

What to inspect, when not expecting – investigating infertility in a beef cow-calf operation

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Introduction

In April of 2016 an outbreak investigation was conducted by Mississippi State University College of Veterinary Medicine's Population Medicine Rotation, within the Department of Pathobiology and Population Medicine. A cow/calf producer had requested aid investigating poor reproductive performance in his herd - approximately 150-200 reproductive females with a reported 70% calf crop annually. Though most concerned with 2016, further investigation found that this was a more chronic issue, with sub-optimal reproductive performance for at least the past six years.

The investigation was undertaken to determine the factors leading to sub-optimal reproductive performance in this herd and help provide recommendations that would help the producer reach his production goals. The approach to this outbreak investigation was modeled after methods of field disease outbreak investigation [26-28]. During an outbreak investigation, it is important to determine the case definition; in this investigation any female that failed to calve following a breeding season exposure to a bull was considered a case. We were fortunate to have detailed reproductive records which were utilized to objectively evaluate the problem. Although we knew if cows failed to have a calf, we did not know if they failed to conceive or lost a pregnancy. An understanding of industry standards of reproductive efficiency in beef cow-calf herds is needed in order to develop benchmarks for an individual herd [10,31].

The purpose of this paper is to describe the investigation of sub-optimal reproductive performance in this beef cattle operation and to illustrate how herd health and performance data can be used to develop a causal hypothesis in herd outbreak situations.

Overview of the operation

The herd of approximately 150-200 mature females and first-calf heifers was housed at four separate locations. First and second calf heifers were kept at the producer's home property.

The sizes of the groups ranged from 15 to 50 head; the cattle were of mixed-breed type, with the predominant breed influence being Angus. Bulls used in 2016 were four Angus and one Charolais. New cattle, other than bulls, had not been recently introduced.

The calving season was stated to be mid-January to the end of March, with bulls being turned out for natural service in mid-April. There was no vaccination program in place. Previously, vaccinations had been given, but due to a lack of obvious benefit the producer had stopped. Deworming is completed annually, but the product which is used was not known. The cattle were maintained in a forage-based system of natural grasses. When sufficient forage is unavailable, primarily in the winter months, grass hay is provided. No grain or protein supplementation was provided, high magnesium mineral was available year-round, and commercially available protein licks are provided in the winter.

Detailed production records for each cow were kept by the producer, and this data set was provided for evaluation of current and past reproductive performance. Having these records available was a very valuable tool and enabled the determination of performance trends over several years.

Infertility specific information

In 2016, 32 of 52 (62%) of cows exposed to a bull delivered a calf in the group for which we obtained records. A calving rate of between 60-70% was seen in most of his herd, down to 30% in a group that had been serviced by a bull that was later found to be infertile.

Two bulls were tested by breeding soundness exam (BSE) after the 2016 calving season. Of the bulls tested, one passed and the other failed. The reason for the bull failing the BSE was not known.

The producer noted some cows with low body condition scores (BCS) in the largest, 52 head, group. These thin individuals were moved to one of the smaller groups. One cow died

before calving, and the other calved but never improved in body condition. During the farm visit: the body condition of the cows was assessed and found to be inadequate (2-3/9) for a majority of the herd; no other obvious signs of concern were noted in the cattle or environment.

The two primary concerns of the producer were the occasional cow with low BCS and infertile bulls. Other than these two areas of concern, the producer was unaware of any other potential causes of infertility in the herd.

Data Summary

Records were obtained for the largest of the four groups of cattle (n=52). This dataset was used as a representative sample to make recommendations for the entire herd. The producer had kept records for each female by year. Not all 52 cows had been in the herd for the whole duration (2010-2016). Calving interval was based off of bull servicing dates, April 15th to the end of June, giving an approximate 75-day calving interval. Based on this 75-day cycle and a goal for 95% of cows to calve annually, a calving incidence of 0.58 is needed to reach a 95% calving rate [10,31]. We determined the 58% rate by taking the industry standard 95% annual calving rate and partitioning that into the 3.5 estrous cycle calving interval used in this herd. The start of the calving interval was taken to be the date of the first calf born in the winter/spring, usually mid-January. An assumption is being made which should be mentioned: we are using calving date to extrapolate conception. The data was transcribed into Microsoft Office Excel Software where basic manipulation and statistics were performed along with construction of graphs. Further analysis was conducted in the Epi-Info Software available free online from the Center for Disease Control website.

Chi-square tests of homogeneity were utilized to compare calving incidence by estrous cycle, pregnancy rates, first estrous cycle conception rates, and the effect of calving the year prior on the incidence of calving. The level of statistical significance was set at $\alpha = 0.05$ for

all analyses. Where chi-square tests showed significance a Chi-square for linear trend was run to determine if the pattern fit a linear trend.

Data Analysis & Results

In 2016 there was a significant decreasing incidence of conception over the four 21-day estrous cycles (Chi-square for trend, P-value = 0.001, Figure 1). None of the calving incidences for any of the 21-day periods were equal to or greater than the 58% benchmark established for this herd.

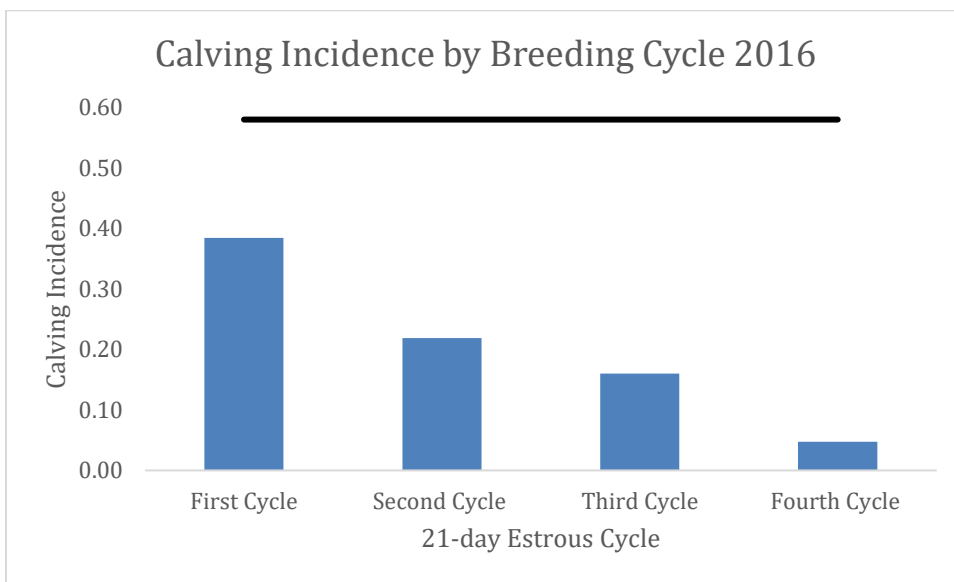


Figure 1 - Calving incidence is shown across four 21-day estrous cycles for 2016. The horizontal bar represents the 58% benchmark needed for 95% of cows to calve within 75-days, P-value=0.001.

Looking across the seven years (2010-2016) a similar pattern emerges as was noted for 2016 (Figure 2). In 2010 no cows calved during the second or fourth periods resulting in only six, rather than seven, columns. The calving rate failed to reach the 58%-benchmark in any estrus period of any year.

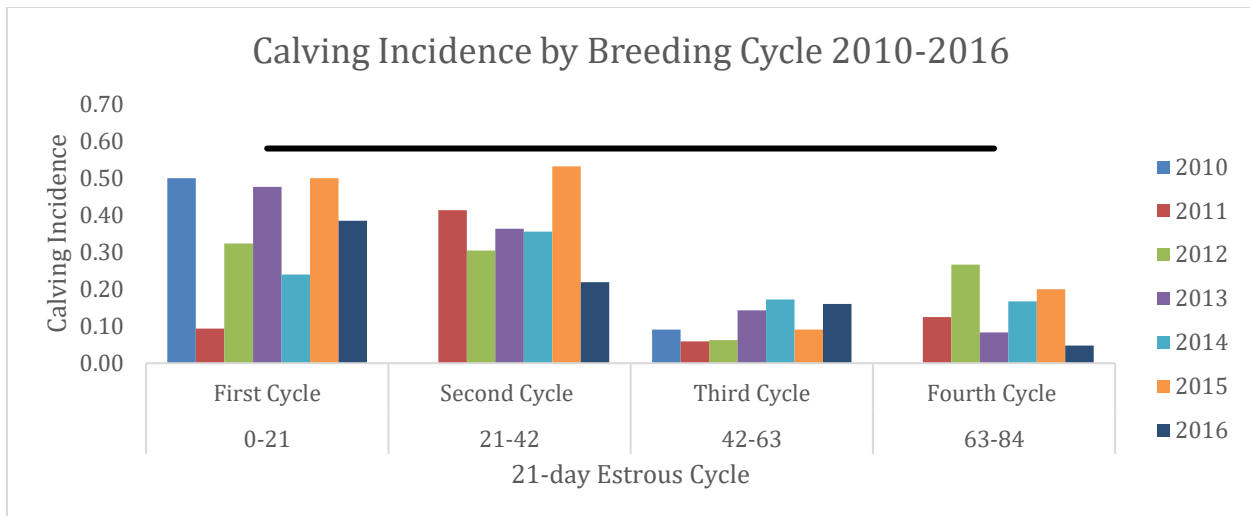


Figure 2 - Calving incidence is demonstrated for the four 21-day estrous cycles for the years 2010-2016. The horizontal bar represents the 58% benchmark needed for 95% of cows to calve within 75-days.

The first estrous cycle calving incidences approach the benchmark of 58% more than any other heat cycle, though it never reached it. There was a statistically significant difference among the calving incidence in the first 21-day estrous cycle by year ($P = 0.001$), but no linear trend ($P = 0.17$) (Figure 3).

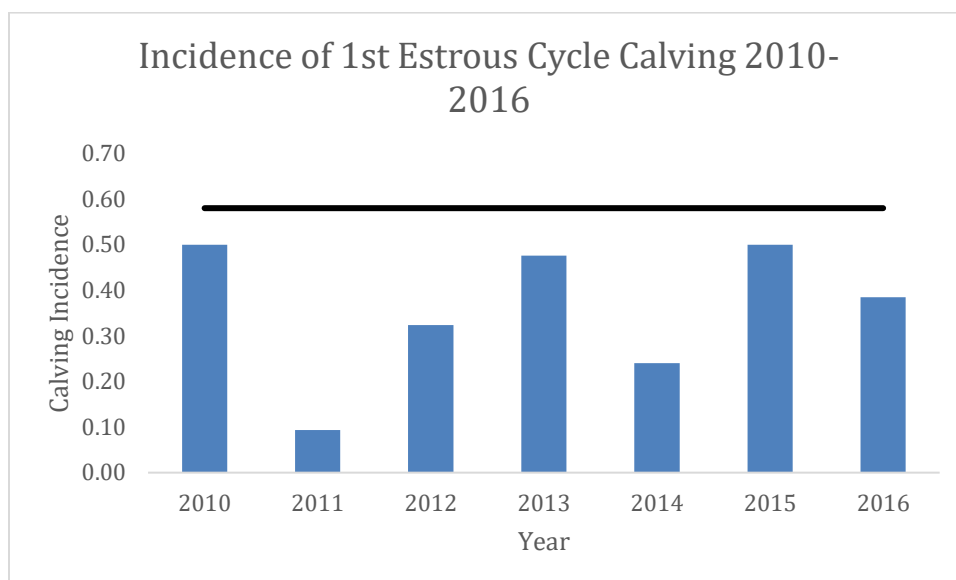


Figure 3 - Calving Incidence for the first estrous cycle 2010-2016. Horizontal bar represents the 58% benchmark needed for 95% of cows to calve within 75-days, $P = 0.001$.

Pregnancy rate is the number of calves born per the number of estrus periods available for conception. Pregnancy rates in this herd differed across years ($P = 0.005$), but no linear trend was noted ($P = 0.40$) (Figure 4).

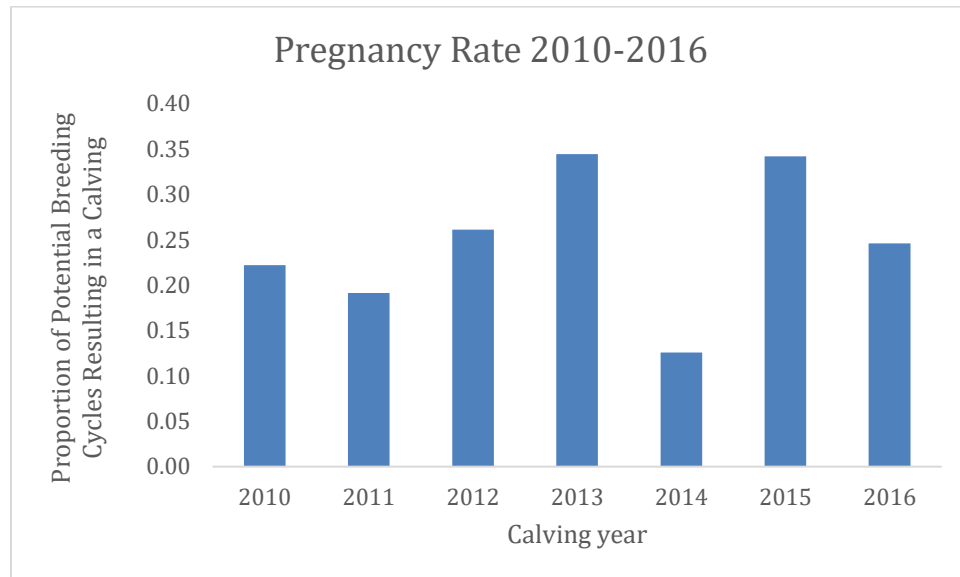


Figure 4 - Pregnancy rates from 2010-2016 showing variation across years, $P = 0.0005$.

The effect of having calved in the previous year on the probability for calving in the subsequent year was tested using a Chi-square analysis stratified by year. Having a calf in the previous year reduced the probability for a cow to calve in the current year (relative risk = 0.70; 95% CI 0.60-0.83; $P=0.002$; Figure 5).

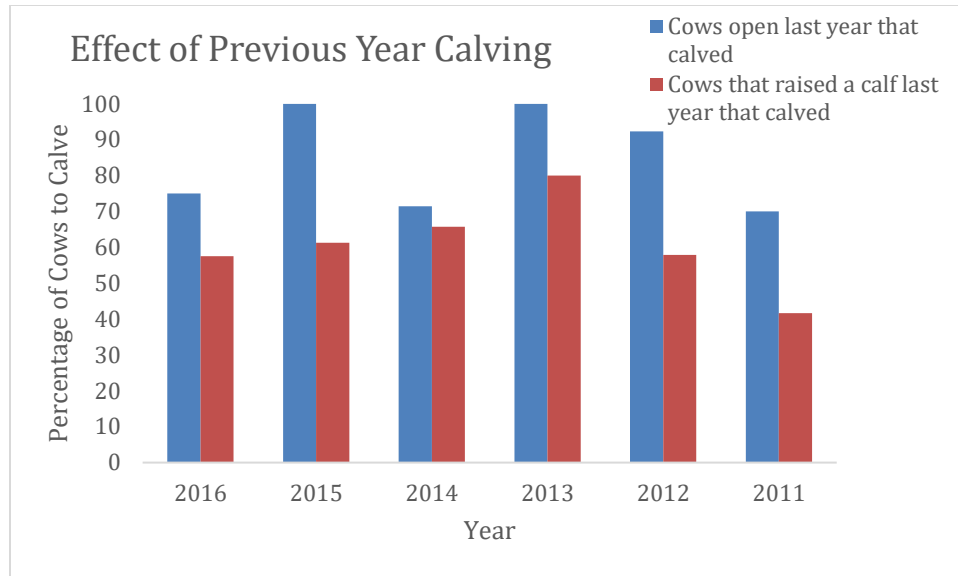


Figure 5 - Percentage of cows that calved after having been open or raising a calf the year prior. There is a difference in the chance of having a calf based on if the cow had a calf the previous year. Risk = 0.70 (95% CI 0.60-0.83) for having a calf if the cow did not calve in the previous year (Two-tailed P = 0.0001).

Epidemiologically Relevant Differential Diagnoses

It is important to approach one's list of differential diagnoses with consideration given to environmental, subject, and disease agent factors [15]. Environmental factors are the conditions in which the animals are living that may affect reproductive potential. These include forage quality, toxins, sanitation, opportunities for infectious disease exposure, stressors, etc. [10,11,25,29]. Subject factors are the characteristics of individuals in the population that may affect reproductive potential. These include age, body condition, and level of immunity against reproductive pathogens. Disease agent factors are those agents, and their level of virulence, which increase the risk of reproductive losses.

Based on the history, observations from the on-farm visit, and results of the data analysis the potential causes of infertility fall into three areas of investigation: causes of reduced bull fertility, causes of cow infertility, and infectious causes of pregnancy wastage. It is certainly possible that more than one of these may be occurring simultaneously. Many causes of poor bull fertility can be identified during the BSE. Causes of cow infertility include body condition and

the importance of energy balance in regard to reproductive success. Relevant infectious disease agents and diagnostic methods will also be covered.

Determining when in the reproductive cycle the losses are occurring would help greatly in narrowing down differentials. Most problems are generally associated with particular stages, especially in regard to infectious disease etiologies, though nutrition and toxins are also important factors to consider that can occur at a particular stage of reproduction [6,7,10-13].

Bull infertility

The chief test used to evaluate bull fertility is the BSE [16,23]. Standardized parameters for a BSE can be found through several sources. In the USA the Society for Theriogenology is the most widely accept [1,8,17]. The purpose of the BSE is to ensure that the bull will be a potential successful breeder. It is important to perform this test close to the start of breeding season to be useful, but also with enough time to allow the purchase of a new bull if there is concern [18]. The focus is on the bull's ability to cover the distance needed to find the cow, physically mount the cow, and deliver live, viable sperm to the sight of fertilization. A thorough physical exam is performed, paying close attention to conformational soundness and structure of the feet [1,23]. Semen is evaluated for motility and morphology [8,17,23]. Further evaluation of the accessory sex glands of the bull should be completed along with measurement and close inspection of the testicles [1]. Bulls may also be tested for common infectious agents, such as *Tritrichomonous foetus*, which can cause bull infertility or more commonly result in venereal transmission to the female [5-7,19].

In this case, a problem with bull power, or sub-infertility, is not supported by the data. It would be expected in a situation with reduced bull power that there would be lower conception rates in the first estrous cycle when there is a larger number of cows for the bull to breed, then as

the number of open cows is reduced through the breeding season the sub-fertile bull would be able to breed an increasing proportion of this smaller total number of cows in estrus. We do know that bull fertility has been a problem in this herd in the past though and should not be completely ruled out. If bull infertility is to blame for this problem, it would have to be that bulls lost fertility later in the breeding season, every year.

Cow nutrition

The cows in this herd have likely had long-term poor body condition, and though the producer did not recognize it as a herd problem, during the farm visit it was evident – with an average BCS of 2-3 noted. The evidence supporting the importance of female nutrition and body condition on reproduction is well established [10,11, 24, 25, 29]. The reproductive female needs to have adequate resources to ensure conception and maintenance of pregnancy is possible. Often one of the largest expenses to the beef cow-calf producer is feed cost; this cost may be associated with supplemental grain and protein, or with maintenance of pasture used for grazing. It is therefore often difficult to get producers to increase these costs for a problem they do not recognize. Negative energy balance, evidenced by inadequate body condition is known to negatively impact fertility and increase the amount of time needed to breed back after calving. The effects of nutritional deprivation on reproductive performance have been extensively studied in dairy cattle but are also well documented in beef cattle [4,10,11,24,25,29].

Supporting evidence from the data analysis includes multiple years of low reproductive performance (Figures 2, 3, and 4). The most telling result is that the cows which calved in the previous year had a decreased probability of calving the subsequent year (Figure 5). This pattern may be an indication that cows which expended more energy raising a calf in the past year were in anestrus and not prepared to conceive the following year. The pattern of decreased calving

incidence as the calving seasons progresses may be explained by the number of cows in too poor of body condition to get bred (Figures 1 and 2). As time progresses, the cows that are capable of cycling have already conceived, leaving those cows not capable of cycling still at risk for conception in the later part of the breeding season. One might expect that with sufficient time cows would increase their body condition allowing for increased conception rates in the later estrous cycles. However, this depends on whether the cows have sufficient opportunity to gain weight. If forage is of inadequate quality or quantity during this period, April to mid-June, then cows may remain infertile.

Infectious losses

Knowing the stage of pregnancy in which reproductive losses are occurring, or if the issue is failure of conception would help narrow down the list of infectious differentials [3,12,15]. To give a detailed report on each of the possible agents that could be affecting this herd is not within the scope of this paper. Based on the absence of observed late term abortions, the patterns seen in calving incidence by estrous cycle, and the multiple years of impaired reproductive performance (Figures 1-4) we might conclude that if infectious agents are involved, it is a disease process that causes early term embryonic loss or failure of conception. Even so this process would have to preferentially affect cows that conceived later in the breeding season. Following is a table showing the most plausible agent from each category (viral, bacterial, and protozoal) and diagnostic methods used for their detection [2,3,15]:

Table 1 - Overview of the most probable etiological agents causing infertility in this herd.

Pathogen	BVDV (Bovine Viral Diarrhea Virus) [20,21]	Campylobacteriosis (<i>Campylobacter fetus</i> subsp. <i>venerealis</i>) [6,14,19]	Trichomoniasis (<i>Tritrichomonas foetus</i>) [5,6,14,19,30]
Type	Virus	Bacteria	<u>Protozoa</u>
Common Signs	infertility, embryonic loss, abortion, weak or malformed calves	low fertility in herds affected	macerated fetus, pyometra secondary to retained placenta
Disease process	viremia then fetal infection (varies by stage of gestation & virus strain)	endometritis leading to embryonic loss	vaginitis to endometritis leading to embryonic loss or abortion
Transmission	transplacental, venereal, oral, contact, <u>persistently infected carriers</u>	<u>venereal (test bulls)</u> , can be oral	<u>venereal (test bulls)</u>
Stage of losses	embryonic loss, first and second trimesters, weak/malformed calves	embryonic loss/early gestation (15-90 days); less commonly 4-6 months	most commonly between 42 and 70 days
Fetal lesions	often autolyzed, lesions difficult to appreciate (teratogenic malformations)	nonspecific, inflammatory infectious process	nonspecific, white flocculant hemorrhagic placentitis
Tissues to sample	fetus, lymphoid tissues, multiple specimen often needed	lung, abomasal contents, placenta	placenta, lung, abomasal contents, uterine fluid

If the problem is due to infectious disease, then the pattern we observed of increased fertility associated with not calving the previous year is suggestive of a process which confers some level of immunity to the cows, after a year of susceptibility - leading to an increased probability of being bred the year after having suffered pregnancy wastage. Trichomoniasis is capable of producing this pattern due to its short-term immunity and primary effects on early gestation [5,6,14,19]. It can infect cows and lead to abortion one year, but possibly provide enough protection to allow them to get bred the following year. It does not fit perfectly though, as it would be expected that the conception rates would be higher in later estrous cycles if this

was the case, as even with immunity from the year prior, cows are thought to have an immune response that would make them less likely to conceive in the first estrous cycle after being introduced to a bull carrying the *Trichomonas* organism [5,6,14,19].

Diagnostic Approach/Considerations

The modern bovine veterinarian has many diagnostic tools available, yet the fact remains that the economics of each investigative measure must be considered [12,15]. If the cost of finding the cause of the infertility is nearly that of the losses due to the infertility itself, then many producers may not be interested. In this case the losses accrued by the producer over the last seven years far outweigh that of paying for an infertility investigation. It is also critical that veterinarians are advocates for these types of investigations and are proponents of their benefits. Often the most important pieces of information in this type of investigation have no inherent cost: getting a good case and production history of the operation and analyzing production records [15]. If any abortions/fetal losses are noted samples should be taken with placenta, fetus, and serum from the cow collected and submitted for diagnostic testing and necropsy examination. The etiologic agents suspected will vary on when the cows are aborting and what signs are noted [2,3,13-15]. The most common infectious agents of abortion vary by region and a good reference is the local veterinary diagnostic laboratory.

It is important to approach the diagnostic lab early that will be used [13]. First, to see what problems are occurring commonly in the area, and secondly to make sure that when an abortion event does occur you will collect the samples necessary [2,13,15]. Looking into past or ongoing infertility issue on nearby operations or the operations from which breeding stock was sourced is also an important consideration.

Often decreased reproductive efficiency will have been occurring for some time before a veterinarian is contacted, and then the producer may look for the most easily implemented

solutions, such as vaccines or other medications. Rarely is this the solution however as management is usually the culprit. Cases of infertility in a beef cow-calf herd can have severe long-term consequences. Late term abortions or retained fetal membranes are easily recognized by producers, but they are more prone to not notice a problem with conception or embryonic loss. The producer may be looking for a simple answer, or solution, and it is therefore important to set expectations and goals at the beginning of the investigation [10,12,14,31]. Reproductive disease investigations can be frustrating; even in cases that present with abortions of suspected infectious cause, a specific agent is isolated only about 50% of the time [2].

Recommendations and Prognosis

The following recommendations were provided to the producer in April of 2016:

- Test all bulls for trichomoniasis prior to turn-out. If any bull tests positive, all bulls should be replaced, because the pathogen is often difficult to culture and is likely being carried in all bulls [1,5,8,14,19,30].
- Pregnancy diagnostics: cows should be checked immediately after bulls are removed to evaluate the conception on first heat cycle, cows should be examined for pregnancy again 40-60 days after pulling bulls to identify all pregnancies, and then they should be checked again 60 days prior to calving [9].
- Vaccinate cows annually against IBR, BVD, Leptospirosis, and Campylobacteriosis. Do so at least 30 days prior to bull turnout [12,13,20-22].
- Monitor body condition and supplement feed to maintain condition during late gestation and lactation and test the feed-value of hay [4,10,11,24,25,29].

Based on the evidence obtained from the history, farm visit, and analysis of reproductive records we have identified a factor most likely responsible for the chronic infertility seen in this herd: inadequate energy in reproductive females. There are likely other contributing factors at play and steps should be taken to ensure bull fertility, proper vaccination, and appropriate general herd health. But the evidence points to body condition and lack of appropriate energy as the primary cause for the poor reproductive performance. The current recommendations for appropriate body condition in beef cows varies by animal age and production system, but a standard benchmark is for cows to be between a score of 5-6 at the time of bull turn-out to

maximize breeding efficiency [10,24,29]. Setting the cows up at the right BCS is a yearlong endeavor because at certain times of the reproduction cycle it is difficult to put weight on cattle effectively, such as during peak lactation and late gestation.

With guidance from their veterinarian, cow-calf producers can learn to assign accurate body condition scores. Numerous factors contribute to cow body condition score, but inappropriate weaning of calves, fertility of pasture and hay meadows, and internal parasite management are some of the most commonly seen issues. A readily available resource for producers regarding beef cow-calf nutrition are state extension agencies - publications are often easily accessed online. Nutritional consulting is a needed service that few veterinarians offer their beef cattle clients. Continued monitoring of reproductive performance as changes are initiated will provide further evidence to support or defame our hypothesis.

Prognosis for return to appropriate reproductive efficiency will depend on the factors responsible for the current infertility. The first step is to improve cow body condition and ensure that the bulls being used are acceptable potential breeders. Next, pregnancy testing should be performed to determine when the largest losses are occurring: at/near breeding, early gestation, or mid-late gestation [7,9]. If nutrition is the primary issue, then once the average body condition is increased to a score of 5-6/9 then improved reproductive performance is expected. The analysis also revealed that a number of cows have been chronic poor producers only having a calf less than 50% of the time they have been in the herd, while there are others that have calved every year, so selective culling may help to remove sub-fertile females. It is important to utilize reproductive data to ensure one is retaining the productive cows and their offspring and culling the poor performers regularly. It is expected that through these two steps: improving average

body condition and culling the worst performing cows that herd fertility will increase to an acceptable level.

To maximize protection from infectious disease vaccination is an important tool, but failures of biosecurity, environmental management, and general herd health will also increase herd susceptibility to infectious disease. Typical vaccination programs focus on BVD, IBR, leptospirosis, and campylobacteriosis, though other agents can be immunized against [6,7,12,13,20-22]. It is not prudent to wait for a diagnosis before starting a vaccination program. The program may need to be altered or supplemented depending on which agents are uncovered but a basic protocol for the region should be begun [6,12,13]. County extension agents and local agricultural universities are an invaluable resource for veterinarians and producers as they often have regionally specific research and recommendations.

Summary

Our investigation found three areas that may be leading to reduced reproductive outcomes, in order of importance: poor female body condition, lack of a vaccination program, and infertile bulls. The recommendations that were provided to the producer in 2016 reflected these major concerns. With our more extensive understanding of the issue, provided by this further analysis, our differentials remain unchanged. However, increased strength has been given to our hypothesis of poor female body condition as the most critical factor in this herds infertility and less to bull infertility.

By ensuring adequate cow body condition at breeding and through gestation and initiating a vaccination program the reproductive performance of this herd will undoubtedly improve. Further evidence suggests that there is a need to cull a portion of cows that are chronic poor performers. If these measures do not result in significant improvement or if abortion events are noted, then the investigation should be continued/redressed as indicated.

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