3% vs. 20%). Beck et al. (1993) evaluated the ability to synchronize ewes by combining a single  $\text{PGF}_{2\alpha}$  injection with a five day intravaginal progestagen pessary which resulted in similar estrous synchronization results (100%) and fertility rates (86%). However, during anestrus the use of progestagens in combination with follicular stimulants such as PMSG is superior to prostaglandin treatments in obtaining adequate fertilization rates (58% vs. 5%) as lack of follicular activity during out-of-season breeding hinders the use of prostaglandins for synchronization of the seasonally anestrus ewe (Rawlings et al., 1983).

Woody et al. (1983) studied the effectiveness of norgestomet implants without the use of follicular stimulants in regards to treatment dose, day of cycle in which treatment was initiated and fertility post treatment.

Ewes treated for 13 days with norgestomet implants containing 6 mg norgestomet had a higher conception rate when compared to 3 mg implants (89% vs. 63%). Furthermore, day of cycle in which norgestomet treatment began influenced the estrus response and conception rates in ewes. Ewes in which norgestomet treatment was initiated on day 13 of the estrous cycle had a lowered estrus response (87%) and conception rate (33%) when compared to ewes treated on day 4 of the estrous cycle (100% and 72%, respectively). These results indicate that efficient estrous synchronization and fertility can be accomplished using 6 mg norgestomet ear implants.

#### Artificial Insemination

Manipulation of the estrous cycle has been key in obtaining proper management practices to either synchronize lambing seasons or more commonly used for purposes of artificial insemination (AI). At present there are two basic procedures used to AI ewes with frozen-thawed ram semen: Laparoscopic AI (LAI) and Transcervical AI (TAI). Insemination of ewes via laparoscopy, was first performed by Killeen and Cafferty (1982). Ewes were surgically inseminated directly into the lumen of the uterine horn 48 hours after progestagen pessary removal. This procedure resulted in a 96% fertilization rate (22 of 23 eggs). Since

the introduction of LAI, many studies have been performed to attain similar fertilization rates. In an overview by Salamon and Maxwell (1995), LAI attempts have reported varying fertilization rates (51% to 89%), however, these results have been more consistent than those for TAI procedures (0% to 79%).

Anderson et al. (1973) first proposed and tested non-surgical intrauterine insemination, and the procedure was later modified by Fukui and Roberts (1978). This non-surgical or TAI method was field tested by Halbert et al. (1990) for commercial use by producers. The trial resulted in lambing rates of approximately 55%, and was considered to be consistent with the results shown when using LAI techniques. Results from Windsor et al. (1994) indicated LAI techniques resulted in higher fertilization rates (48% vs 26%) when compared with TAI, however, penetration through the cervix into the uterine body when performing TAI resulted in similar fertilization rates (40% for TAI vs. 42% for LAI). While these results are promising, much research is needed to examine the factors limiting the ability to accomplish higher rates of fertilization when using AI.

Timing of insemination is also a key factor in achieving optimum fertilization rates while utilizing AI. Inseminating the ewes at a time closer to ovulation gives

rise to higher fertility rates. Findlater et al. (1991) suggested that frozen-thawed semen placed in the reproductive tract of the ewe via laparoscopy at either 19h before or 4h after ovulation resulted in pregnancy rates of 50% or greater.

### Ultrasonography

Ultrasonographic techniques have been utilized in the sheep to collect data on follicular and luteal development, uterine motility and pregnancy (Kahn, 1992; Schrick and Inskeep, 1993; Schrick et al., 1993). Pregnancy was accurately determined in ewes using transrectal ultrasonography techniques by day 25 after fertilization (Schrick and Inskeep, 1993). Furthermore, the diameter and number of follicles and CL present on the ovaries of ewes were observed by Schrick et al. (1993). On day 4 or 5 after estrus CL were detectable on the ovary of the ewe as luteal tissue was observed via ultrasonography. Numbers of newly detected follicles  $\geq 2$  mm during the first 16 days of the estrous cycle were highest on day 2, then decreased and remained relatively constant thereafter. Schrick et al. (1993) observed 11-12 different follicles  $\geq 4$  mm which were capable of ovulating if luteal regression had occurred in the ewe.

These techniques required the use of an Aloka 500 with



a 7.5 MHz human prostate transducer (linear array; Corometrics Medical Systems, Inc., Wallingford, CT). During ultrasonography each ewe was placed in dorsal recumbancy in a tilting squeeze chute (Hulet, 1972; Lindahl, 1972). A liberal coating of 3% carboxymethylcellulose, used as coupling medium, was applied to the transducer before insertion into the rectum (Ginter, 1986). The transducer sheathed with a piece of PVC pipe (2 cm X 35 cm) was inserted until the uterine horn(s) were observed. Then the transducer was rotated 90° clockwise and 180° counter-clockwise across the reproductive tract until both ovaries were scanned. Use of this technology gives researchers the ability to accurately record and trace activity on the ovary of the ewe during their estrous cycle. In the present study, these ultrasonographic techniques were used to accurately determine the timing of ovulation during estrus and pregnancy 45 to 60 days post breeding by observing the reproductive tract in the ewe.

#### Radiotelemetric Detection of Estrus

Detection of estrus via radiotelemetry has been greatly studied and utilized in cattle (Dohi et al., 1993; Senger, 1994; Stevenson et al., 1996). It allows around-the-clock monitoring for accurate and precise detection of the onset of estrus when the animal is mounted. A patch holding a

pressure sensitive device adhered to the rump of the female transmits a signal to a main computer program via a fixed radio antenna. The computer program records the identity of the female, time of day and duration of mount when the device is activated. Stevenson et al. (1996) found that the use of this heat detection device increased the efficiency of detecting estrus in estrus-synchronized heifers that had fewer and/or shorter duration of standing events which were missed by visual observation. Not only is the activity of the female measured, but the activity of the male and his movement through the herd might also be monitored with this device.

#### Evaluation of Libido in Rams

Evaluation of sexual behavior in rams to predict their efficacy at breeding has been of particular interests to behaviorists as well as physiologists. Price et al. (1992) determined whether serving capacity or ejaculation rate could be predicted in experienced rams by observing pre-copulatory behaviors when exposed to estrous ewes. They found that the pre-copulatory activities occurred in proportion to ejaculation rate, suggesting that the mating potential of rams can be estimated under conditions that preclude copulation.

The use of serving capacity tests to predict the

success of pen breeding has been evaluated as well (Perkins et al., 1992). In serving capacity tests rams were evaluated by the number of mounts and ejaculations within a given time frame. Rams were scored as high in libido if greater than 10 mounts and(or) six ejaculations occurred within 30 minutes when introduced to estrous ewes. Rams were given a low libido score if little ( $\leq 2$  mounts) or no interest was shown by rams when introduced with estrous ewes. High libido scored rams had greater sexual activity, more live lambs born per ewe and a higher lambing percentage than did low libido scored rams. Thus, using serving capacity tests as a means for evaluating performance in pen breeding as well as determining the productivity of an individual ram can be accomplished.

#### Purpose of Study

Norgestomet is an inexpensive (\$1.00 to \$3.00 per ewe) synchronization method, however, little is know about its effectiveness when utilized without a follicular stimulant. Secondly, little information is available on the effect of norgestomet treatment on the timing of estrus initiation, ovulation and the interval between onset of estrus and ovulation. This information could be very useful for improving the efficacy of AI when utilizing norgestomet as the preferred synchronization method. The use of ultrasound

technology allows for follicles on the ovaries of ewes to be observed through the estrus cycle and ovulation can be observed without causing adverse environmental effects. Furthermore, the HeatWatch system allows for exact detection of estrus initiation in the ewe. With the use of these two tools, an accurate estimation of the timing of estrus initiation, ovulation and the interval between onset of estrus and ovulation can be precisely observed.

The HeatWatch System will also be utilized to evaluate the sexual performance of high and low libido scored rams in a preliminary study.

The objectives of the present study are to evaluate the performance of ewes synchronized with norgestomet and its effect on (a) timing of estrous onset, (b) timing of ovulation, (c) the interval from onset of estrus to ovulation (d) fertility rates and (e) number of lambs born per ewe.

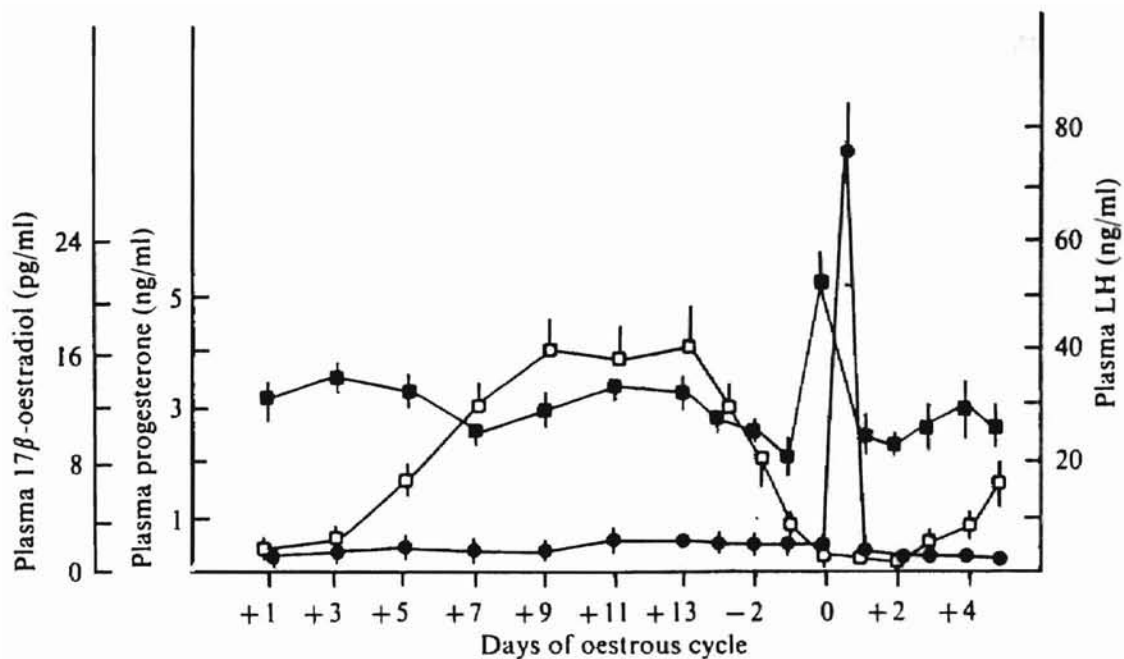


Figure 1. Mean concentration in jugular vein plasma of estradiol-17 $\beta$  (■), progesterone (□) and LH (●) in six ewes during the estrous cycle. Day of estrus is day 0. Each vertical line represents the standard error of the mean (Taken from Pant et al., 1977).

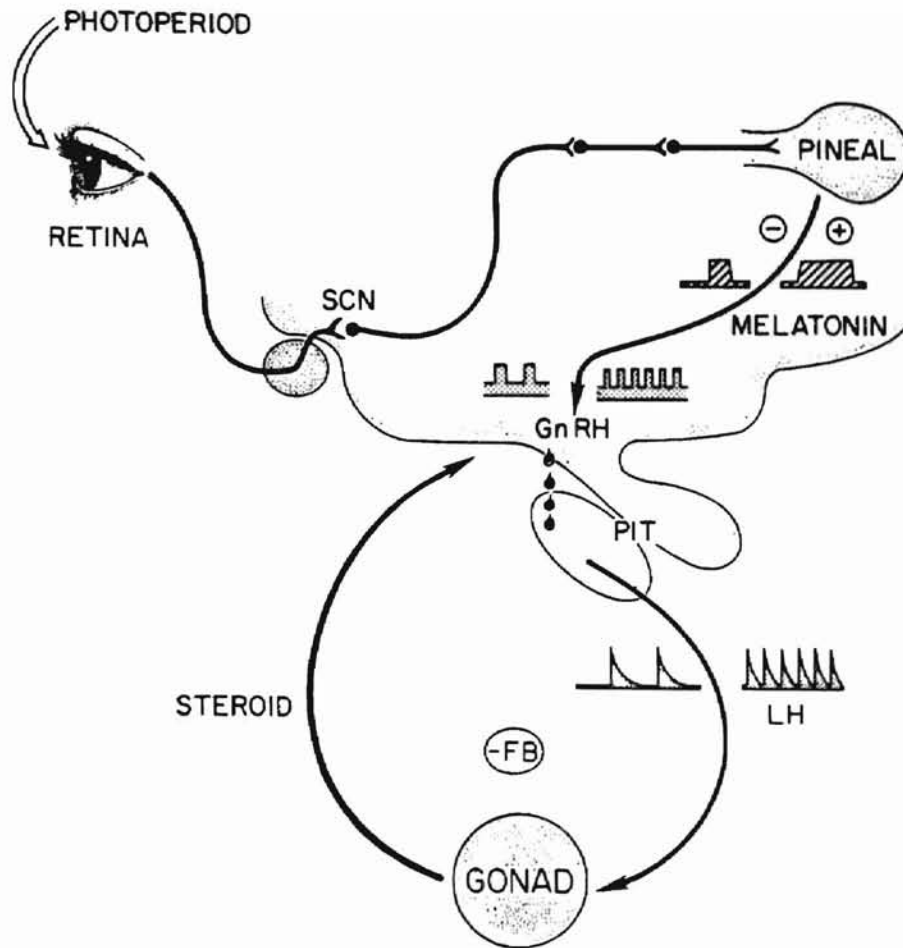


Figure 2. Postulated photoneuroendocrine pathway to the LH pulse generator of the ewe. It is through this pathway that an external signal (light) serves to modulate the capacity of the pulse generating mechanism to respond to an internal signal (steroid negative feedback). This interaction then determines the reproductive state (Taken from Karsch et al., 1984a)

### Chapter III

#### ULTRASONIC EVALUATION FOR THE TIME OF OVULATION IN EWES TREATED WITH NORGESTOMET AND PREGNANT MARE SERUM GONADOTROPIN DURING THE SPRING AND FALL BREEDING SEASONS

##### ABSTRACT

Utilization of progestins and follicular stimulants have provided reasonable techniques for estrous synchronization, but time of ovulation relative to removal of the progestin is not clearly established. Objective of the present study was to monitor follicular development and time of ovulation in ewes during natural and synchronized estrous cycles. Ovaries of sixty Dorset and Rambouillet X Dorset ewes were evaluated during Spring and Fall breeding seasons (30/season). Ewes were randomly assigned to one of three treatment groups (n = 20/group): 1) Control (C) were given a 5 mg i.m. prostaglandin F<sub>2α</sub> injection 9 days apart; 2) implant only (I) received a norgestomet implant for 10 days; and 3) implant + pregnant mare serum gonadotropin (PI) received a norgestomet implant for 10 days with 500 IU i.m.

injection pregnant mare serum gonadotropin (PMSG) at implant removal. The exact time of estrous onset was detected with the HeatWatch Estrus Detection System. Ovaries were monitored via rectal ultrasonography every six hours to determine time interval from implant removal to onset of estrus (EST), implant removal to ovulation (OVUL), and onset of estrus to ovulation (INT). Only 7 out of 20 (35%) C ewes responded to the synchronized estrus, compared to 13 out of 16 (81%) I and 14 out of 16 (88%) PI ewes ( $P < 0.10$ ). Onset of estrus and INT were not effected by breeding season ( $P > 0.10$ ), however OVUL was longer ( $P < 0.10$ ) during Fall breeding (79.3h) compared to Spring breeding (70.8h) season. Onset of estrus to ovulation was reduced ( $P < 0.05$ ) in the C ewes (20.9h) in comparison with I (35.9h) and PI (35.7h) ewes. Treatment mean OVUL was prolonged ( $P < 0.10$ ) in the I group (79.4h) compared to the PI group (70.6h), and mean EST was also lengthened ( $P < 0.10$ ) for I ewes (43.5h) compared to PI ewes (34.9h). Present data indicates that ovulation occurs on average 70 to 80 hours after implant removal in ewes treated with norgestomet, and PMSG advances time to ovulation from implant removal.

Key Words: Estrous Synchronization, Ultrasound, Ewes, Ovulation, Norgestomet.



## INTRODUCTION

In the sheep industry, use of artificial insemination (AI) could greatly enhance the efficiency of genetic gain as has been established in the dairy, beef and swine industries. While AI of ewes has been utilized, wide variability in conception and fertility rates have been problematic and hindered its acceptance in the sheep industry. Laparoscopic AI (LAI) (Killeen and Caffery, 1982) and transcervical AI (TAI) (Andersen et al., 1973; Fukui and Roberts, 1978) are two artificial insemination techniques that have been utilized with some success in ewes. Laparoscopic AI has been effective, however, cost and required surgical intervention for LAI has slowed reception for use by commercial sheep producers. Transcervical AI could potentially be the most convenient practice for commercial producers to use (Halbert et al., 1990), but ease of the procedure using the recommended equipment and inconsistencies in fertility rates must first be revised before it will become widely practiced.

In an overview by Salamon and Maxwell (1995) causes of reduced fertility rates in ewes with AI were ram semen preservation, ram variation, ewe body condition, environment, breeding season, insemination method, depth of insemination, inseminator experience, timing of insemination

and synchronization technique. The present study is focused on the effects of synchronization techniques on the timing of events leading to ovulation during the estrous cycle of the ewe. Utilization of progestins and follicular stimulants have provided reasonable techniques for estrous synchronization (Hulet and Foote, 1967; Quirke, 1979; Beck et al., 1987), but the time of ovulation relative to removal of the progestin is unclear when utilizing different treatment regimes. Breeding of ewes has been accomplished, on a timed insemination basis between 50-65h from removal of the progestagen implant (Eppleston and Maxwell, 1995).

Findlater et al. (1991) suggest that inseminating ewes 19 h before to 4 h after ovulation yielded conception rates of 50% or better when LAI was utilized to inseminate ewes following progestin and PMSG treatment. Therefore, determining time of ovulation relative to either implant removal or onset of estrus may be key in accomplishing optimum fertility rates when inseminating ewes with frozen-thawed ram semen.

The goals of this study were: 1) To observe the time interval from onset of estrus to ovulation and its variability among ewes, 2) To observe the time interval from onset of estrus to ovulation in ewes synchronized with progestagens or a combination of progestagens and follicular stimulants.

## MATERIALS AND METHODS

Sixty, 5 to 7 year old, Dorset and Dorset x Rambouillet cross ewes were synchronized and onset of estrus and ovulation were recorded during 1996 Spring and Fall breeding (n = 30 per season). Ewes were divided into three treatment groups (n = 20/group). Control (C) ewes were treated i.m. with 5 mg of prostaglandin F<sub>2α</sub> (Lutalyse, Upjohn Company, Kalamazoo, MI) 12 and 3 days prior to the beginning (implant removal = day 0) of the trial. An implant only group (I) were implanted with used Syncro-Mate-B (G.D. Searle and Co., Chicago) norgestomet implants for 10 days (Fitch et al., 1986). A third group of ewes (PI) were implanted with used norgestomet implants for 10 days and treated with 500 IU i.m. PMSG (Equinex, Ayerst Laboratories, Montreal, Quebec, Canada) at implant removal.

On the day of implant removal, the ovaries of each ewe were observed transrectally (Schrack et al., 1993) (Figure 1A, B and C), and then at onset of estrus as indicated by the HeatWatch System every six hours using an Aloka 500 with a 7.5 MHz human prostate transducer (linear array, Corometrics Medical Systems, Inc., Wallingford, CT) while in dorsal recumbancy (Figure 2A and 2B). A liberal coating of 3% carboxymethylcellulose, used as coupling medium, was applied to the transducer before insertion into the rectum

(Figure 2C). The transducer sheathed with a piece of PVC pipe (2 cm X 35 cm) was inserted until the uterine horn(s) were observed. Transducer was rotated 90° clockwise and 180° counter-clockwise across the reproductive tract until both ovaries were scanned. Ovaries were evaluated at the onset of estrus, and every six hours until disappearance of the largest follicle(s). Location of ovary, and size and location of the follicle on the ovary were mapped and recorded.

Exact time for initiation of estrous behavior was detected by activation of the HeatWatch system (DDX Inc., Boulder, CO). Mounting by a vasectomized ram was recorded by a radiotelemetric, pressure sensitive device placed in a pocket on the patch which was adhered with K-mar adhesive on the rump of the ewe (Figure 3A and B). This device transmitted a radiotelemetric signal to the main computer program via a fixed radio antenna. The computer program recorded the identity of the ewe, time and duration mounted.

Onset of estrus (EST), ovulation (OVUL) and interval from onset of estrus to ovulation (INT) were examined for effects of season, treatment group and season by treatment interaction (Table 1) by analysis of variance using GLM and least-square means procedures of SAS (1985).

## RESULTS

Only 7 out of 20 (35%) C ewes responded to the PGF<sub>2α</sub> synchronized estrus (Table 2), compared to 13 out of 16 (81%) I and 14 out of 16 (88%) PI ewes ( $P < .10$ ). Only 16 I and 16 PI ewes were analyzed due to norgestomet implant loss in eight of the norgestomet treated ewes. The response to norgestomet treatment is comparable to other reports having accomplished response rates as high as 90 to 100% (Spitzer and Carpenter, 1981; Woody et al., 1983; Fitch et al., 1986). Onset of estrus and time interval from estrus to ovulation were not effected by breeding season ( $P > .10$ ), however, time to ovulation after implant removal was longer ( $P < .10$ ) during Fall breeding (79.3h) compared to Spring breeding (70.8h) in I and PI ewes. The mean estrus to ovulation interval was reduced ( $P < .05$ ) in the C ewes (20.9h) in comparison with I (35.9h) and PI (35.7h) ewes (Table 1). Time of ovulation from implant removal was prolonged ( $P < .10$ ) in the I group (79.4h) compared with the PI group (70.6h), and mean time of estrus onset after implant removal was also longer ( $P < .10$ ) for I ewes (43.5h) compared with PI ewes (34.9h).

Distribution for the occurrence of estrus and ovulation over time from implant removal are displayed in Tables 3 and 4. Onset of estrus had occurred in all ewes by 48h and

ovulation had completed at 84h post-implant removal giving rise to a tighter synchrony when compared to I ewes. A majority of ewes in the study (23 out of 33) had an INT of 36h or less (Table 5).

## DISCUSSION

Detection of estrus via radiotelemetry has been greatly studied and utilized in cattle (Dohi et al., 1993; Senger, 1994; Stevenson et al., 1996). In the present study, the system was utilized to more precisely determine onset of estrus in ewes in order to determine the exact time for onset of estrus relative to implant removal. With the exception of detecting estrus in two ewes (Table 4) the HeatWatch Estrus Detection System proved to be a very useful tool in more precisely determining the time for estrus onset and could be utilized in sheep industry as it is in the cattle industry for AI purposes. Furthermore, it is important to note that the patches adhered to the ewes have much less movement and are more effective when applied on shorn ewes which have thirty days or less wool growth.

The most significant results from this study indicate onset of estrus and ovulation is advanced and tightened when using a progestin in combination with PMSG. Response to norgestomet combined with PMSG in the ewe is similar to that

of progestagen-impregnated pessaries and controlled internal drug release devices (CIDRs) combined with PMSG (Maxwell and Barnes, 1986) as all ewes exhibiting estrus did so by 48 h post- progestagen removal. Walker et al. (1989) indicated that ewes treated with progestagen-impregnated pessaries and PMSG commenced ovulation within 57h and all ewes had ovulated by 81h when ovaries of ewes were observed via laparoscopy. This parallels with the time of ovulation in this study as follicle rupture commenced within 54h and all ewes had ovulated by 82h post-implant removal and PMSG treatment. The INT was similar among I and PI ewes, however, it should be noted that there was a large variation among the ewes as times ranged from 12 to 62h in I ewes and 17 to 53h in PI ewes. A majority of ewes in I (69.2%) and PI (57.1%) treatment groups had an INT of 36h or less. The wide variability of this interval might explain the variation in conception rates reported by other researchers following timed AI (Salamon and Maxwell, 1995).

Eighty-one percent of the I ewes responded to the norgestomet treatment during the spring breeding season. This can be attributed to the use of fall lambing ewes in the study as this particular flock had previously displayed a 70% fall lambing rate. However, the controls response (20%) to  $\text{PGF}_{2\alpha}$  treatment was somewhat less. A 5 mg dose appeared to not be effective in causing luteolysis of the

CL. Reports from Henderson et al. (1984) suggest 10 to 20 mg of PGF<sub>2α</sub> is required for luteal regression in the ewe.

Findlater et al. (1991) suggested that frozen-thawed semen placed in the reproductive tract of the ewe at either 19h before or 4h after ovulation resulted in pregnancy rates of 50% or greater. Present data indicates that ovulation after implant removal occurred on average between 70 and 80 hours. Today, a majority of timed AI is performed 57 to 65 hours after implant removal. Data from the present study indicates that insemination is occurring at a time appropriate for sufficient fertility rates to be obtained.

Control ewes had a tendency to have a shortened INT (range=16-27h) compared to norgestomet treated ewes. This would indicate that insemination of PGF<sub>2α</sub> treated ewes should occur sooner after the detection of estrus. Results from this study suggest 10 to 12 hours after estrous detection would be an appropriate time to obtain optimum fertility rates.

#### IMPLICATIONS

Using norgestomet and a combination of norgestomet and PMSG provides reasonable techniques for synchronizing estrus in the ewe. However, it should be noted how each of these synchronization techniques effect the timing of estrus onset and ovulation when utilizing these techniques for AI



purposes. Artificially inseminating ewes from a basis of estrus onset rather than a timed insemination may give rise to higher, more consistent conception rates.

Figure 1. A) Displays the 7.5 MHz and the bisected 2 cm X 35 cm PVC pipe used as a modified sheath before assembly. B) Displays the assembled sheath and probe. C) Displays the Aloka 500 ultrasound machine and 7.5 MHz probe with sheath as used in the present study after assembling probe.

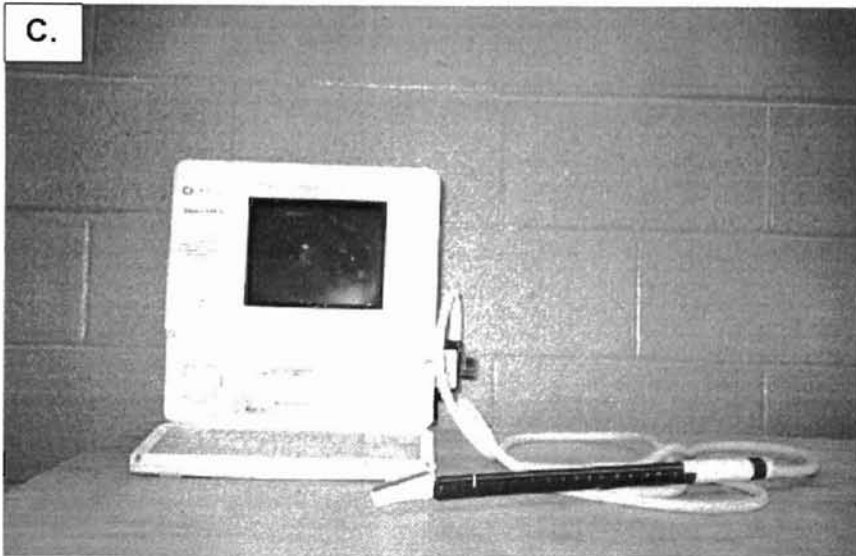
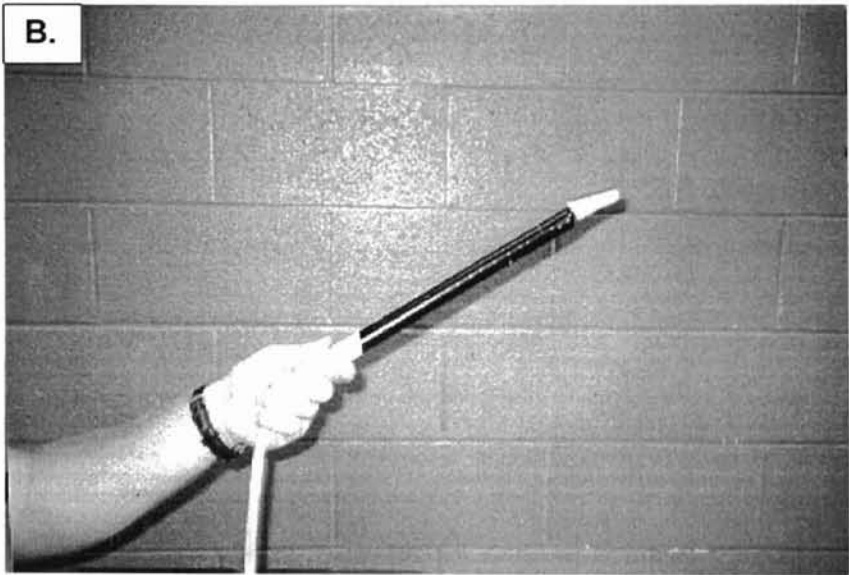
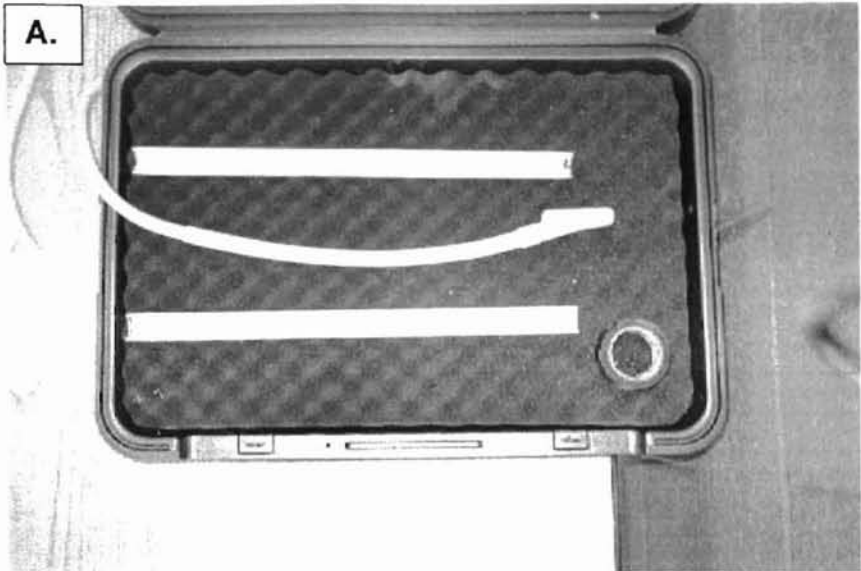


Figure 2. A) Displays the squeeze chute used for maintaining the ewe in dorsal recumbancy. B) Displays the ewe in the chute in dorsal recumbancy. C) Displays the technique used to observe the reproductive tract of the ewe while in dorsal recumbancy. This technique allows for minimal movement of the ewe when used to observe the reproductive tract.

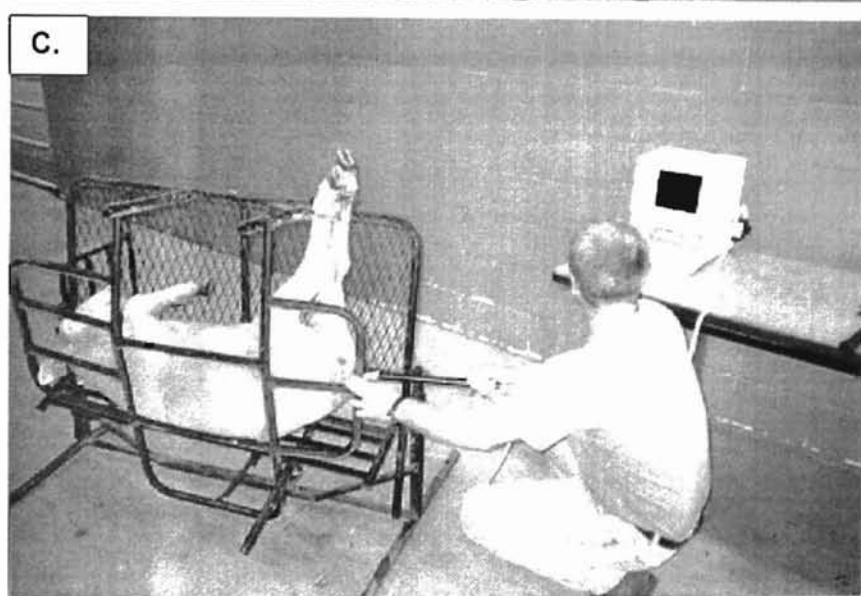


Figure 3. A) Displays the HeatWatch system patch and transducer before inserting the transducer into the pocket on the patch. B) Displays the proper application of the HeatWatch system patch and transducer to the rump area of the ewe.



Table 1. Analysis of variance table illustrating effects of season and treatment and appropriate interactions on onset of estrus (EST), ovulation (OVUL) and the interval from onset of estrus to ovulation (INT).

Source	Degrees of Freedom
Season	1
Treatment	2
Season x Treatment	2
Residual	54



Table 2. Percent response to synchronization and means for EST<sup>a</sup>, OVUL<sup>b</sup> and INT<sup>c</sup> by treatment in mature ewes<sup>d</sup>.

Treatment	No.	% estrus	EST (h)	OVUL (h)	INT (h)
Control	20	35 <sup>e</sup>	N/A	N/A	20.9 <sup>e</sup> 16-27 <sup>g</sup>
Implant only	16	81 <sup>f</sup>	43.5 <sup>e</sup> 26-69 <sup>g</sup>	79.4 <sup>e</sup> 56-100 <sup>g</sup>	35.9 <sup>f</sup> 12-62 <sup>g</sup>
Implant+PMSG	16	88 <sup>f</sup>	34.9 <sup>f</sup> 24-45 <sup>g</sup>	70.6 <sup>f</sup> 54-82 <sup>g</sup>	35.7 <sup>f</sup> 17-53 <sup>g</sup>

<sup>a</sup>EST = implant removal to onset of estrus.

<sup>b</sup>OVUL = implant removal to ovulation.

<sup>c</sup>INT = onset of estrus to ovulation (OVUL-EST=INT).

<sup>d</sup>5 to 7 years in age.

<sup>e,f</sup>LS Means within a column lacking a common superscript letter differ (P < .1).

<sup>g</sup>Ranges of data.

Table 3. Distribution for timing of estrous onset in ewes from implant removal by treatment and season.

Treatment	No.	Hours to estrus from implant removal					
		24	36	48	60	72	84
<b>I</b>							
Fall	7	0	0	3	3	1	0
Spring	6	0	3	1	0	2	0
<b>PI</b>							
Fall	6	0	3	2	0	0	1 <sup>a</sup>
Spring	8	1	5	2	0	0	0

<sup>a</sup>Animal taken out of study because of error in HeatWatch system detection.

Table 4. Distribution for timing of ovulation in ewes from implant removal by treatment and season.

Treatment	No.	Hours to ovulation from implant removal					
		48	60	72	84	96	108
<b>I</b>							
Fall	7	0	0	1	4	1	1
Spring	6	0	1	1	2	2	0
<b>PI</b>							
Fall	6	0	1	3	2	0	0
Spring	8	0	2	5	1	0	0

Table 5. Distribution for interval from onset of estrus to ovulation in ewes by treatment and season.

Treatment	No.	Hours from estrous onset to ovulation					
		12	24	36	48	60	72
C							
Fall	5	1 <sup>a</sup>	4	0	0	0	0
Spring	2	0	0	2	0	0	0
I							
Fall	7	0	2	4	0	0	1
Spring	6	1	0	2	2	1	0
PI							
Fall	6	1 <sup>a</sup>	0	3	1	1	0
Spring	8	0	2	3	2	1	0

<sup>a</sup>Animal taken out of study because of error in HeatWatch system detection

## Chapter IV

### SYNCHRONIZATION OF EWES WITH NORGESTOMET IMPLANTS

#### ABSTRACT

The efficacy of a single norgestomet implant treatment for estrous synchronization of ewes was evaluated during the natural breeding season. Eighty-seven Dorset, Montadale, Hampshire and Suffolk ewes ranging in age from 1 to 9 years were implanted with norgestomet implants during the natural breeding season (September, 1995). Ewes were blocked by breed and implants were removed from the ewes at either 12, 14 or 16 days and time to estrous onset was recorded at twelve hour intervals. The mean time to estrus was 43 hours with a range of 24 to 72 hours. Time to estrus varied among the different ages of the ewes. Ewes one year of age had lengthened time to estrous onset when compared to ewes 2-9 years of age (51 vs 41h). Seventy-seven (88.5%) of the ewes showed estrus post-implant removal. Of the 77 that showed estrus, 58 (75.3%) of the ewes conceived at the synchronization. The conception rates were different across age as they varied from 39% (1 year of age) to 90% (5 years

of age). The overall lambing percentage of the ewes was 154%. Lambing percentage was lowered as implant treatment was lengthened as ewes with implants removed at 12 days had a lambing percentage of 181% compared to 16 day implant treated ewes which had a lambing percentage of 128%. Results from this study indicate age of ewe and length of norgestomet implant treatment influence the success of estrus synchronization in the ewe when using a single norgestomet treatment during the natural breeding season.

#### INTRODUCTION

Many studies have incorporated the use of norgestomet implants for out of season breeding with pregnant mare serum gonadotropin (PMSG) (Boland et al., 1979; Fitch et al., 1986; Tritschler et al., 1991). The pregnancy rates from these studies have indicated that results are comparable to that of natural mating (60 to 65%). Little information is available concerning the use norgestomet implants without PMSG to synchronize ewes during the natural breeding season. Woody et al. (1983) studied the effectiveness of norgestomet implants without the use of follicular stimulants in regards to treatment dose, day of cycle in which treatment was initiated and fertility post-treatment. Ewes treated for 13 days with norgestomet implants containing 6 mg norgestomet

had a higher conception rate when compared to 3 mg implants (89% vs. 63%). Furthermore, day of cycle in which norgestomet treatment began influenced the estrus response and conception rates in ewes. Ewes in which norgestomet treatment was initiated on day 13 of the estrous cycle had a lowered estrus response (87%) and conception rate (33%) when compared to ewes treated on day 4 of the estrous cycle (100% and 72%, respectively). These results indicate that efficient estrous synchronization and fertility can be accomplished using 6 mg norgestomet ear implants.

The present trial was designed to test the efficacy of using norgestomet implants as a synchronization tool during the natural breeding season without the use of follicular stimulants such as PMSG. Using norgestomet to synchronize lamb crops would allow the commercial producer to obtain uniformity in his lambs while also giving him the ability to regulate the lambing season at a relatively low cost.

#### MATERIALS AND METHODS

Eighty-seven Dorset, Montadale, Hampshire and Suffolk ewes were utilized in this trial in September of 1995. Ewes were implanted subcutaneously in the ear with used Syncromate-B norgestomet implants (G.D. Searle and Co., Chicago) (Fitch et al., 1986) and were randomly assigned to

treatment groups according to implant treatment length. The implants were pulled at either 12, 14 or 16 days from insertion. Ewes were then exposed to rams wearing marking harnesses which were given a Breeding Soundness Exam prior to the study. Hours to first expression of estrus was recorded in twelve hour intervals according to when ewes were marked by the rams. No more than five ewes were exposed every other day to each ram post-implant removal in order that rams would not be over worked during breeding.

Ewes were checked for pregnancy at 60 days following the day that the first ewe marked by the ram. Pregnancy was determined by ultrasound utilizing an Aloka 210DX unit equipped with a linear array, 5 MHz transducer (Corometrics Medical Systems, Inc., Wallingford, CT). The date of lambing and type of birth was recorded with the corresponding identity of each ewe.

General Linear Models and Least square means for SAS (1985) were used to measure the effects of treatment (TRT), age of ewe (AGE) and breed of ewe (BRD) on time to estrus (TTB), total ewes conceiving to synchronized estrus (CR), ewes lambing to first synchronized estrus (PR), and lambing percentage (LP-no. of lambs born per ewe) (Table 1).



## RESULTS

Ewes were randomly assigned to one of three treatment groups (Day 12, 14 and 16) within age (1 to 9 years) and breed (Dorset, Hampshire, Montadale and Suffolk) as presented in Table 2. Least square means for TTB, PR, CR and LP by TRT, BRD and AGE are presented in Table 3.

There were no breed effects ( $P > 0.05$ ) on TTB, PR or CR, and pregnancy rates did not differ ( $P > 0.05$ ) due to TRT, BRD or AGE. Montadale, Dorset and Hampshire breeds did not differ in LP, but the LP of the Suffolk (115%) breed was lowered when compared to Montadale (162%), Dorset (168%) and Hampshire (160%) breeds ( $P < 0.05$ ). Treatment affected LP ( $P < 0.05$ ) as 12 day implanted ewes had a higher percentage lamb crop (180%) when compared to 14 and 16 day implanted ewes (144% and 128%). There was also a treatment effect ( $P < 0.05$ ) on TTB as 14 day treated ewes had a lengthened TTB when compared to 16 day treated ewes (46h vs. 39h). Age of dam affected TTB and CR ( $P < 0.05$ ). In ewes one year of age TTB was 51 h as the other ages which ranged from 38-45 hours. While CR for yearling ewes was 39.1%, which is significantly lower compared to 56.4%-89.9% for 2-9 year olds.

Distribution of times for time to first estrus expression (TTB) for AGE, BRD and TRT are presented in

Tables 4 through 6. Ten of the ewes in this trial did not show a response to norgestomet treatment. Eight of the ten ewes which did not express estrus, were yearling maiden ewes. It should be noted that a majority (<50%) of all ewes regardless of age, breed or treatment had initiated estrus by 48 hours.

#### DISCUSSION

Estrus response to norgestomet treatment in the present study was similar to reported findings of other research (Boland et al., 1979) when using norgestomet only as treatment for synchronization as estrous response was 88.5% (77 out of 87) compared to 66.7% (8 out of 12).

While time of initiation of estrus (TTB) was variable among TRT (39 to 46h) and AGE (38 to 51h), it was similar to that of earlier reports as the mean TTB was 43h compared to 38h to 46h of reports using norgestomet implants (Boland et al., 1978; 1979; Fitch et al., 1986).

Conception rate (total ewes conceiving to synchronization) was 66.7% (58 out of 87) and pregnancy rate (ewes lambing to first synchronized estrus) were 75.3% (58 out of 77) for the present study. These numbers are similar to other reports which had a CR of 50 to 63% and a PR of 60 to 65% when using norgestomet implants in combination with

follicular stimulants (Spitzer and Carpenter, 1981; Fitch et al., 1986; and Tritschler et al., 1991). Therefore, it is possible to use norgestomet only for synchronization within the natural breeding season to achieve adequate fertility rates.

Lambing percentage (LP) was dependent on length of treatment. As implant length increased the LP decreased from 180% using 12d implants to 128% using 16d implants. This decrease in lambing percentage is due to the aging of ovulatory follicles, and is similar to reduced fertility associated with persistent dominant follicles observed in cattle when using exogenous progesterones (Savio et al., 1993b).

The mean LP was 157% in the present study which is similar to 181% reported by Fitch et al. (1986) and 144% of Tritschler et al. (1991). However, there is an indication that parity of ewes, particularly maiden ewes, adversely effected the success of attaining synchrony when using norgestomet only. This was evidenced by a lengthened TTb and 32% (8 out of 25) maiden ewes were unresponsive to norgestomet treatment.

The lower LP (115%) in the Suffolk ewes (usually a very prolific breed) may be attributed to the fact that of the 18 Suffolks that lambed in this trial eight of them (44%) were yearling maiden ewes compared to 2 of 11 (18%) Dorsets, 3 of

13 (23%) Hampshires and 4 of 35 (11%) Montadales in the study. Kelly (1984) has shown that age of ewe is a consistent factor in embryonic loss. Although fertilization rates in maiden ewes and older ewes has been recorded as being similar (86% vs 88%), the proportion lambing following transfer of cleaved ova was markedly lower (39% vs 75%) indicating substantial embryonic wastage in maiden ewes. However, in the present study fertilization success was due to whether there was an estrous response in maiden ewes rather than a problem with early embryonic death.

#### IMPLICATIONS

Synchronization of ewes during the natural breeding season is possible using a single norgestomet impregnated implant. Close attention to dose, length of treatment and age of ewe should be taken into consideration when utilizing this technique in a commercial situation as these factors may effect the synchronization capabilities of the norgestomet implant.

Table 1. Analysis of variance table for the effects of treatment, breed and age and appropriate interactions on time to estrus (TTB), total ewes conceiving to synchronized estrus (CR), ewes lambing to first synchronized estrus (PR), and lambing percentage (LP).

Source	Degrees of Freedom
Treatment	2
Breed	3
Age	5
Treatment x Breed	6
Treatment x Age	10
Breed x Age	15
Treatment x Breed x Age	30
Residual	15

Table 2. Number of ewes per treatment by Age and Breed.

	Implant Treatment Length (days)		
	12	14	16
<b>Age</b>			
1	10	8	7
2	5	5	5
3	6	8	3
4	4	3	1
5	4	5	4
6	1	1	0
7	2	2	1
8	1	0	0
9	0	1	0
<b>Breed</b>			
Dorset	4	5	4
Hampshire	5	6	3
Montadale	16	16	9
Suffolk	8	6	5

Table 3. Least square means for estrous initiation (TTB), pregnancy rate (PR), conception rate (CR) and lambing percentage (LP) by Treatment, Breed and Age.

Groups	n	TTB <sup>c</sup> (hr)	PR <sup>d</sup> (%)	CR <sup>e</sup> (%)	LP <sup>f</sup> (%)
<b>Treatment</b>					
12d	33	43 <sup>a,b</sup>	68.8	69.3	180 <sup>a</sup>
14d	33	46 <sup>a</sup>	72.4	66.0	144 <sup>b</sup>
16d	21	39 <sup>b</sup>	71.3	70.0	128 <sup>b</sup>
<b>Breed</b>					
Dorset	13	43	64.1	60.2	168 <sup>a</sup>
Hampshire	14	45	72.4	71.7	160 <sup>a</sup>
Montadale	41	38	80.3	72.1	162 <sup>a</sup>
Suffolk	19	44	66.5	70.0	115 <sup>b</sup>
<b>Age</b>					
1	25	51 <sup>a</sup>	59.6	39.1 <sup>a</sup>	155
2	15	43 <sup>b</sup>	64.8	65.1 <sup>a,b</sup>	142
3	17	45 <sup>b</sup>	87.3	88.1 <sup>b</sup>	163
4	8	38 <sup>b</sup>	68.6	72.0 <sup>a,b</sup>	146
5	13	41 <sup>b</sup>	85.0	90.0 <sup>b</sup>	146
6-9	9	38 <sup>b</sup>	59.5	56.4 <sup>a, b</sup>	153

<sup>a,b</sup>Those in the same column with different superscripts differ significantly ( $P < 0.05$ )

<sup>c</sup>TTB = time to estrus initiation

<sup>d</sup>PR = ewes lambing to first synchronized estrus divided by total number of ewes responding to synchronization in each treatment, breed or age

<sup>e</sup>CR = ewes conceiving to synchronized estrus divided by total number of ewes in each treatment, breed or age

<sup>f</sup>LP = lambing percentage of ewes lambing

Table 4. Distribution for time to estrus initiation (TTB) by age of ewe.

Ewe Age	No.	Hours from implant removal to estrus					
		24	36	48	60	72	NR <sup>a</sup>
1	25	0	2	11	1	3	8
2	15	1	6	6	2	0	0
3	17	0	5	10	1	0	1
4	8	2	4	2	0	0	0
5	13	3	6	3	1	0	0
6-9	9	4	1	2	1	0	1
Totals	87	10	24	34	6	3	10

<sup>a</sup>NR = no response to implant treatment



Table 5. Distribution for time to estrus initiation (TTB) by breed of ewe.

Breed	No.	Hours from implant removal to estrus					
		24	36	48	60	72	NR <sup>a</sup>
Dorset	13	2	3	4	1	1	2
Hampshire	14	0	4	5	4	0	1
Montadale	41	8	14	12	1	1	5
Suffolk	19	0	3	13	0	2	1
Totals	87	10	24	34	6	3	10

<sup>a</sup>NR = no response to implant treatment

Table 6. Distribution for time to estrus initiation (TTB) by Treatment (TRT).

TRT	No.	Hours from implant removal to estrus					NR <sup>a</sup>
		24	36	48	60	72	
12	33	6	6	17	0	2	2
14	33	1	9	13	3	1	6
16	21	3	9	4	3	0	2
Totals	87	10	24	34	6	3	10

<sup>a</sup>NR = no response to implant treatment

## Chapter V

### A PRELIMINARY STUDY: A NEW WAY OF EVALUATING SEXUAL BEHAVIOR IN RAMS

#### ABSTRACT

The sexual behavior of two low and three high libido scored rams was evaluated during the natural breeding season. Each ram was penned with 11 to 12 estrous synchronized ewes for a period of four days. Number of mounts and frequency of mounts were recorded via the HeatWatch System. The results of this study indicate proficient mating differences in the behavior of sexually mature rams.

#### INTRODUCTION

Evaluating sexual activity in the ram is an important aspect of reproduction in the sheep industry. Past studies have used libido and serving capacity tests to evaluate the reproductive performance of the ram (Perkins et al., 1992; Price et al., 1992). Libido tests evaluate the sexual aggressiveness (mounts/amount of time), while serving

capacity tests evaluate the sexual performance (ejaculations/ram/amount of time). A study by Perkins et al. (1992) found that the mating performance of rams can be predicted by using libido and serving capacity tests.

The purpose of this study was to monitor the ram's mating activity using the HeatWatch system. The HeatWatch System allows for continuous recording the total number of mounts per ram, total number of mounts per ewe and total number of ewes mounted during the test period.

#### MATERIALS AND METHODS

Thirteen rams were libido tested by utilizing a similar protocol described by Price et al. (1992). All rams had previously passed a Breeding Soundness Exam prior to the serving capacity tests. Each ram was introduced to five synchronized estrous ewes, and number of mounts and ejaculations during a 15 minute period were recorded. Rams were scored as "High" libido (HL) rams (n=3) if at least 5 ejaculations or 7 mounts were achieved, while rams that had less than 2 mounts or 0 ejaculations were considered "Low" libido (LL) rams (n=2).

After libido score was established, each ram was placed in a pen with 11 to 12 ewes two days prior to implant removal for adjustment to the new environment in order that

accurate estrous detection might be accomplished upon initiation of the trial. Synchronization was achieved by implanting the ewes with norgestomet implants for 12 days. Implants were removed in half of the ewes in each pen on the initial day of synchronized estrous and the remainder of the ewes in the pens on the following day. Rams were penned in with ewes for four days post-implant removal.

Mounting was recorded with a HeatWatch System (DDX Inc., Boulder, CO) transducer and patch adhered to the rump of each ewe. A main computer program recorded the identity of the ewe and time at which each mount took place. The duration of the mount necessary to record to the main computer was set for one second. Total mounts per ram, number of ewes mounted and number of mounts per ewe were analyzed.

Forty-five days following the last day of breeding, ewes were monitored for pregnancy via ultrasound by using an Aloka 210 with a 5.0 MHz transducer (linear array, Carometrics Medical Systems, Inc., Wallingford, CT). Pregnant and open ewes were noted and fertility rates were calculated for each of the rams.

## RESULTS

Ram comparisons are presented in Table 1. Two rams,

one LL (#55) and one HL (#59), were removed from the trial for comparisons. Mounting behavior data was inaccurate for these rams as ultrasound pregnancy figures indicated more ewes pregnant than were mounted. However, the other three rams mating behavior was distinct from one another and resulted in applicable comparisons. The other LL ram (#51) mounted only three times, mounting a total of three ewes and resulted in no confirmed pregnancies. One HL ram (#57) mounted a total of 50 times, covering 9 ewes and resulted in 7 pregnant ewes. A second HL ram (#61) mounted a total of 12 times covering 3 ewes resulting in 2 pregnancies.

#### DISCUSSION

Results from this trial indicate variation in the mating behavior of rams. Three types of rams can be distinctly identified: 1) a ram (#57) that is highly aggressive and efficient (several mounts and ewes mounted resulting in seven out of nine pregnancies), 2) a ram (#61) that is highly aggressive but inefficient (several mounts on one ewe resulting in 2 pregnancies), and 3) a ram (#51) that is not aggressive nor efficient (few mounts and ewes mounted resulting in no pregnancies).

Though ram #55 was a low libido scored ram in human observed serving capacity tests, it should be noted that his

pregnancy rates were equal to that of the #57 ram (highly aggressive and efficient ram). This indicates that being able to analyze the mounting activity of rams without the presence of humans might allow for more accurate evaluation of rams and their sexual behavior in actual field conditions.

The use of the HeatWatch system in this study gave researchers the ability to evaluate an individual ram's sexual behavior over a 24 hour per day period without being present for observation. With this technology frequency of mounts and movement from ewe to ewe can be evaluated.

Some minor problems were detected with the HeatWatch system as some of the pregnancy rates did not correspond with the ewes mounted in this trial. It was observed that longer fleece could hinder the transducer's effectiveness. When using this tool, the researchers of this trial recommend four weeks of fleece growth or less to properly utilize the HeatWatch system when using it with sheep.

#### IMPLICATIONS

A producers ability to evaluate and select for highly aggressive and efficient breeding rams could prove to be a very cost effective tool in sheep reproduction. Scoring the mating behavior in breeding rams needs to be researched

further to make it a useful tool in ram selection.



Table 1. Mounting data of high and low libido mature rams.

Ram ID	Libido score	MPR <sup>a</sup>	MPE <sup>b</sup>	NEM <sup>c</sup>	UPG <sup>d</sup>
51	low	3	1.0	3	0
55 <sup>e</sup>	low	6	1.5	4	7
57	high	50	5.6	9	7
59 <sup>e</sup>	high	9	9.0	1	6
61	high	12	4.0	3	2

<sup>a</sup>Mounts per ram during trial

<sup>b</sup>Mounts per ewe mounted during trial

<sup>c</sup>Total number of ewes mounted per ram during trial

<sup>d</sup>Number of ewes per ram pregnant to ultrasound at 45 days

<sup>e</sup>Rams not analyzed in study due error in mount detection.

## CHAPTER VI

### SUMMARY AND CONCLUSIONS

Two studies were performed to evaluate the performance of ewes synchronized with norgestomet and its effect on (a) timing of estrous onset, (b) timing of ovulation, (c) the interval from onset of estrus to ovulation (d) fertility rates and (e) number of lambs born per ewe. A preliminary study was also performed to evaluate the sexual performance of high and low libido scored rams using the HeatWatch System to monitor mounting activity around-the-clock.

Eighty-seven ewes consisting of four breeds and ages ranging from 1 to 9 years in age were utilized to determine whether norgestomet implants were efficient for synchronizing estrus and lambing in ewes. Seventy-seven (88.5%) of the ewes respond to synchronization by exhibiting estrus. Of the ewes that exhibited estrus, 58 (75.3%) of these ewes lambed to the synchronized estrus. These synchronization and pregnancy rates are very acceptable in accomplishing a synchronized lambing season. Because of this fact, utilizing norgestomet within the breeding season could give producers the opportunity to maintain uniformity in their lamb crops, thus reducing time and management

during the lambing season and allow for greater profitability in the production of lambs.

The ultrasound study focused on the timing of events during the follicular phase of the estrous cycle in ewes following treatment with norgestomet, norgestomet in combination with PMSG and PGF<sub>2α</sub>. The most significant findings of this study is that norgestomet used in combination with PMSG tends to tighten synchrony and advance timing of estrous initiation and ovulation in ewes when compared to the other treatments and should be the synchronization technique utilized when artificially inseminating ewes on a timed AI basis. Additionally, the interval from onset of estrus to ovulation is similar (approximately 36h) in ewes treated with norgestomet only or norgestomet in combination with PMSG. However, ewes treated with PGF<sub>2α</sub> tend to have a shortened interval (21h) and should be adjusted for if one utilizes this synchronization treatment before artificially inseminating ewes. The present study indicates that artificially inseminating ewes at a set time from initiation of estrus rather than a timed AI from implant removal should give rise to higher fertility rates as estrus response to treatments were highly variable among the ewes in the present study. This, however, should be studied further before concrete recommendations can be made.

The use of the HeatWatch system proved to be a very useful tool for monitoring both ewes and rams in the studies in which they were utilized. Precise detection of estrous initiation allowed for accurate determination of the timing of events during the estrous cycle in ewes. This technology could be used in sheep as it is in cattle to increase fertility rates while being utilized to AI at a set time after the initiation of estrus. In addition, the HeatWatch System could be used as an alternative to more accurately rate or score a rams sexual performance for pasture and pen breeding systems as the preliminary study has indicated. The system might be useful, as well, in monitoring a group of rams in pasture mating systems by not only applying the HeatWatch System patches to the rumps of ewes, but also by applying the patches to the breast of each rams. This would allow for 24 hour monitoring of the rams' behavior, and allow researchers the ability to observe hierarchy breeding by rams penned together. Continued study needs to be done to evaluate the feasibility of this monitoring system.

In conclusion, the current practices utilized for AI in sheep are most likely the best at this time. Utilization of technology such as the HeatWatch System, while expensive, might give rise to higher fertility rates in the future. Likewise, the development of a sound and consistent TAI method should increase the use of AI, and benefit the sheep

industry as it has in the other agricultural animal industries.

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