A BETTER UNDERSTANDING OF AI

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Take Home Messages

- Analyzing records in a systematic fashion should be the first step in evaluation of the AI program. Percent successful on first service, monthly trends for percent successful on first service, and percent successful stratified by inseminator should be used to evaluate AI technique and fertility level of the herd.
- Four general categories are used to classify the factors that determine the ultimate outcome of conception per insemination: 1) female fertility; 2) male fertility; 3) environmental factors; and 4) techniques used in the AI of cattle.
- Efficient and accurate detection of estrus that determines the timing of insemination, proper semen handling and thawing procedures combined with deposition of semen in the uterine body are skills that determine the outcome of an AI program.

Introduction

The AI of cattle has been important in reducing disease transmission, allowing for genetic selection, and ultimately increasing the health, longevity, and milk yield of dairy cattle. Increased milk yield, dependence on human labor for the detection of estrus, and increased herd size have combined to furnish an environment that challenges management's ability to maintain an acceptable level of fertility. Fertility is a broad category that includes semen quality, accuracy of both semen handling and placement in the female reproductive tract, timing of insemination, condition of the female reproductive tract, ambient temperature, breed, season, nutritional status and recent changes in body condition.

During the past 50 years that AI has been practiced in the U.S. the fertility of virgin heifers has remained relatively constant at approximately 65% first service conception; whereas, the first service conception rate for lactating cows has decreased approximately 33% from 60 to 40%. Conception per insemination is the outcome resulting from a multitude of factors that interact in an intricate fashion. Four general categories will be used to classify the factors that determine the ultimate outcome of conception per insemination or fertility. The four areas that will be discussed are female fertility, male fertility, environmental factors, and techniques used in artificial insemination. Female fertility refers to any factors directly related to the heifer/cow that may alter her probability of becoming pregnant, including condition of the female reproductive tract, nutritional status, changes in body condition status from calving to insemination, age, and breed. Male fertility factors associated with AI will be the primary focus of this section. Environmental factors will concentrate on effects of ambient temperature and techniques involved with artificial insemination will include accuracy of estrus detection, timing of insemination, semen handling and semen placement in the reproductive tract.

Evaluation of Fertility

Analyzing records in a systematic fashion should be the first step in evaluation of the AI program. Often conception problems occur within sub-populations of animals within a herd, which are reflected to varying degrees in the overall herd performance. Comparing "normal" and problem populations can aid in identifying factors influencing fertility. Adequate sample size is important for sound interpretation of reproductive performance especially when dealing with binomial data where pregnant and open are the only two outcomes available. It is necessary to determine if changes in conception are real or just due to a small number of breedings being evaluated. The bottom line is that abundant numbers of observations are required for reasonably precise estimates of fertility.

What is normal? What should be expected as normal conception rate values in dairy cattle? The mean first service conception rate for Florida DHI herds over the past 12 months was 34 ± 13% and the distribution across herds is shown in Figure 1. The major factor influencing fertility in dairy herds is first calving. Heifers have conception rates close to theoretical optimal values. First calving reduces the conception rate 35 to 50%. The question is why this tremendous reduction after parturition. Many herds experience at least 50% of cows calving having one or more postpartum diseases. Typically their chance of conception for cows experiencing problems at calving is half that of normal cows (Lucy, 2001). The conception rate in normal cows and the proportion of normal cows in a herd determine the fertility of the overall herd. Factors such as body condition loss, heat detection errors, and less than optimum semen handling and placement, makes it readily apparent why many herds have conception rates less than 25%.

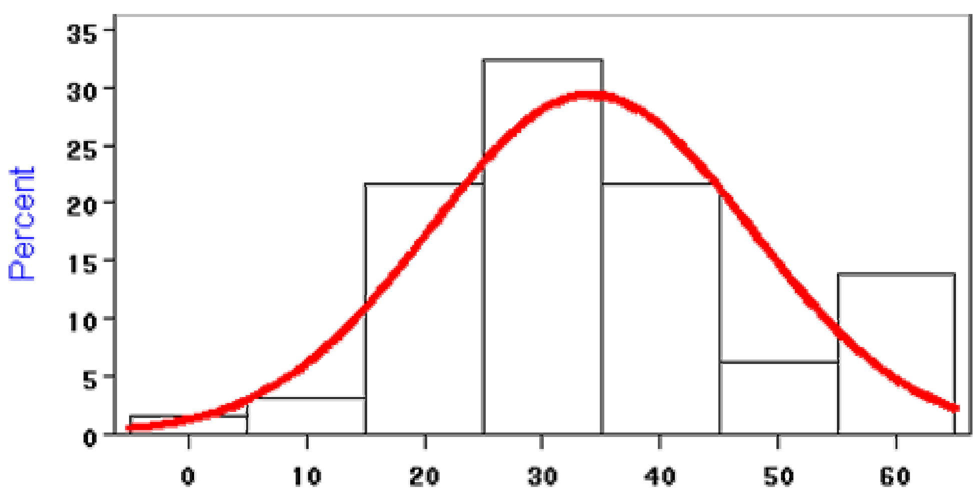

Figure 1. Distribution of the percent successful on first service for Florida DHI Holstein herds.

Environment Effects on Fertility

Fertility in dairy cows is depressed during the summer months in warm areas of the world (Hansen, 1997). This depression is caused essentially by heat stress. Experimental application of heat stress reduced fertility and increased embryonic mortality, while alleviation of heat stress during the summer increased fertility. The magnitude of the seasonal depression in fertility is influenced by environmental factors that define the extent of heat stress and internal factors of the cow that determine her ability to regulate body temperature during heat stress. There is a greater reduction in fertility during the summer for lactating cows than for non-lactating heifers. High milk yield exacerbates the effects of heat stress on fertility. The major reason for high milk yield provoking the effects of heat stress on fertility is related to the increased metabolic rates and decreased thermoregulatory ability for cows with high milk yield. Berman et al., (1985) found that among cows exposed to temperatures within the range of 70 to 92°F, rectal temperature increased 0.03°F for each 2.2 lbs of fat-corrected milk a cow produced above 53 lbs per day.

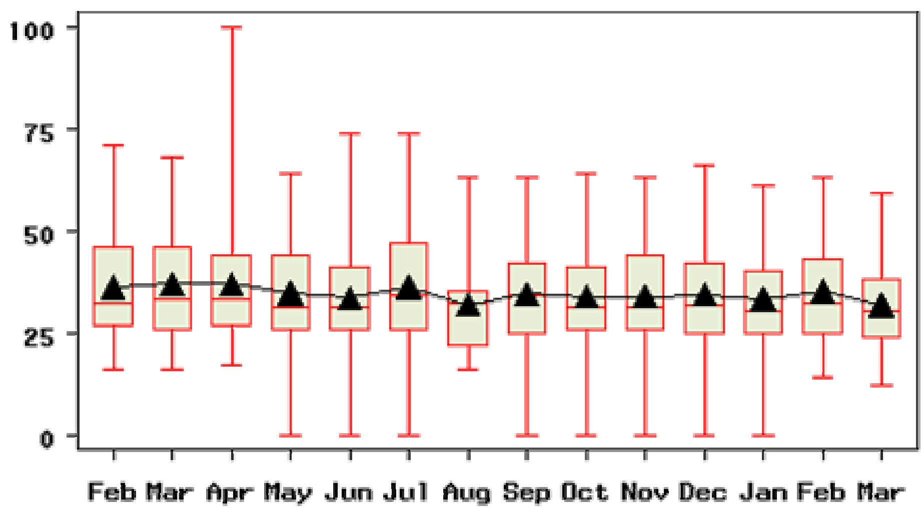

Figure 2. Monthly trend of percent successful on first service for Florida DHI Holstein herds.

Male Fertility

Discussion of fertility differences or influences attributed to the male component will be limited to AI service. Thus, what is the range in fertility of sires available from AI organizations? Estimated relative conception rate (ERCR), is a measure of conception rate of a service sire relative to service sires of herdmates and are calculated by the DHI processing center DRMS in Raleigh, NC. These conception rate summaries are available from the DRMS website at http://www.drms.org and updated twice yearly during May and November.

ERCR is expressed as a percent difference of conception rate from the average AI service sire of herdmates. For example, ERCR values published in the November 2001 summary for Holstein bulls ranged from -5 to +5. Holstein sires summarized between 1998 and 2000 ranged from -11% to +5%. This means that the average service to the most fertile bull produced a calf 5% more often than a service to the average AI service sire of herdmates. Similarly, a service to the least fertile bull produced a calf 11% less often. The average value was 0 and there were approximately equal numbers of positive and negative values. Although the values in 2001 had a range of 16 percentage points, 91% of the bulls were in the range of -3% to +3%. Bulls within this range are of acceptable fertility and most selection emphasis within this group should not be for fertility.

Techniques Involved with AI

This section involves the intervention of man into the actual breeding procedure of cattle. First the female must be accurately identified in estrus, then the timing of insemination becomes the next step in the procedure, finally semen handling, thawing, and placement finishes the process.

Accuracy of estrus detection. Accuracy of estrus detection is defined as the percentage of cows identified in estrus that are indeed in true estrus. Inaccuracies occur when cattle are inseminated at times other than true estrus. Examining the frequency distribution of interestrual intervals has been shown to be helpful in documenting errors in the detection of estrus. Two characteristics specific to estrus detection error are: (1) more than 10% of the interestrual intervals are between 3 and 17 days, and (2) cows are verified pregnant or to calve to a breeding prior to the one last recorded.

In a large field study involving herds in the Northeast U.S., Reimers et al. (1985) reported that 5.1% of the cows presented for insemination were not in estrus based on high milk progesterone levels. The error rate varied from 0 to 60% among herds and 10% or more of the cows inseminated were not in estrus from 30% of the herds. Numerous other studies using milk progesterone analysis have shown that 5 to 15% of cows are inseminated when they are not in or near estrus.

To make the evaluation of estrus detection accuracy with milk progesterone analysis worthwhile, a minimum of 20 cows should be sampled on the day of insemination. When compared with a standard progesterone sample, the milk obtained on the day of insemination should have low progesterone. Progesterone levels are low for about seven days around the time of estrus. Thus, low progesterone indicates the cow is either in or near heat, but progesterone levels cannot be used to precisely time insemination.

Onset of estrus and optimum timing of insemination. Since 1995, the HeatWatch® system (DDx Inc, Denver, CO) has been used exclusively for the detection of estrus at the Virginia Tech Dairy Cattle Center. The distribution of the onset of estrus is shown graphically in Figure 3 for heifers (n = 400) and cows (n = 990). There has been no difference in the onset of estrus for ether cows or heifers, thus an approximate equally numbers of individuals start estrus at any given time. There was a tendency for more cows to begin standing activity between midnight and 3:00 AM which corresponded to the AM milking time and for more heifers to initiate standing activity between 6:00 and 9:00 AM which coincided with the only period of grain feeding the heifers received. In a field study comprised of 17 herds and 2661 estrus periods that used the HeatWatch® system to detect estrus, Dransfield et al., (1998) also reported an equally distribution of the onset of standing activity during the day.

Optimal time of AI was determined from 2661 services performed across 17 herds (Dransfield et al., 1998). Each farm selected a 3-hour interval to inseminate cows identified in estrus during the previous 24 hours. This allowed for inseminations to occur at all intervals from onset of estrus to insemination. Pregnancy status was determined by data for return to estrus and palpation of the uterus for pregnancy determination 35 to 75 days following AI. Time from first standing event to AI affected the probability of pregnancy; highest conception rates for AI occurred between 4 and 15 hours after the onset of standing activity (Figure 4). A higher probability of pregnancy was revealed for cows greater than 100 days in milk, exhibiting more than 2 standing events and inseminated during the months of March, April, or May**.** Timing of insemination should be based on the frequency of detection. A general recommendation for timing of insemination is that when only one daily period is used for the detection of estrus insemination should occur soon after the detection period has ceased, when two observation periods are used approximately 12 hours apart insemination should occur 4 to 8 hours following detection, when three observation periods are used approximately 8 hours apart insemination should occur 4 to 12 hours following detection.

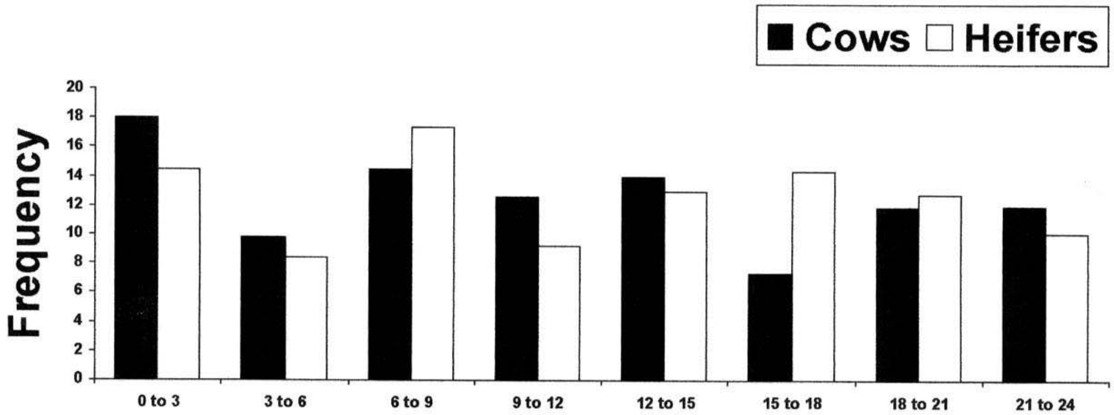

Figure 3. Distribution of the onset of standing activity determined by the HeatWatch® system for cows and heifers located at the Virginia Tech Dairy Centre.

Semen handling prior to thawing. The primary objective in handling semen properly is to conserve the fertile life of sperm until deposition in the female. This is accomplished by minimizing exposure of semen to temperature fluctuations and contamination with other compounds especially water and soap. The semen storage tank is a large, metal, vacuum-sealed liquid nitrogen refrigerator encased within an extremely efficient insulation system. With proper attention and handling, most liquid nitrogen semen storage tanks give years of trouble–free service, but all storage tanks will eventually fail. To ensure maximum holding time, the tank should be kept in a cool and dry location away from direct sunlight, in a clean and well-ventilated area away from drafts, elevated above concrete to prevent corrosion, and where it can be seen daily. Particular attention must be given to the neck and vacuum fitting. Accumulation of frost on these areas indicates that the vacuum insulation has been lost and liquid nitrogen has been evaporating rapidly.

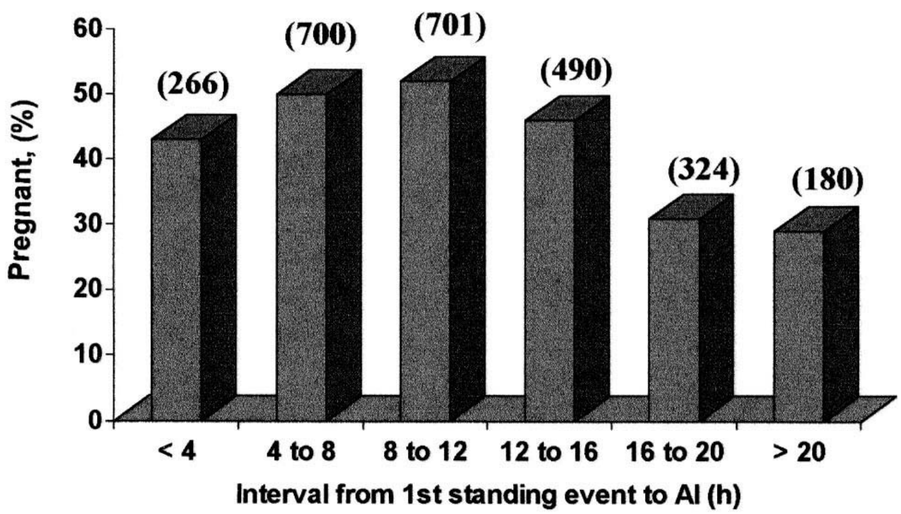

Figure 4. Percentage of cows pregnant by various intervals from onset of estrus to insemination for 2661 services performed in 17 herds using the HeatWatch® system to determine the onset of estrus (Dransfield et al., 1998).

In addition to the obvious error of permitting a liquid nitrogen storage tank to go dry, stored semen may also be exposed to adverse high temperatures when straws are being removed for thawing. An accurate inventory and location of the semen stored in the liquid nitrogen tank is important to prevent exposure of semen during searching for the straw prior to removal for thawing. Thermal injury to sperm is permanent and cannot be corrected by returning semen to the liquid nitrogen. For optimal maintenance of sperm viability, canisters and canes containing semen should be raised into the neck of the tank only for the time required to retrieve a single straw. This time should not exceed 8 seconds.

Semen thawing. The recommendation for thawing of semen frozen in straws is not the same for all AI organizations. For optimal results, the recommendations of the semen processor should be followed. However, the situation on most farms is that semen from many AI organizations is being used and only one thawing procedure is being performed. The National Association of Animal Breeders has recommended that **when in doubt, the straw should the thawed for a minimum of 45 seconds in 90 to 95 °F water.**

Multiple semen straw thawing. To facilitate the insemination of many cows in a timely manner, AI technicians routinely thaw multiple straws simultaneously. It has been reported that using a thermostatically controlled water bath allows batch thawing of straws (up to 10 at a time) without compromising semen quality when the ambient temperature is approximately 70°F (Brown et al., 1991). When thawing more than one straw simultaneously, do not allow straws to touch in the water bath because re-freezing occurs in straws that touch causing sperm injury as seen by decreased motility. Reports by Goodell (2000) and Lee et al. (1997) suggest that thawing more than two straws at once results in reduced conception rates of the third and fourth insemination in sequence (Tables 1 and 2).

Table 1. Retrospective study examining the effect of batch-thawing semen straws on the conception rates in a Colorado dairy herd.

Semen straw	Number of cows inseminated	Number of cows diagnosed pregnant	Conception rate, (%)
1	62	30	48
2	58	24	41
3	53	9	17
4	7	1	14

(Adapted from Goodell et al., 2000)

Table 2. Effects of batch-thawing semen straws on the conception rates in a Holstein dairy herd located in Hawaii.

Semen straw sequence of insemination	Conception rate, (%)
1	48
2	41
3	37
4	25

(Adapted from Lee et al., 1997)

A glaring flaw in the studies by Goodell et al. (2000) and Lee et al. (1997) was that too few cows were inseminated to draw conclusions on fertility (279 cows combined). In contrast, Sprenger et al. (2001) utilized over 5,000 inseminations and reported no difference in conception rates of straws 1 to 6 following batch-thawing by a professional AI technician (Table 3). The thawing of multiple semen straws simultaneously is not recommend for the inexperienced AI technician or when the temperature of the thawed semen can not be maintained near the final temperature of thawed semen (approximately 95°F). Expertise of the AI technician is critical to the success of any reproduction program. **A general recommendation is to thaw no more straws than can be deposited into cattle within 10 minutes.**

Semen handling after thawing. Regardless of what type of water bath is used, all water should be thoroughly removed from the straw before it is cut. Osmotically, exposure of the semen in a 0.5 ml and especially the 0.25 ml straw to as little as one drop of water can result in irreversible cell injury. A concern with warm-water thawing is the danger of cold shock caused by mishandling of the semen following thawing. Cold shock occurs when semen is thawed and then subjected to cold ambient temperatures prior to insemination. Because of potential cold shock, many investigators have questioned the wisdom of the warm-water thaw, particularly when breeding cows in cold weather. Fleming and coworkers (1976) studied the effects of thaw rates and cold shock on sperm viability measurements three hours after initial thawing. Both sperm motility and acrosomal integrity were adversely affected by cold shock when compared to a 95°F thaw without cold shock; however, air thaw and 40°F water thaw were still inferior to the 95°F thaw following cold shock.

Table 3. Effect of batch-thawing semen straws on the conception rates in New York dairy herds serviced by a professional AI technician.

Semen straw sequence of insemination	Number of cows inseminated	Number of cow diagnosed pregnant	Conception rate, (%)
1	1,260	397	32
2	1,250	411	33
3	1,129	386	34
4	917	300	33
5	696	219	32
6	464	133	29

(Adapted from Sprenger et al., 2001)

Semen placement. The highest quality semen placed in the healthiest cow at just the right time will not produce a calf if the breeding technique is not performed properly. The mechanics of passing the insemination device through the cervix will not be covered here. For further information an AI organization that provides AI training should be consulted. Practice is required to develop the skill, which should be learned and periodically reviewed with the assistance of professionals. One of the most critical components of the insemination technique is depositing the semen anterior to the cervix. The major reason why sperm numbers can be markedly lower for frozen and thawed semen used in AI is that the cervix, which is the major barrier to sperm transport, is bypassed in correct semen deposition.

Many studies have compared semen deposition near the greater curvature of the uterine horns with conventional deposition into the uterine body. Although Senger et al. (1988) and Lopez-Gatius (1996) reported increased conception rates when semen was deposited in the uterine horns rather than the uterine body, Williams et al. (1988), McKenna et al. (1990), and Hawk and Tanabe (1986) found no difference in fertility when comparing uterine body and uterine horn inseminations. It is not clear why a few studies have shown fertility advantage following uterine horn insemination while others have not. A possible explanation for the difference may be related to the minimization or elimination of cervical depositions with horn placement. Cervical deposition accounts for approximately 20% of attempted uterine body depositions (Peters et al., 1984). Macpherson (1968) reported that cervical insemination resulted in a 10% decrease in fertility when compared with deposition in the uterine body. Clearly, all AI technicians must develop sufficient skills to recognize when the tip of the AI pipette remains in the cervix. **To maximize conception rates, AI technicians must continue to manipulate the reproductive tract until the tip of the AI pipette is past the cervix and deposition into the uterus can be accomplished.**

Factors Influencing Cow Fertility

Female fertility refers to any factors directly related to the heifer/cow that may alter her probability of becoming pregnant, including uterine condition, nutritional status, recent changes in body condition status, and health. Many research studies have shown that fertility increases as interval from calving to first service increases up to 70 days postpartum. Repetitive estrous cycles are necessary to confer fecundity. Conception rate and estrus expression improves with each sequential estrous cycle from first ovulation to the third estrous cycle. For maximum fertility cows should be on the third or higher estrous cycle when AI commences (Ott et al., 1986). Hopefully, first ovulation will transpire 17 to 30 days postpartum so that the third ovulation will occur by 50 days postpartum when uterine involution and repair will also be complete. Increased negative energy balance may delay first ovulation 60 to 75 days or longer extending the postpartum effects of recovery of the uterine environment.

Postpartum problems. As stated earlier the biggest factor influencing fertility in dairy herds is first calving, but every calving places the cow at risk for metritis, retained placenta, dystocia, milk fever and other metabolic diseases that contribute to the decline in fertility of the population. Typically cows that experience a postpartum problem will have conception rates one half that of normal cows (Lucy, 2001). Ketosis and lameness do not appear to have the magnitude of metritis in reducing conception rates; however, many studies have identified these conditions as having a significant impact on fertility. The impact of retained placenta on conception rate may be dependent upon the development of secondary disease, such as metritis or ketosis. Lameness has had varying effects on fertility and may depend on the time postpartum when it occurs and the severity of the problem.

Metritis and systemic metritis may be perceived as conditions associated with hygiene and stress at calving. Retained placenta, milk fever, uterine prolapse, and grass tetany are directly associated with dry cow feeding and mineral content in dry cow rations. Ketosis, laminitis, fatty liver, and ovulatory dysfunction, particularly anestrus, may be viewed as metabolic dysfunctions associated with energy balance. In addition, excessive body condition loss should be detected as a problem with energy management that will reduce fertility. **The magnitude and duration of negative energy balance depends more on dry matter intake than milk yield.**

Nutrition and reproduction. Minimizing metabolic problems is contingent upon management of energy and protein, not only in rations fed, but also in body tissue stores. Excessive energy or protein in the dry cow diet can create metabolic problems as readily as low energy or low protein diets. Excessive tissue mobilization associated with low dry matter intake in the late dry period is a risk factor for significant parturient problems. In addition mineral supplementation, particularly Ca, Mg, and Se are important in the control of milk fever and retained placenta.

Prevention of excessive mobilization of body fat in the first 4 weeks of lactation is of primary importance for subsequent fertility. Cows will tolerate a loss of approximately 1 body condition scoring unit in the first 4 weeks after calving; more extreme condition loss will predispose her to lower conception rates at first service as seen in Table 4. Researchers in Florida (Moreira et al., 1998) have shown that the pregnancy rates to timed AI was approximately 12% lower for cows with a body condition score less than 2.5, compared to cows with a body condition score greater than 2.5. The mobilization of body fat observed post-calving actually begins prior to parturition, as seen from profiles of serum lipids. One unit change in body condition score represents about 120 lbs of body weight change and about 400 Mcal of energy.

Table 4. Effect of body condition change from calving to breeding on first service conception rate.

Change in body condition score	Conception rate	95% Confidence interval
(X)	(%)	
.75	55.9	49.8 – 61.9
.25	49.5	47.5 – 51.6
0	46.3	38.3 – 54.6
-.25	43.2	41.2 – 45.2
-.75	37.0	31.4 – 42.9
-1.5	28.6	19.5 – 39.6

(Adapted from Ferguson et al., 1996; n = 531 cows, Body condition scale 1 and 5)

Conclusions

Efficient reproduction in a dairy herd is a primary determinant of profitability and the fertility level obtained by a herd is an important component of reproductive efficiency. Factors which determine the fertility levels of a dairy herd are numerous and often complex in nature. Although it may be difficult to diagnose various causes of fertilization failure they are usually related to an AI technique failure or some source of stress experienced by the lactating cow.

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