

THE EFFECTS OF GONADOTROPIN RELEASING HORMONE
ON CONCEPTION IN POSTPARTUM ANESTRUS ANGUS
BEEF COWS AND HEIFERS

A Thesis

Presented to the

Faculty of the Graduate School of
Angelo State University

In Fulfillment of the

Requirements for the Degree

MASTER OF SCIENCE

by

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December 2012

Major: Animal Science

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ACKNOWLEDGEMENTS

I first would like to thank God for all the blessings in my life. Also I would like to extend my deepest gratitude and thanks to Dr. Micheal Salisbury, my thesis advisor and chairperson, for his guidance and support through my graduate stay at Angelo State University. I would also like thank the other members of my advisory committee: Dr. Gil Engdahl, Dr. Kirk Braden, and Dr. Charlene Bustos. I would also like to acknowledge and give a huge thanks to Dr. Kirk Braden, Kris Ede and Robert Cope for always believing in me, for all the advice and support through the thick and thin while coaching and my graduate work. In addition, I would like to thank all the faculty and staff in the Agriculture Department, as well as the graduate students for all the help through my study.

And lastly, I have to thank my family Mom, Dad, Kay, Randy, Chris, Sandra, Amy and William for all the support they gave to me during this time of change in my life. In addition, I would like to extend a thanks to my second family Mr. and Mrs. Robert and Abban Lastovica for believing in me, when I chose to go back to school and further my education. And finally, I am so very grateful for my best friends Jerred and Melissa and all my friends that I made here at Angelo State University for their support.

ABSTRACT

The objective of this study was to determine if a pre synchronization gonadotropin releasing hormone (GnRH) injection would increase conception percentages in postpartum anestrous angus beef cows and heifers. Sixty-four Angus females (42 multiparous cows and 20 heifers) from the Angelo State University's Management, Instruction and Research Center were randomly assigned to two treatments groups at the beginning of the study. The control group were administered a two shot prostaglandin estrous synchronization protocol. The treatment group were administered a GnRH injection thirteen days prior to the two shot prostaglandin synchronization protocol. However, only 30 females showed signs of estrus and were artificially inseminated. Results in cows and heifers were similar, and no differences were found among the two treatment groups ($P>0.05$). Differences might be noted with a larger number of females that show signs of estrus. Conception was recorded by the use of ultrasound, and is a viable management tool.

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INTRODUCTION

Through past research and technological advancements, the world of reproductive physiology has advanced leap years in terms of overall mindset and practice in the beef industry. Advancement factors such as synchronization protocols, genetic markers, artificial insemination and nutrition have been vital tools for small producers in the commercial beef sector to stay with large scale producers. The most limiting factor in the animal industry for all producers is reproductive efficiency. In basic terms it is the number of cows that conceive during a given breeding season that dictates the overall profitability of the herd. One of the largest problems the beef industry faces is the occurrences of females not resuming normal estrous cycles postpartum. This period is referred to as postpartum anestrous. This problem causes the calving season to be stretched out over a longer period of time, which consequently will become a deterrent to your calf crop uniformity. With the problem of reproductive inefficiency in the beef cattle, figuring out ways to shorten the period postpartum with having a higher percentage of females cycling at the start of breeding season will give insight to correcting the problem. With a solid understanding of this it should lead to higher revenue in the end for your production scheme.

In trying to approach the reason for long periods of postpartum anestrous that can hinder the overall production in beef cattle, one factor to explore is jump starting the

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estrous cycle. A majority of other studies have shown that applying an increase in the nutritional plane of the female in order to reach the desired body condition score prior to calving leads to optimal results for females resuming normal estrous cycles post calving.

Keeping economics in check with the current high prices of feedstuffs, could it be possible to mimic the endocrine system that it is ready to resume cycling after calving with a simple hormonal treatment?

OBJECTIVES

1. Determine the effects of pre-synchronization Gonadotropin Releasing Hormone injection on postpartum estrous cycles in heifers and mature cows
2. Determine the effect of pre-synchronization Gonadotropin Releasing Hormone injection on conception rate in heifers and mature cows

LITERATURE REVIEW

Postpartum anestrous in beef cattle can be defined as the period of time post calving that cows do not show signs of cycling. During the bovine anestrous period, normal follicular waves do occur, but standing estrus and actual ovulation does not occur (Perry, 2004). Anestrous period in cattle can range from 14-180 days in postpartum cows. Normal ranges in mature cows range from 30-90 days and for young cows it ranges from 60-120 days postpartum (Anderson, 2010). There are many causes of this delay after calving that can cause females to become out of sync with their contemporary group. With these problems it can cause the breeding season to sometimes be delayed and cause a longer calving season. This problem is referred to as the most challenging factor that reduces reproductive efficiency in beef cow calf operations (Anderson, 2010).

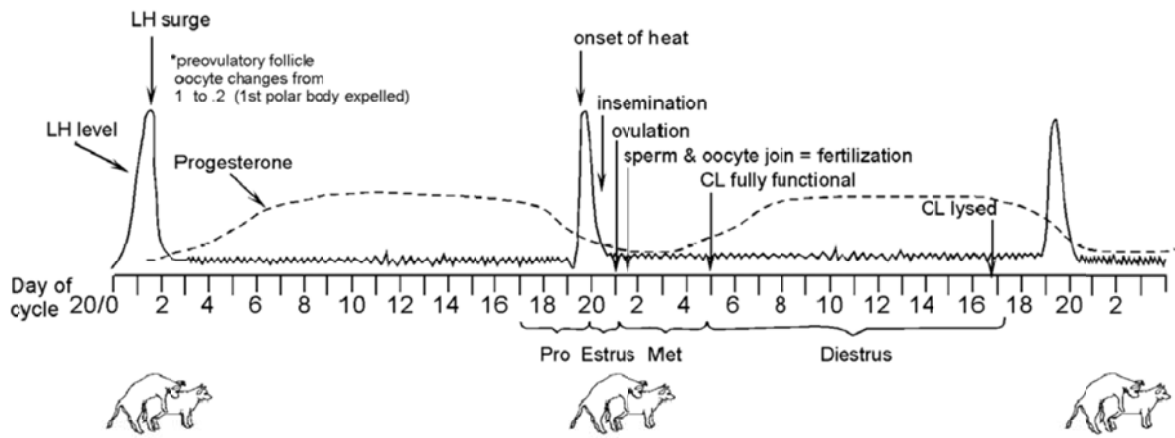
Many different factors causing a delay will affect the anestrous period in cattle, including but not limited to presence of a suckling calf, age of female, body condition score (BCS) and the difficulty of calving. It has been said that the presence of a calf is one of the most influential factors that plays a role in how long before estrous cycles resume. In a study Williams (1990) stated, “the continual presence of a suckling calf prolongs anestrous and the delays the re-initiation of estrous cycles”. Researchers also state that the removal of the calf temporarily or permanently usually initiates estrous within a few days (Williams, 1990). The effect of the suckling calf on the female is imperative to understand, because it puts an external stress on the overall nutrition requirements of the female for re breeding purposes. The age of the female is another pertinent factor because the producer needs to know if the

nutritional intake is being put towards growth of the individual or to uterine repair. Nutrients are utilized by cows according to an established priority of needs (Short and Adams, 1988). The first priority is maintenance of essential body functions to preserve life in the animal. Once that requirement is met, the remaining requirements accommodate growth (Stevenson et al., 2002). Because of these additional requirements for growth it is generally why younger cows have a longer anestrus period when compared to mature cows (Short and Adams, 1988). In older females growth requirements have been previously met, thus nutrients consumed can be put towards lactation and towards resuming estrous cycles (Stevenson et al., 2002). This is why it can be linked that younger, still developing females typically produce less milk and have longer anestrus periods post calving. The animal "Body Condition Score" (BCS) truly goes hand in hand with the previously stated nutrient requirements. The animal's BCS prior to calving and after calving will play a vital role in the overall length of the anestrus period. According to Hall (2004), "the most critical period in the production year is the last 60 to 90 days before calving. Not only are dramatic changes about to occur in the female, but this period also sets the stage for the reproductive success or failure in the months that follow." This period is imperative to the success of the producer because on average a cow will drop a full BCS in the period postpartum due to the demands of lactation that the calf is putting on the female. Therefore, being in the proper BCS at the time of calving is imperative so that the female can remain at the optimum BCS at the beginning of the breeding season in hopes to minimize the anestrus period post calving. The difficulty of parturition, referred to as dystocia, is one of the last factors that plays a role in the length of anestrus. The occurrence of dystocia can cause direct injuries to the

reproductive tract. If damage does occur there will have to be extensive repairs done to regain normal estrous cycles. The process of these repairs is known as uterine involution. Uterine involution is a process defined as the regressing changes in female reproductive organs that return the uterus to a normal non-pregnant state (Gray et al., 2003). In cattle this uterine involution is complete when the uterine horns return to their normal position, size and tone of a normal cycling female (Casida et al., 1968). This process must be successful so that the animal can return to its normal cycling patterns. If this process is unsuccessful, it can result in a longer anestrous period. With these factors of why postpartum anestrous can occur, it validates the reason of why reproductive inefficiency is a major concern amongst cow calf producers.

The use of estrous synchronization in bovine has been one of the biggest advancements in the beef industry. This technology has allowed producers the opportunity to capture the economic benefits of artificial insemination (AI). Synchronization also allows the producer the benefit of narrowing their calving season to a smaller window and market their offspring as a more uniform package. This technology can be achieved by the usage of progestogens, progestogen-prostaglandin combinations, prostaglandins alone, progestogen-estrogen combinations, and gonadotropin-prostaglandin combinations with or without progestogens (Larson and Randle, 2008). But to fully understand the benefits of estrous synchronization, you must have a firm grasp of the bovine estrous cycle.

The bovine estrous cycle is typically 21 days in length and comprised of four different periods: estrus, metestrus, diestrus, and proestrus (Figure 1). During the period of



* Two full cycles and the start of a third.
 Figure 1. Schematic of stages of the estrous cycle, serum progesterone concentrations, and serum luteinizing hormone (LH) concentrations .

Image taken from Larson and Randle, 2008.

estrus this is the time that the cow or heifer is receptive to the male by standing to be mounted. This period usually lasts about 20 hours in length. During this period of the cycle the corpus luteum (CL) has already been lysed and the progesterone levels in the blood are at a very low level. In this period luteinizing hormone (LH) spikes to a high level, which initiates ovulation to occur. Estrogen levels are of decreasing levels from the high levels that were just prior to estrus (Larson and Randle, 2008).

Ovulation occurs during metestrus, about 10-15 hours after the end of estrus. During this period the CL is in the early development stage. Levels of progesterone are still low but slightly increasing. This is occurring because the CL is small in size and doesn't have the power to produce large amounts of progesterone. During this stage prostaglandin F₂ α is not effective in lysing the growing CL (Larson and Randle, 2008).

The period of the CL is referred to as diestrus and typically last about 12 days. At this time the CL is producing increasing amounts of progesterone in the early stages of diestrus. During diestrus the concentration of progesterone plateaus in the blood stream (Larson and Randle, 2008). In the event that fertilization does not occur, PGF₂ α will lyse the CL and initiate a return to estrus.

The last stage of the estrous cycle is the period of proestrus. This period typically lasts about 2 to 3 days. This is the period of the regression of the CL and the final growth phase of the ovulatory follicle (Larson and Randle, 2008). In this stage the progesterone levels begin to decrease and the LH and estrogen positively feed off each other to produce a

rapid increase in estrogen production. During this stage, treatment with PGF2 α will not affect the animal's normal return to estrus.

Periods of the estrous cycle are controlled by the actions of the hypothalamus which sends signals to the anterior pituitary gland to release hormones to initiate certain parts of the estrous cycle to begin. The hypothalamic hormone, Gonadotropin releasing hormone (GnRH) causes the anterior pituitary gland to release LH and follicle stimulating hormone (FSH), in which these two hormones act as the main regulators of the estrous cycle (Larson and Randle, 2008). Having the information that GnRH is the catalyst for the production of FSH and LH in the cow; leads us to believe that GnRH could be the driving force behind possibly jump starting cycles earlier post parturition to cut down the time lost due to anestrous. The secretion of LH and FSH from the anterior pituitary gland is important in cattle, because LH allows for the maturation and production of the tertiary follicles. This is necessary for the maintenance of the CL and production of progesterone by the CL. In addition FSH allows for the maturation of secondary follicles to tertiary follicles (Larson and Randle, 2008).

With GnRH being the catalyst for the release of LH and FSH, which allows for the maturation and recruitment of follicles in the estrous cycle, this could allow producers to synthetically jump start the estrous cycle in anestrous females with a simple intermuscular injection. By possibly doing this, cow calf producers could perceivably cut their losses on non-cycling females at the beginning of the breeding season.

The purpose of this study is to determine if an injection of GnRH prior to an estrus synchronization protocol could jump start the endocrine system to allow for assurance

that the female would cycle at the beginning of the breeding season. If so would this increase the effectiveness of your estrus synchronization protocol and profitability of your operation?

MATERIALS AND METHODS

Within the Angelo State University purebred Angus herd, the mature cows, first calf heifers and yearling heifers were randomly assigned to one of two treatments in an alternating fashion blocked by parturition date. Due to the number of females that will be utilized in the study, females were separated into two uniform groups. Group one consisted of all the mature cows and a portion of the early calving first calf heifers, and group two will consist of the remaining first calf heifers and all of the yearling heifers. The two treatment groups for the study are 1) control; two shot prostaglandin protocol (DuPonte and Lee, 2007), and 2) treatment; Intermuscular Gonadotropin Releasing Hormone injection 13d prior to the two shot prostaglandin protocol.

The two working groups were placed on a wheat field prior to synchronization protocol and breeding. The treatment groups (2) within the two working groups received an intramuscular injection of Gonadotropin releasing Hormone (2 mL of Cystorelin; Merial, Duluth, GA) 13 days prior to the synchronization protocol. Both working groups (control and treatment groups) were then subjected to the two shot prostaglandin (5 mL of Lutalyse; Pharmacia & Upjohn Company, New York, NY) synchronization protocol normally used at the Angelo State University Management, Instruction and Research Center.

Once the second shot of prostaglandin was given, estrus detecting sensors (Heat Watch®, Cowchips LLC, NJ) were placed on the cows and heifers, and females were then returned to a wheat field adjacent to the cattle breeding barn. The sensors sent mount data to a receiver that was then be downloaded to a computer for determination of actual time of

estrus. Then twelve hours following determination of the initiation of estrus, cows and heifers were artificially inseminated with predetermined semen.

Animal I.D, time and date of standing estrus and numbers of mounts were recorded at the time of A.I. service. Once the two groups were A.I. serviced, they were put back into a native grass pasture at the Angelo State University ranch with Angus bulls that serviced any non-pregnant females that did not conceive at the time of A.I. and to mate with any females that did not express estrus during the time of data collection. One hundred eighty days after A.I. breeding, pregnancy was determined by ultrasound (ALOKA 500, Aloka Co. LTD, Wallingford, CT) via rectal palpation.

STATISTICAL ANALYSIS

Number of mounts and time post injection will be analyzed using PROC GLM procedure of SAS (SAS Institute, Cary, NC) with individual cow or heifer serving as the experimental unit with multiparous cow or heifer serving as a block. Conception rates were analyzed using Chi² and categorical models in SAS (SAS institute, Cary, North Carolina) and individual animal serving as the experimental unit. Treatments will be considered different at $P \leq 0.05$.

RESULTS

Sixty-two registered Black Angus females from the Angelo State University Management, Instruction and Research Center (n=62), were used in this research trial. All sixty-two females were subjected to either the treatment of a GnRH injection prior to the two shot prostaglandin synchronization protocol (22 multiparous cows and 10 heifers) or the control which received the two shot prostaglandin synchronization protocol (20 multiparous cows and 10 heifers). Of the sixty-two cows only thirty showed signs of estrus and were artificially inseminated (16 multiparous cows and 14 heifers) and included in the analysis. All effects will be discussed by both multiparous cow and heifer.

Cows

As shown in Table 1, the control group consisting of five multiparous cows and the treatment group consisting of eleven multiparous cows. Within the multiparous cows for the effect of days postpartum (DPP) until estrus, the control group had a mean of 62.15 days and the treatment group had a mean of 56.45 days. Numerically it appears that there is a difference between the control and treatment groups, but there were no statistical differences ($P=0.20$) between the treatments for this effect. With the data collected for number of mounts received between time of estrus and A.I. amongst the control and the treatment groups of cows. The control group displayed a mean of 20.8 mounts and the GnRH treatment group displayed a mean of 21.72 mounts before time of artificial insemination. With the means being numerically similar, there was no difference ($P=0.93$) between the treatments. To see if there would be a decrease in number hours between the last injection of prostaglandin and

Table 1. Estrus activity, and conception rates in Angus cows and heifers treated with or without Gonadotropin Releasing Hormone (GnRH) prior to a modified two shot Prostaglandin F 2 alpha (PGF_{2α}) estrous synchronization protocol

| | Control ^a | Treatment ^b | SE ^c | P-value |
|---------------------|----------------------|------------------------|-----------------|---------|
| Cows ^d | | | | |
| n | 5 | 11 | | |
| DPP ^e | 62.15 | 56.45 | 3.21 | 0.20 |
| Mounds ^f | 20.80 | 21.72 | 8.98 | 0.93 |
| Hours ^g | 81.10 | 65.22 | 10.57 | 0.23 |
| Conception, % | 80.00 | 90.90 | | 0.55 |
| Heifers | | | | |
| n | 7 | 7 | | |
| Mounds | 11.71 | 17.28 | 4.59 | 0.40 |
| Hours | 69.21 | 61.78 | 8.53 | 0.54 |
| Conception, % | 71.40 | 85.70 | | 0.52 |

^a Control consist of the animals that were just subjected to the two shot prostaglandin synchronization protocol.

^b Treatment consist of the animals that were administered GnRH thirteen days prior to the two shot prostaglandin synchronization protocol.

^c Most conservative Standard Error

^d Cows indicate that they have had at least one calf born

^e DPP=days postpartum at the time of the first prostaglandin injection

^f Mounds=actual times the female was mounted prior to artificial insemination

^g Hours=hours between last prostaglandin injection and estrus

the time of estrus amongst the two groups we found the following means. The control group had a mean of 81.1 hours and the GnRH treatment group had a mean of 65.22 hours. There appears to be differences, but there was a high standard error (Table 1) that resulted in no difference ($P=0.23$). For conception rate among the multiparous cows the control group had 80% conception rate and the GnRH treatment group had a 90.9% conception rate. No differences ($P=0.55$) were found among the groups.

Heifers

As shown in Table 1, the control group consisting of seven heifers and the GnRH treatment group consisting of seven heifers. In the data collected to determine if the GnRH would increase the strength of the heat in terms of the number of mounts the female would receive we found that the control group had a mean of 11.71 mounts and the GnRH treatment group had a mean of 17.28 mounts. Again there was no difference ($P=0.40$) between the treatment and control. For the effect of number of hours between the last injection of the prostaglandin and the time of estrus for the heifers, to see if the GnRH injection would shorten and tighten the period after the completion of the protocol, we found the following means. For the control group of heifers the mean was 69.21 hours and for the GnRH treatment group 61.78 hours. For the effect of hours after last prostaglandin injection being shorter, there showed no difference ($P=0.54$) among the two the groups. In terms of conception rate between the two groups of heifers, the control group had a conception rate of 71.4% and the GnRH treatment group had a conception rate of 84.7%. With the conception rates being numerically similar, there was no difference ($P=0.52$) among the groups.

DISCUSSION

If this trial was repeated with a larger group of females, it is likely the results may show a difference in the benefit of using GnRH as reproductive aid among anestrous females. Typically when an estrus synchronization protocol is used you can see about 75% to 90% of the females show signs of estrus within five days (DeJarnette, 2004). With that being said, if more of the females on trial would have responded to the modified two shot prostaglandin protocol (48%), it is likely that there would have been more of a statistical difference since that would have increased our sample size. If this trial would be replicated again, adding the aspect of individual body condition scores to the females going into the trial would benefit the research. With the recent years of droughts that Texas and the southwest has been experiencing, I feel that this could possibly benefit producers that may have high risk/ low body condition females in their herds. So with incorporating that aspect into the trial, it could maybe shine some light on to possibly increasing the conception rate amongst those high risk females.

IMPLICATIONS

This study found no differences among the treatments between using the standard two shot prostaglandin synchronization and utilizing a GnRH injection 13 days prior to the synchronization protocol. However, more studies done with larger numbers of females, with assigned body condition scores, would be more beneficial to the research to determine if the addition of GnRH prior to the synchronization protocol would show higher conception rates amongst anestrous cows and heifers.

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