

Estrus Activity After Prostaglandin Injection using Heatime Activity Monitoring Collars
used at Cal Poly Dairy

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Abstract

The objective of the study was to determine activity using an activity monitoring system, Heatime. Data was collected from cows at the Cal Poly dairy from December 30, 2011 to January 6, 2012 whose results are from the previous program. This program involved prostaglandin in the form of Lutalyse given on Friday and cows were time bred the following Monday, 72 hours after the prostaglandin injection. Data collected from January 13, 2012 to current shows the new trial program which involved an injection of 5cc Lutalyse Monday and Tuesday, a vet check on Wednesday, time bred on Thursday, and a possible injection of Gonadotrophin- releasing Hormone (GnRH). Cows that have been checked by the veterinarian and are not showing signs of ovulation are given the additional GnRH injection. The results of both were compared to see if the additional injection caused immediate estrus cycling and showed signs of peak activity on the Heatime system more consistently than the previous program. Results showed that while not all cows came into estrus before or on the day of insemination, there were some consistencies in peak detection. The data showed that methods in which the two injections of Lutalyse were given on consecutive days, followed by time breeding on the fourth day had more instances where estrus was not demonstrated as peaking within the expected period. Although the results from those cows also given an injection of GnRH were shown to have higher instances of cows demonstrating peak estrus activity, it did not demonstrate enough to show that it is the best method. Limited results show that the new 4 day program without the incorporation of GnRH are not effective in creating peak activity and that the previous method of a single injection of Lutalyse may be the most beneficial. Factors that are possibilities for this outcome include time as a constraint, as well as inconsistent weather conditions over the duration of the new protocol. To see if

results will change, data needs to be observed using more cows and a longer period of time as well as including more cows from the previous protocol.

Table of Contents

Abstract-----	2
List of Tables-----	6
Introduction-----	7
Review of Literature-----	9
<i>Natural Hormones Released During Estrus</i> -----	9
<i>Signs of Estrus</i> -----	10
<i>Breeding Methods</i> -----	11
<i>Ovsynch Program</i> -----	12
<i>Detection Methods</i> -----	12
Methods and Procedures-----	14
<i>Data Collection</i> -----	15
<i>Data Processing</i> -----	15
Results and Discussion-----	17
<i>Results</i> -----	17
<i>Results Interpretation</i> -----	18
<i>Critical Analysis</i> -----	19

Conclusions----- 22

Citations----- 24

List of Tables

Table 1-----	19
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Introduction

Dairy farmers must do everything possible to be efficient with regards to reproduction expenses. Unfortunately, being efficient isn't always easy. The dairy industry is very limited to cutting costs due to the amount of resources it takes to function properly. Unfortunately, on the road to being efficient, trial and error may be the only method available, which can be expensive. Reproductive efficiency is a key component to running a successful dairy, to not only save money, but have a quicker turnover rate of heifers entering the milking string.

Old methods utilized to maximize reproduction rested solely on hormonal manipulation, visual detection methods, and time consuming estrus detection methods. This process may be expensive as well as ineffective. Newer protocols are introducing estrus detection via systems showing levels of activity sensed by detection monitors which are recorded on a computer program. The Heatime collar system designed by SCR Engineering may be a simple solution for these time consuming methods. The system is practical for dairies with up to 600 cows needing monitoring and the company prides themselves on their "user friendly" product. Each collar contains an H-Tag which reads movement by sending a signal to the ID unit which registers in the Data Flow or Micro Dairy Logic management program. The beneficial Heatime system runs 24/7, monitoring cows in 2 hour increments and requires no prior knowledge or training on how to use the program, as the system records and analyzes information automatically. SCR's website shows comparisons between Heatime systems and other motion devices, displaying that SCR's product had a 95% detection rate compared to the alternative's detection rate of 70%. In 2010, the Heatime system was installed at Cal Poly's dairy to be used on all

cows available to be bred that were currently enrolled in the Ovsynch program. (SCR Precision Dairy Farming, 2012)

The Ovsynch program that was being used up till January 16th of this year required one injection of Lutalyse Friday morning and time bred the following Monday morning regardless of estrus activity. With this protocol, cows that showed signs of estrus prior to Monday were bred early in hopes to breed at peak activity. A current observation is being done on a new protocol which requires two injections of Lutalyse, as well as an injection of GnRH if necessary, a veterinary check, and time breeding all within a four day period. The objective of this study is to evaluate if the new method is more effective in terms of causing proper estrus peak and how the Heatime system aids in determining such. Observing data from 48 cows of both methods beginning in 2011 to current is what the data will be based on.

Literature Review

Reproductive efficiency is a key aspect in every dairy, but developing the right program is a constant struggle for dairymen. While there is not a perfect program that can produce 100% reproductive efficiency, there are programs which may be much more beneficial and can produce improved pregnancy rates.

Natural Hormones Released During Estrus

In selecting a program, it is important to understand the natural hormonal change each cow must go through to begin ovulation. Before the ovulation process begins, the initial step involves the corpus luteum (CL) regressing. Once this process has occurred; levels of progesterone decrease, causing luteinizing hormone (LH) and follicle stimulating hormone (FSH) to be released within the body. The release of these hormones causes follicle maturation, which is the basis for ovulation. During the follicle's developing period, the dominant follicle then releases Estradiol. This stage is significant because the key hormones stimulating estrous behavior are secreted and those hormones are then responsible for estrus. Estradiol is responsible for collaborating with the hypothalamus and the pituitary gland; its job is to initiate secretion of hormones responsible for the commencement of ovulation. While the pituitary gland is responsible for the release of LH as well as several other hormones, the hypothalamus is responsible for the release of GnRH. As increased amounts of LH are secreted, ovulation is initiated. After ovulation has begun, the cow will initiate signs of estrous with prostaglandin levels at extreme highs. The pituitary gland responds to this high hormone level by reducing the level of GnRH secreted. As hormones have reached these normal levels for estrous and

the cow has reached peak activity, it is of optimal timing to utilize the breeding method of choice. (Boer, et al., 2009).

Kommadath, et al. performed a study to determine genes in the brain associated with estrus activity. In the study, genes observed were AVP, OXT, POMC, CGA, and MCHR1. The CGA gene was reviewed to be responsible for encoding three necessary hormones for estrus; Follicle Stimulating Hormone (FSH), Leutenizing Hormone (LH), and Thyroid Stimulating Hormone (TSH) and therefore was demonstrated to be present in high amounts when estrus was evident. When OXT was researched, it was found to be associated with the hippocampus (HC) as well as with the dorsal hypothalamus (DH). Similarly, when the gene AVP was evaluated it was determined to be associated with the HC and the amygdale (AM). These three genes were found to be associated with causing mounting and standing to be mounted. Hormones responsible for this AVP gene are Progesterone and estrogen, which in turn cause similar responses as oxytocin which heightens sexual arousal and partnering relationships. The genes MCHR1, POMC and a few others are known to control emotional and sexual impulses. (Kommadath, et al., 2011)

Signs of estrous

When a cow is in estrus, she demonstrates a number of different signals. Some signals are generally more noticeable than others, such as mounting. Standing to be mounted is the primary sign of estrous which can be easily overlooked (Roelofs, et al., 2005). There are also cases in which cows do not demonstrate any activity, but biologically are still going through the same ovulation stages. Dairyemen in the past have

commonly used these signs of estrous as a way to indicate optimal breeding time, but have found that relying on this method alone has often been inaccurate. Roelofs, et al., did an experiment of behavioral signs of estrous and ovulation times, in which observations included: cows sniffing the vulva of others, chin resting on another cow's back, mounting, mounting head of another cow, standing to be mounted, and level of restlessness. Their research revealed that the most prominent signs were sniffing the vulva of other cows as well as resting their chin on other cows (Roelofs, et al., 2005). When multiple cows were in estrus biologically, they expressed these behaviors more, whereas when there was only one cow in estrus, these behaviors were not expressed as frequently (Roelofs, et al., 2005). This research indicates that estrus can easily be missed if the visual signs have not been viewed by an employee. Results of missed estrous cycles can cause a dairy to lose money invested in hormones with attempts to synchronize the herd.

Breeding methods

Yoshida, et al., conducted a study where they utilized three different test groups and gave cows different injections of prostaglandin ($\text{PGF}_{2\alpha}$) followed by observation of the duration until estrus began. In these observations, it was determined that groups using CIDR Select Synch and groups using Select Synch both had high incidents of cows in estrus within five days of the program initiation. Those entering estrus were then observed and concluded that 74% of the estrous cycles occurred between 4-12 hours and peaked at 8 hours (Yoshida, et al., 2009). This study demonstrated that administering $\text{PGF}_{2\alpha}$ is efficient in ensuring synchronization for preferred breeding programs.

Ovsynch Program

The Ovsynch program typically uses two different injections of GnRH and one injection of PGF_{2α}. It has been noted that the program is most efficient when the PGF_{2α} is administered between the GnRH (El- Zarkouny et al., 2004). The purpose in utilizing PGF_{2α} is for regression of the corpus luteum and to begin ovulation so that a cow can be bred (Peters and Pursley, 2003). Peters and Pursley have found that while the Ovsynch program is very efficient, ovulation synchronization rates are between 80-90%, which leaves room for improvement to achieve more pregnancies (2003). This room for improvement Peters and Pursley mention does not necessarily need to be viewed as flaws within the Ovsynch program. The numbers demonstrate that when compared to previous methods of attempting synchronization, Ovsynch has been shown to be one of the most efficient methods to improve pregnancy rates. In addition to using the program, systems such as Heatime can be beneficial to make up where Ovsynch may be lacking in estrus activity earlier or later than expected, so as to increase pregnancy rates.

Detection Methods

A study was performed by Peralta, et al. to compare two different methods of heat detection systems with a visual observation method. HeatWatch and ALPRO were the two systems being observed in the study in addition to a visual observation done three times daily. Results from the study showed that visual observation had the highest rate of detection accuracy, followed closely by HeatWatch. From the research, it was determined that a combination of programs proved to have more successful conception rates than any of the three programs alone. A program involving HeatWatch followed by visual

observations 3 times a day had the highest rates overall, coming in at 26.9 +/- 4.6. Issues found within these heat detection programs lay within the method in which the system actually reads an interaction as estrus activity. The HeatWatch system specifically detected standing estrus, while the ALPRO system registered more mounting incidents. (Peralta, et al., 2005)

Methods and Procedures

The Cal Poly dairy's educational setting has allowed for students and faculty to review a variety of areas to improve reproduction efficiency methods and protocols. Our goal at the dairy has always been to determine the most efficient way of running a business, without jeopardizing limited resources or funds, and demonstrate useful tools to improve production and reproduction. From this idea, it is inferred that a key factor in becoming as resourceful as possible is reproductive efficiency. The first step to achieving increased reproduction rates is to accurately create a successful breeding program. Currently, there is no perfect program that can synchronize all cows 100% of the time and detect as well as inseminate within the appropriate window for optimal ovulation. Trial and error programs are continuously being performed to determine the most successful estrus detection program.

Previously, Rich Silacci, Cal Poly's herd manager has used the Ovsynch program, visual observation detection methods, and regular veterinary palpation checks to determine the estrus period. While the program has proven beneficial to the point of increasing herd size, Rich Silacci and staff considered an easier approach with a hopeful increase in pregnancy rates. The Heatime estrus detection system was one solution to catch missed estrus activity when employees missed observational periods, as well as provide statistical data saved in the Micro Dairy Logic program that could be later studied. After having this system for over a year period, another modification in reproduction practices is now being evaluated to improve efficiency. The modification of drugs administered to regress the corpus luteum and cause ovulation on a determined

schedule has been done to achieve this goal, in hopes that the two protocols (Heatime and drug modifications) can work simultaneously.

Data Collection

Cows that had the status *open* were eligible to undergo the new breeding protocol, but not every cow with this status was able to participate simultaneously due to limited supplies. Those selected to participate were assigned a collar which could be read by any of the ID units located in the transition pens above water troughs, as well as in the milking parlor. The period which the data was collected began on December 30, 2011 and ended on February 23, 2012. To give partial information on the previous program, six cows are listed, which were injected with Lutalyse on Friday mornings and time bred on Monday mornings. The remaining forty-two cows follow the current trial program which has Monday and Tuesday morning Lutalyse injections and Thursday morning timed breeding. The method used in determining which cows to use in the study was to take those cows selected by the herd manager that were given injections and then to see which of those each week showed any activity on the Micro Dairy Logic system. Cows will be documented based on the time they were given the shot and how long it took for them to peak, if they did.

Data Processing

The affect of how each cow responded to the injection of Lutalyse and GnRH (if administered) will be processed. The efficiency of peak activity read by Heatime collars will be monitored and the corresponding measure for those cows which have alerted the system as high activity. Dates for each injection will be taken into consideration, as well

as dates of noticeable estrus peak and dates of timed AI (TAI). Data evaluation may show a majority of cows demonstrating peak estrus prior to TAI or on the same date, and after the initial Lutalyse injection.

Results and Discussion

Results

Data was observed on a new trial protocol being run at the Cal Poly dairy to view its efficiency compared to previous methods. The method used previously was to administer only Lutalyse on Friday and then breed the following Monday. The trial method begins with an injection of GnRH Intramuscularly (IM) Wednesday, Lutalyse IM the following Monday morning, ending with a second Lutalyse injection on Tuesday morning, and then time bred Thursday morning, regardless of heat detection. The amount of Lutalyse and GnRH injected to each cow was 5 cc which is the desirable amount for each day to induce estrus. Because the protocol has been in effect since January 16, 2012, there is very limited data, and I was therefore only able to use forty-eight cows with usable data. Twenty- six of those observed were Holsteins and the remaining twenty-two were Jerseys.

Results show that from the week of January 1st, four Jerseys from the group were given shots Friday December 30th. This group peaked accordingly with the Lutalyse shot and was bred on the 2nd of January. There was only one Holstein showing consistent behavior from week two. She was given the initial shot on the 6th and then bred on the 9th, with a peak activity between a Standard Deviation (STD) of 24 to 25. The third week of data collected demonstrated that more cows provided consistent peak information. During this week, 3 Holstein and 1 Jersey all peaked on the 15th at a STD above 30 and then bred the same day, as well as 3 of them bred the following day. For the 4th week of January, GnRH shots were given on the 18th and Lutalyse was given on the 23rd and bred

on the 26th, although data we would like to see would demonstrate peak activity to be after the 23rd, only 2 cows (one of each breed) showed appropriate peak activity, whereas the other 3 peaked prior to the initial injection. The 5th group of cows was given injections on the 25th and 30th, 3 of which peaked between a STD of 15 to 24 on February 2nd and were also bred that same day. On February 6th, Lutalyse shots were given, 5 out of 14 showed appropriate peaks, 4 of which were Jerseys. Readings showed that STD ranged from 7-30 and that all but 1 that peaked did so on the day which they were also force bred, February 9th. The week of February 13th only had 2 cows show peak activity out of 6 which both had levels below 10. These cows were bred on the 16th and were not given GnRH. The remaining 9 cows only had 5 registered heats ranging from a STD of 2.5- 21.5. This group had inconsistent days in which they registered as peak activity.

Results Interpretation

Results show that many cows that have been introduced to this new method are having observable peaks ranging from 2.5 to 30+. Currently, there are issues with this method because there are several cows not demonstrating estrus appropriately as Table 1 clearly demonstrates. The data shows that peak estrous activity for the first two weeks of the program occur before the initial injection of Lutalyse administration. These are then bred the following day to have it in the 48 hour available period after peak activity. Table 1 shows there were several cows prior to the addition of GnRH which demonstrated peak activity through the Heatime system appropriately in regards to time of injection. So far, there are also consistent results showing the addition of GnRH may be beneficial, but more data must be reviewed before a conclusion can be drawn about the complete effectiveness. Data from Table 1 does not show peak activity to be

Critical Analysis

After viewing data from a new protocol at the Cal Poly dairy, it is evident that the results suggest this method to be overall efficient. Although not all cows given the injections demonstrate peak activity on the Micro Dairy Logic system, it does not necessarily mean they are not in estrus but, they are not demonstrating estrus activity as clearly as others. The system can therefore only pick up physical estrus, rather than the actual hormonal change. This does not mean the system is not viable, because it still is able to detect activity that may not always be noticed, instead there is not a 100% guarantee that estrus in all cows can be detected.

Table 1: Dates and levels showing estrus activity. Dates from shots given from Dec 30-Jan 6 are from the previous protocol and shorts given from Jan 13- current are from the current program

	Cow #	GnRH	Lut Shot (time)	Estrous Activity	Estrous Peak	Breeding
1	265		30-Dec	1-Jan	13.2	2-Jan
2	397		30-Dec	2-Jan	22.5	2-Jan
3	399		30-Dec	1-Jan	37.5	2-Jan
4	393		30-Dec	1-Jan	10.2	2-Jan
5	2358		6-Jan		no peak	9-Jan
6	2426		6-Jan	8-Jan	26.6	9-Jan
7	410		13-Jan	15-Jan	32.5	15-Jan
8	2303		13-Jan	15-Jan	30+	16-Jan
9	2340		13-Jan	15-Jan	30+	15-Jan
10	2411		13-Jan	15-Jan	43.9	15-Jan
11	322	18-Jan	23-Jan	26-Jan	29.7	26-Jan
12	349	18-Jan	23-Jan		no peak	26-Jan
13	2429	18-Jan	23-Jan		no peak	26-Jan
14	2348	18-Jan	23-Jan	10-Jan	12.3	26-Jan
15	2343	18-Jan	23-Jan		no peak	26-Jan
16	187	25-Jan	30-Jan	2-Feb	24.2	2-Feb
17	203	25-Jan	30-Jan	2-Feb	17.3	2-Feb
18	2266	25-Jan	30-Jan	2-Feb	15.3	2-Feb
19	2354	25-Jan	30-Jan			2-Feb
20	2362	25-Jan	30-Jan		no peak	2-Feb

Table 1: Continued. All shots given on this section of the table are from the current program

21	238		6-Feb	11-Feb	7	9-Feb
22	311		6-Feb	9-Feb	32	9-Feb
23	331		6-Feb	9-Feb	26.5	9-Feb
24	398		6-Feb		no peak	9-Feb
25	405		6-Feb	9-Feb	7.8	9-Feb
26	409		6-Feb		no peak	9-Feb
27	2338		6-Feb	9-Feb	26.2	9-Feb
28	2369		6-Feb		no peak	9-Feb
29	2394		6-Feb		no peak	9-Feb
30	2420		6-Feb		no peak	9-Feb
31	2430		6-Feb		no peak	9-Feb
32	2445		6-Feb		no peak	9-Feb
33	94731		6-Feb		no peak	9-Feb
34	357		13-Feb		no peak	16-Feb
35	399		13-Feb	16-Feb	6.1	16-Feb
36	2359		13-Feb		no peak	16-Feb
37	2414		13-Feb		no peak	16-Feb
38	2422		13-Feb		no peak	16-Feb
39	2436		13-Feb	15-Feb	8.9	16-Feb
40	298	15-Feb	20-Feb		no peak	23-Feb
41	304	15-Feb	20-Feb	23-Feb	12.5	23-Feb
42	338	15-Feb	20-Feb	15-Feb	17.1	23-Feb
43	345	15-Feb	20-Feb	16-Feb	5.8	23-Feb
44	368	15-Feb	20-Feb		no peak	23-Feb
45	2300	15-Feb	20-Feb	23-Feb	21.5	23-Feb
46	2431	15-Feb	20-Feb		no peak	23-Feb
47	2443	15-Feb	20-Feb		no peak	23-Feb
48	2444	15-Feb	20-Feb	24-Feb	2.5	23-Feb

Conclusion

Results have demonstrated that the new protocol where cows receive two injections of Lutalyse, some receive GnRH and are time bred 3 days following the initial injection may show better signs of estrus. The amount of cows demonstrating estrus on the Dairy Logic system are increasingly more than when compared to those that were previously receiving one injection of Lutalyse and then bred 3 days later. Cows observed on the previous program showed to have some cows not registering peak activity on the system, but which may have been in estrus before the breeding date and simply not demonstrated activity. The data has shown that the second shot of Lutalyse ensures that the cow enters the estrous cycle and the GnRH ensures the same in the few cows that do not respond to the initial shot.

Reasons for results demonstrating the high number of cows not registering estrus activity via the Heatime system indicate cows are showing secondary signs of estrus. The Heatime system will only register specific movements and determine they are demonstrating peak levels of activity, which leaves room for error. Biological indications, such as releasing vaginal mucous as well as ovulation cannot be registered by the system because the program cannot detect these kinds of indications.

Results can be more conclusive if more data can be collected on cows that are introduced to the new protocol. Currently, there is more data on previous procedures, which makes it very difficult to discuss trends in the current protocol and have more effective comparisons between the two. Larger numbers of cows to be observed would

also make the study more efficient, but in this case, limited data was the cause for limitations.

Citations

- Boer, H.M.T., R.F. Veerkamp, B. Beerda, and H. Woelders. 2009. Estrous behavior in dairy cows: identification of underlying mechanisms and gene functions. *Animal*. 4:3, 446-453. doi: 10.1017/S1751731109991169
- El- Zarkouny, S.Z., J.A. Cartmill, B.A., Hensley, and J.S. Stevenson. 2010. Pregnancy in dairy cows after synchronized ovulation regimens with or without presynchronization and progesterone. *J. Dairy Sci.* 87: 1024-1037.
- Kommadath, A., H. Woelders, B. Beerda, H. A. Mulder, A. AC de Wit. R. F. Veerkamp. M. FW te Pas, and M. A. Smits. 2011. Gene expression patterns in four brain areas associated with quantitative measure of estrous behavior in dairy cows. *BMC Genomics*. 12:200.
- Peralta, O.A., R.E. Pearson, R.L. Nebel. 2005. Comparison of three estrus detection systems during summer in large commercial dairy herd. *A. Reprod. Sci.* 87: 59-72.
- Peters, M.W., and J. R. Pursley. 2003. Timing of final GnRH of the Ovsynch protocol affects ovulatory follicle size, subsequent luteal function, and fertility in dairy cows. *Theriogenology* 60: 1197-1204.
- Roelofs, J.B., F.J.C.M. van Eerdenburg, N.M. Soede, and B. Kemp. 2005. Various behavioral signs of estrous and their relationship with time of ovulation in dairy cattle. *Theriogenology*. 63:5, 1366-1377.

SCR Precise Dairy Farming. 2012. Heatime Comparison. Accessed Mar. 8, 2012.

<http://www.scrdairy.com/HeatComparison.asp>

Yoshida, C., M. Yusuf, and T. Nakao. 2009. Duration of estus induced after GnRh-PGF_{2α} protocol in dairy heiger. An. Sci. J., 80:6, 649-654. doi: 10.1111/j.1740-0929.2009.00694.x.