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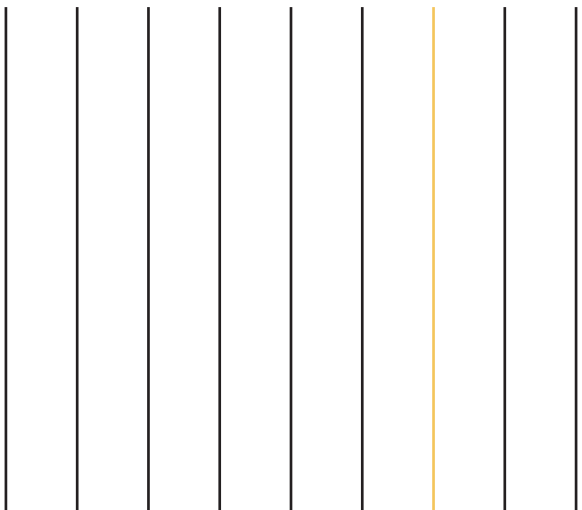

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CONFERENCE EVALUATION

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BOVINE LEUKOSIS VIRUS CONTROL, FARMERS PERCEPTIONS, NEW STRATEGIES & OLD REMEDIES

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LARGE ANIMAL

Bovine Leukosis Virus control, farmers perceptions, new strategies, and old remedies.

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In North America, almost 90% of the dairy and beef herds are infected with BLV with the within-herd prevalence of approximately 40% and 55%, respectively. These numbers are very similar in Canada.
Investigation of within-herd BLV prevalence rates among Canadian dairy herds with the convenience samples revealed a median within-herd BLV prevalence of 40-50%.
The natural hosts of BLV includes cattle (Bos taurus, Bos indicus) and its closely related species water buffalo (Bubalus bubalis), and yak (Bos grunniens). However, experimental infection is possible in diverse hosts including sheep (Ovis aries), goats (Capra aegagrus hircus), and rabbits (Oryctolagus cuniculus). The broad host range of BLV can be attributed to the expression of cationic amino acid transporter 1 (CAT1)/solute carrier family 7 member 1 (SLC7A1) in these hosts' cells which functions as the receptor for BLV.

The transmission of BLV virus can occur horizontally or vertically. A free BLV virus is unstable in the environment, therefore natural infection with BLV occurs primarily by exchange of BLV-infected cells present in the bodily fluids such as milk, blood, and colostrum.

Horizontal transmission
In dairy farms, iatrogenic procedures such as using blood infected needles provide ample opportunity for transfer of BLV-infected cells. Reuse of needles have been associated with increased BLV herd prevalence, indicating the risk of transferring BLV infection being high when injecting a BLV-infected animal followed by a non-infected one with the same needle. Rectal palpation experiment conducted with reuse of palpation sleeves that had been used in BLV-infected animals demonstrated a high risk of seroconversion in BLV-negative animals, as opposed to using new sleeves for every animal. However, single use of needles or rectal palpation gloves failed to reduce BLV incidence in a separate study. This suggests that the invasiveness of the procedure and the extent of infected blood exchange opportunity may contribute to the risk of BLV transfer. Additional potential routes include herd management procedures such as dehorning and tattooing. Use of gouge dehorners without sterilization has been associated with increased BLV prevalence in epidemiological studies. Electric dehorning or sterilizing dehorners after use might minimize the BLV transmission risk.
Hematophagous arthropods may pose BLV transmission risk dependent on the geography and season, which contributes to the insect population and biting incidence. Experimental infection by inoculating mouth parts of hematophagous flies that fed on blood from a BLV-positive cow was able to cause seroconversion in BLV-negative cattle. However, the role of these flies in transmitting BLV under natural grazing conditions remains to be elucidated. Direct close contact between animals is indicated as a risk, however, the exact mechanism involved is not clear. Detection of proviral DNA in saliva and nasal secretions of BLV-

infected cattle indicates that close contact between animals may pose a risk, but this remains to be validated.
Use of semen from BLV-seropositive bulls is not regarded as a substantial risk as artificial insemination with ejaculates from BLV-seropositive bulls failed to induce infection in BLV-seronegative herd. Breeding heifers with BLV-seropositive bulls failed to demonstrate the role of semen in BLV transmission. However, BLV proviral DNA has been detected in the vaginal secretions and smegma of bulls, thus breeding routes cannot be completely ruled out as a risk for BLV transmission.

Vertical transmission
Perinatal transmission of BLV can occur from a BLV-infected dam to its calf and the risk is greater if the maternal BLV proviral load is high. Evidence of identical BLV genomic sequences in the dam and its infected calf highlights the possibility of intrauterine BLV transmission.
The frequency of milk and colostrum-borne BLV transmission was reported to be much lower than other direct contact routes. Intraperitoneal inoculation of leukocytes harvested from the colostrum of a BLV-infected Holstein was able to establish infection in sheep, suggesting the infectious potential of BLV-infected colostrum/milk. However, milk and colostrum from BLV-infected dam also impose a preventive role as these anti-BLV maternal antibodies are detected for 3 to 9 months in calves. This can be confirmed by the results from an in vitro experiment that demonstrated colostrum containing significantly higher antibody titer than serum but lower proviral load than blood. In order to acquire this passive immunity while minimizing the transmission risk, a simple treatment of freeze-thawing of colostrum can be recommended.

Pathogenesis
Bovine leukemia virus causes enzootic bovine leukosis (EBL), and primarily targets CD5+ IgM+ B-lymphocytes, wherein it integrates its reverse-transcribed genetic material into the host's genome, forming a provirus and inducing a lifelong, persistent infection. Additionally, BLV provirus integration in other cells such as T-lymphocytes, monocytes, granulocytes, and mammary epithelial cells have also been reported, however, the tumor cells are only specific to the CD5+ IgM+ B-cells.
Bovine leukemia virus and HTLV-1 are closely related, and often these two models are studied to understand the initial phases following infection with both viruses. In the HTLV-1 model, the viral spread occurs through cell-to-cell contact following entry. After primary infection, the virus replicates either by an infectious cycle or clonal expansion. The infectious cycle involves new target cell infection through cell-to-cell transfer of viral particles, reverse transcription of the viral RNA, integration of the DNA copy of virus into the host chromosome forming a provirus, viral protein expression, and virion budding. The clonal expansion mechanism involves mitotic division of the cells harboring the integrated provirus.
Early BLV infection is characterized by opposing forces: i) BLV favoring proviral integration into genes or promoters leading to clonal expansions, and ii) a massive depletion of the proviruses integrated next to the transcribed regions as a result of increased viral

expression and increased exposure to the host immune response. The interplay of these opposing forces drives the BLV proviral load establishment in the host.

Progression of BLV infection

Classically, the progression of BLV infection was categorized into different stages: aleukemic stage, persistent lymphocytosis (PL) stage, and lymphoma stage. This framework implied that the persistent lymphocytosis and lymphoma stage resulted after a gradual progression of BLV infection and the expansion of blood lymphocytes. However, this theory has been challenged by recent findings which indicate that the persistent lymphocytosis stage can manifest shortly after infection and does not necessarily require a slow, gradual progression of BLV infection. Considering the importance of BLV proviral load in the current BLV diagnostics, it is necessary to understand how quickly the BLV provirus establishes itself following infection and whether the proviral load remains consistent or fluctuates over time. This information is useful for monitoring in BLV control programs. Longitudinal experimental studies have indicated that the proviral load is established shortly after infection and remains relatively stable over time. However, experimental studies may not accurately represent a natural infection due to the variation in the size of the inoculum. A natural BLV infection longitudinal study has suggested that the fluctuations in lymphocyte count over time may not necessarily be a consequence of gradual disease progression, and proviral load does not demonstrate significant increments with time.

Impact of BLV infection

Following a BLV infection, the host's immune system is activated, engaging both humoral and cell-mediated immune responses. This results in persistent antibody production throughout the host's lifetime. However, a gradual reduction in helper T-cells (CD4+) and cytotoxic T-cells (CD8+), along with disruptions in the proliferation and apoptosis of blood lymphocytes, adversely impacts the immune and vaccination responses in the host. The suppressed immune system renders BLV-infected animals more vulnerable to secondary infections. Cattle infected with BLV, exhibiting elevated white blood cells (WBC) and lymphocytes, have a higher incidence of subclinical mastitis compared to BLV-seronegative cows or BLV-seropositive cows with normal WBCs and lymphocytes. The severity of mastitis is also higher among BLV-seropositive cows with high proviral loads. Additionally, BLV infections results in a 30% incidence of persistent lymphocytosis and 5-10% of these lead to cases of lymphoma among infected animals, severely impacting animal welfare.

The assessment of BLV's impact on the milk production of individual animals and at herd level has varied results. Apart from milk production, BLV infection influences cow longevity, with BLV-infected cattle reportedly having a higher likelihood of leaving the herd earlier than their non-infected counterparts.

Diagnosis

A significant proportion of BLV-infected animals (70%) do not exhibit visible clinical signs. In such circumstances, BLV detection becomes challenging without specific clinical tests.

Historically, age-dependent normal reference intervals for lymphocyte counts were established in Danish cattle, to screen for leukemic cattle in 1963. Although this method was successful in eradicating EBL from Danish herds, the application of this method in the current North American dairy herds may be complicated because of the breed, genetic changes, increased production, and environmental differences. Additionally, with only 30% of BLV-infected cattle demonstrating lymphocytosis, relying entirely on lymphocyte monitoring will not detect all infected animals.

A more reliable BLV diagnostic strategy includes detecting the host's immune response against the virus through serological tests and detecting the proviral genome using polymerase chain reaction (PCR) tests. Serological assays, such as radioimmunoassay (RIA), agar-gel immunodiffusion (AGID), and enzyme-linked immunosorbent assays (ELISA), can be used to detect the antibodies against BLV, commonly anti-gp51 and anti-p24, targeting envelope glycoprotein and viral capsid protein, respectively. These antibodies are expressed throughout the host's lifetime following BLV infection. ELISA is reliable and flexible as it can be used to screen various sample types including serum, milk, and colostrum. Commercially available BLV ELISA kits have demonstrated a relative sensitivity of 100% and relative specificity of 95-100%, making ELISA a readily available BLV diagnostic test.

Another BLV detection method involves detection of a segment of the proviral DNA. Various PCR methods such as conventional PCR, nested PCR, real-time quantitative PCR (qPCR), and direct blood-based PCR, have been applied to amplify targeted BLV proviral sites. Experimental studies have indicated that BLV proviruses and antibodies can be detected as early as 24- and 36-days post-infection, respectively. This implies proviral detecting methods enables the identification of BLV infection earlier than antibody detection. However, PCR methods require a complicated sample processing and stringent protocols to avoid cross contamination, which increases the testing cost and cannot be performed without proper laboratory facilities.

The host genome can get integrated with multiple copies of BLV proviruses. Quantitative PCR (qPCR) methods enables quantification of BLV proviral load, which is expressed as the number of BLV proviruses per denominator such as quantity of DNA or endogenous genes. Multiple approaches to quantifying BLV proviral load are implemented, with differences existing in the choice of target BLV gene for amplification, qPCR assays employed, and methods used in proviral load calculation. Categorization of BLV-infected animals into high (HPL) or low (LPL) proviral loads is rendered to be crucial as HPL cows are considered a higher risk of transmitting the virus than LPL cows. Additionally, quantifying BLV proviral load serves as a method to monitor infection status and infectivity in BLV infected animals.

BLV control

Over the years, various control strategies were tried, sometimes organized, but most of those failed due to difficulties in maintaining biosecurity, encountering practical challenges, or just plain 'control fatigue' which comes down to losing interest. None of these programs were mandatory, and generally small scale. It is not easy to maintain a set of best management practices 24/7, 365 days/year, especially when the only perceived benefit is to see a set of numbers (=infected animals) on a sheet of paper (=laboratory

results) go down. Only in highly infected cow herds, observable results could motivate farmers and their personnel, as the number of cows with leukosis will decline (slowly) over time when the prevalence declines as well. This is an important contributing factor to the limited success for BLV control, apart from the fact that it has been shown multiple times (especially for BVDV control) that voluntary control of a livestock virus has a low chance of success.

Which factors could motivate the farmer to actively work on BLV control? Economic incentives are most important. Clarifying to the farmers and the farmer community what a BLV infection level of on average 40% of the herd would mean in lost revenue (decreased milk production, reduced longevity and slaughter value) would be helpful. On the other hand, making clear what the interventions (increased labour, purchase of material, or lower efficiency) would cost paints a more realistic picture in which the pros and cons of BLV control can be weighed. In our economic study, which focused on the 'average farm' in Alberta, Canada, we collected all the economic data and included the potential benefit of BLV control, and the costs associated with interventions in our analysis. Different control strategies were compared, and in all cases BLV control led to a net positive financial result. Apart from the financial motivators, concerns for public health and animal welfare, striving for improvement of milk quality, peer pressure and/or higher levels of education and good realistic information will motivate farmers to increase their efforts for control. Veterinarians and other stakeholders could assist in this process by taking away barriers that could hinder control, assist in design of farm specific intervention or through the provision of incentives for delivering BLV negative milk or meat.
It should be noted however, that identification of the best management practices for BLV control is not always straight forward. Many of the 'logical' transmission routes are still scientifically debated or unproven. Routes such as mixed colostrum provision, reusing rectal palpation sleeves, or using needles for multiple cows are the first that come to mind as a transmission risk, however, it is difficult to quantify their contribution to the spread of this virus.

UPDATES ON BOVINE VIRAL DIARRHEA VIRUS INFECTIONS & CONTROL, A CANADIAN PERSPECTIVE

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Updates on bovine viral diarrhea virus infections and control, a Canadian perspective.

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The virus

Bovine viral diarrhea viruses are enveloped, single-stranded RNA viruses of the genus Pestivirus within the Family Flaviviridae. The genome of BVDV is about 12.3kb. The virus contains both 5' and 3' untranslated regions (UTRs) that flank a single, large open reading frame (ORF) which encodes the viral proteins. The 5'UTR is a highly conserved, non-protein coding region that has an important secondary structure, it is an important target for diagnostic use, most PCRs will detect this region of the genome. It functions mainly as a ribosomal entry site and is essential for infectivity. Similarly, the 3'UTR is highly structured and conserved and plays a vital role in viral RNA replication. The first protein encoded by the ORF is Npro, a viral protease that, along with a variety of host proteases, co- and post-translationally cleaves viral proteins apart and alters the host type I interferon antiviral response. The structural proteins follow and include the capsid protein (Cap) and three envelope-embedded surface glycoproteins (Erns, E1 and E2). The capsid forms a structure around the viral RNA in a mature virion. Erns lacks a transmembrane region and is secreted from infected cells, binds to host cell surface proteins and has an RNase activity. E1 is a surface glycoprotein involved in host cell binding and entry in conjunction with the E2 glycoprotein. Both E1 and E2 contain antigenic sites recognized by the host immune response, however E2 appears to be more dominant in this role. Although Erns and E1 can both induce antibodies, the majority of neutralizing adaptive immune responses target E2. p7, which encodes a protein required for infectivity and produces an ion channel in other *Flaviviruses*, sits between the structural and non-structural genes. The non-structural genes include NS2, NS3, NS4A, NS4B, NS5A and NS5B. NS2 and NS3 perform multiple functions as a single polypeptide (serine protease and helicase) and are essential for viral replication, but their most notable property in the context of persistent infection is how they act as the genetic basis for differentiating cpBVDV and ncpBVDV strains. A wide range of mutations have been shown to result in this biotypic conversion including deletions, duplications and rearrangements of the viral genome, single point mutations and recombination with other BVDV genomes or with host RNAs. It is ultimately the independent expression of NS2 from NS3 that is the hallmark of a cpBVDV strain and therefore to the development of mucosal disease. The spontaneous generation or exogenous introduction of a cpBVDV strain that is antigenically similar to a persistent, ncpBVDV strain in a PI animal will eventually be fatal.

The main host cell receptor for these surface proteins is CD46. Upon host cell binding, the viral envelope fuses with the host cell membrane and ejects its contents into the host cell cytoplasm. The viral RNA is uncoated, and the host cell machinery begins to translate the viral proteins. New virions are produced on the endoplasmic reticulum, transported through the trans-Golgi network, and released from the host cell by exocytosis.

Genetic variability and phylogenetics of BVDV

BVDV is divided into two main genotypes, BVDV1 and BVDV2 with each being further divided into sub-genotypes. Originally the naming of pestiviruses was based on the area or animal species the virus species was discovered in, however, the increase in new discovered viruses of this group made it necessary to come up with a new nomenclature: *Pestivirus* A, *Pestivirus* B (BVDV type 1 and 2), *Pestivirus* C etcetera. It should be noted that 21 *Pestivirus* A subtypes (BVDV1a-u) and 4 *Pestivirus* B subtypes (BVDV2a-2d) are discovered thus far. There is considerable genetic variation both within and between sub-genotypes. Furthermore, while numerous sub-genotypes of BVDV1 exist there are generally only a few found in any given geographic region. This genetic variation has implications for the phenotypic differences between viral isolates with antigenic differences being of particular importance with respect to vaccine development. In North America, BVDV1a, BVDV1b and BVDV2a circulate widely with BVDV1b likely being the most prevalent.
PI animals are integral to the ongoing transmission of BVDV but their role in driving genetic diversity is not clear. Although they are known to generate and maintain herd specific strains it has also been found that the population of viral genomes within a single, PI animal is highly diverse. The interaction between the refining selection of the PI animal's immune system and this diversity is not well understood but it may play a role in the ongoing evolution of BVDV.

Transmission and clinical implications

Animal production systems face many challenges with respect to raising and maintaining economically viable animal populations. One of the chief concerns of these operations are diseases which can significantly impact the production potential of animals and, as a result, the bottom line of the operation. Diseases leading to morbidity in a population can be challenging to address, particularly when infections remain subclinical and difficult to identify without laboratory diagnostic testing. BVDV is a viral infection of cattle found worldwide in both dairy and beef cattle operations where BVDV is a major production limiting pathogen. Although the precise behaviour of this pathogen will vary due to the differing nature of dairy and beef farms, BVDV has a notable and negative impact in both situations.

Although there have been cases of BVDV outbreaks associated with high mortality, fortunately this is uncommon. Most infections are subclinical and can circulate relatively unnoticed on a farm for years. Most infections are transient in nature with less than 1% being persistent. Transient infections result from the transmission of the virus from an infected host to a susceptible host. These mostly occur horizontally but may also result from vertical transmission *in utero*. These infections result in a viremia of approximately two to three weeks followed by the development of a robust, neutralizing immune response that clears the virus and produces a long-lasting immunity against the infecting strain. While viremic and during recovery afterwards the animal will experience the production limiting effects that make this such an important pathogen. These may include reduced performance metrics such as milk production and daily weight gain. Immunosuppression

is particularly noteworthy since it can lead to a variety of secondary infections. The most notable is probably bovine respiratory disease (BRD) which has a significant production limiting effect as well as mortality, particularly in feedlots. Although both biotypes have immunosuppressive capabilities, non-cytopathogenic BVDV (ncpBVDV) appears to be more potent than cytopathogenic BVDV (cpBVDV) in this regard due to the lack of type I interferon synthesis during infection. This immunomodulation of the host is proposed to play a key role in establishing persistence by supressing both the innate and adaptive immune systems. It may also be partially due to differential host cell tropism of the biotypes. Transient infections are responsible for the majority of economic losses on a farm since they represent the bulk of infection. However, on their own they would not be able to perpetuate BVDV infections for years. The virus would infect all susceptible animals and the resulting adaptive immunity would protect the herd against reinfection with that strain. PI is necessary for the long-term maintenance of BVDV in a herd.

Persistent infections

PI is the result of an *in-utero* infection with a ncpBVDV strain during the first ~125 days of gestation. During development of the fetal immune system, the viral antigen is recognized as a self-antigen. The resulting calf is immunotolerant to the infecting strain of virus and will not produce an immune response capable of clearing the infection. While other outcomes are possible from *in utero* transmission of BVDV (transient infection, abortion, and mummification), PI is the most notable and epidemiologically important. Immunotolerance of PI animals is the hallmark of such infections. These animals have very similar immune cell populations (except during mucosal disease (MD)) and have functional antigen presenting cells but seem to have a strong BVDV tolerance in their CD4-positive cells. Although they may produce adaptive immune responses to BVDV strains other than the initial one they are generally very permissive to BVDV infection. As such, the calf will develop a blood serum viremia of between 10^6 and 10^7 $TCID_{50}$/mL that can vary throughout their life. This viremia fuels high levels of viral shedding (10^4 $TCID_{50}$/g of feces and 10^6 $TCID_{50}$/mL of mucosal secretions) which continuously challenges the rest of the herd with BVDV and drives ongoing transient infections. Although the PI animal's immune system can respond against BVDV strains that are antigenically distinct from the infecting strain and therefore refining the population of viruses in the host, these animals also seem to act as a source of novel viral variants that may contribute to evading the rest of the herds adaptive immune responses and re-infecting them. In this way, a very small number of PI animals (often <1% of a herd) can maintain BVDV within a herd indefinitely.

PI animals are not easily identified in a herd but generally do not perform as well as their immunocompetent peers. As with transient infections, they have poorer performance metrics and are more susceptible to secondary infections that can lead to premature mortality. They are also capable of spontaneously developing fatal MD. MD results from the introduction of a cpBVDV strain that is antigenically similar to the persistent, ncpBVDV strain. The cpBVDV strain could be exogenous (from a vaccine for example) or due to spontaneous mutation of the ncpBVDV strain. MD is characterized by the development of lesions along mucosal surfaces and the digestive tract, diarrhea, and weight loss. It is

typically fatal within about two weeks of the appearance of clinical signs. PI animals do not always develop MD and can live to reproductive age. PI dams will produce PI calves themselves, resulting in multiple generations of related PI animals. PI bulls may also have BVDV in their semen which can induce PI in calves following breeding or artificial insemination. In summary, PI is essential to maintaining BVDV infections in a herd and plays an integral role in spreading the virus.

Vaccines

The main aspects of successful BVDV control programs include biosecurity, virus elimination and ongoing monitoring. Specific measures to control and eradicate BVDV revolve around first eliminating sources of infection in a herd and then preventing BVDV from re-infecting the herd. Clearing BVDV from a herd in regions with high infection rates relies on vaccination and identification and removal of PI animals. Most vaccines in use today are modified live, multivalent vaccines. In addition to a variety of other pathogens, the vaccines commonly used in North America contain both BVDV1a and BVDV2a strains. The primary goal of vaccination is to prevent a PI from emerging. To avoid the accidental induction of PI through the use of vaccines if a pregnant dam is vaccinated, and to elicit better antigen presentation, cpBVDV strains are usually used in vaccine production. There have also been several ncpBVDV vaccines that are protective as well, although they do not seem to be widely used. The differential immune responses resulting from Th1/Th2-like regulatory mechanisms to different biotypes also have important implications for the choice of biotype in a vaccine. Most of these vaccines result in a robust adaptive immune response against the target strain and heterologous strains to varying extents. They also yield a net positive economic benefit to the vaccinating farm although this will vary significantly from region to region.

The use of cpBVDV strains in vaccines to eliminate PIs from a group of animals (for example in feed yards) has thus far always been unsuccessful. It is not possible to induce MD using vaccines in all PIs that are present in a group of calves.

Larger scale, phylogenetic studies utilizing isolates collected over years and from across Canada demonstrate a marked diversity of viral isolates. This is true both with respect to the genotypes and sub-genotypes in circulation as well as the variation observed within these genetic groupings. The identification of BVDV1a, BVDV1b and BVDV2a in Canada emphasizes the need for vaccines to address all three sub-genotypes. While multivalent BVDV1a/BVDV2a vaccines are common in Canada, they rely on potentially suboptimal cross-protection against BVDV1b. Given the number of BVDV1b isolates identified through convenience sampling in our studies and the high prevalence of BVDV1b found using more robust sampling methods there is a pressing need to design vaccines that more explicitly protect against the diversity of viruses currently in circulation.

Other control methods

While vaccination is an effective tool for reducing economic losses at the herd level and can be useful if properly implemented, it must be used in combination with other tools as it is not capable of eradicating BVDV alone. This is largely because vaccines do not

always produce a sufficient immune response towards BVDV to entirely prevent vertical transmission of the virus and the genesis of new PI calves. Although the herd will experience a reduced burden from the resulting PI calves thanks to the vaccine-induced immunity, there is a high probability that the virus will persist in the population. To fully eradicate BVDV on a farm, animals must be tested for viremia and positive animals re-tested at least two weeks later to confirm PI. These animals must be removed from the herd and biocontainment barriers put in place. Biocontainment measures are also integral to successful eradication campaigns. A combination of tools over long periods of time is required to declare a region or farm BVDV-free.

BVDV in heterologous hosts

Bovine Viral Diarrhea Virus infections were discovered in sheep, swine, goat, many wild herbivores (deer, moose etc), camelids, but also rabbit and hare for example. The epidemiological contribution of these animals to the virus maintenance in populations is unclear. It can be expected that BVDV PIs in any wildlife species will have a difficult time surviving long enough to contribute to the circulation of the virus in the wildlife population. Any domesticated animal that can be infected and excrete the virus have a higher probability infecting the bovine herds, but good data on these 'spill back' infections is lacking. Many species of wildlife have pestivirus antibodies, however, most of these are derived of one of the 8 Border Disease Virus species. It should be noted that in the past the circulation of BVDV in swine has provided challenges to correctly diagnose Classical Swine Fever Virus infections during outbreaks, and several herds were eliminated due to the cross reactivity of BVDV and CSFV induced antibodies. Currently more specific tests should be able to avoid those situations.

IMPROVEMENT OPTIONS & DEVELOPMENTS IN BOVINE VACCINES & VACCINATION STRATEGIES

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Improvement options and developments in bovine vaccines and vaccination strategies

This text is not meant to discourage the use of vaccines, but to discuss the best strategies of vaccine use in bovines and provide realistic expectations about their contribution to infectious disease management in beef and dairy herds.
It should be noted that vaccination is not the same as immunisation. The administration of a vaccine doesn't mean that the animal can immunologically respond, will be able to resist an infection with a pathogen, or even that the disease associated with that pathogen will be milder or absent when it gets infected. Therefore, the choice of vaccines, the optimization of the circumstances for the animal to respond to a vaccine, the timing of vaccination(s), and the reduction of the risk of getting infected, amongst other things, need to be considered to ensure the best outcome possible. The best outcome could be 'prevention of disease' whereby infection is still possible, but the animal is not getting a disease, or is less affected, or 'prevention of infection' which would be ideal.

As probably every veterinarian knows, the AVMA and other organisations have identified a set of 'core' and 'non-core' vaccines.

The core vaccines are

- Infectious Bovine Rhinotracheitis virus (IBRV) (Bovine herpesvirus 1)
- Bovine Viral Diarrhea Virus (BVDV)
- Parainfluenza Virus (PI3)
- Bovine Respiratory Syncytial Virus (BRSV)
- Clostridial Vaccines (*C. hemolyticum* and *tetani* are not considered core, but are considered risk-based)

These vaccines are our best defensive tools we currently have in our toolbox. However, there is significant room for improvement, and specifically to improve the knowledge of the diseases and the pathogens. Further studies on the role of these pathogens and the interaction with their hosts are necessary to better understand the way interventions could be implemented and improved.

The BHV1, PI3V and BRSV viruses all infect the respiratory tract at different levels and with various severity. While BHV1 can also cause a systemic infection leading to for example abortions or mastitis, it is not uncommon to see reproductive tract pathology depending on the tropism of the infecting strain. Vaccinating against BHV1 will have a positive effect in the prevention of reproductive tract ailments, but the respiratory tract is not sufficiently protected. Studies have clearly indicated that BHV1 vaccines will not prevent outbreaks but could aid in the prevention or mitigation of clinical disease. PI3V and BRSV are almost exclusively infecting the lung tissue. The contribution of PI3V to the development of the bovine respiratory disease complex is far from clear. Therefore, the level of protection derived from PI3V vaccines is also a matter of debate. Most cows will be antibody positive

for this virus, without having displayed any type of disease that can be associated with this virus. The interaction of this virus with bacterial pathogens are also poorly studied. BRSV however, can cause calf mortality and severe clinical outcomes. Its role in BRDC pathogenesis is again not very clear.

As a rule of thumb, vaccines for respiratory tract pathogens are not necessary the most effective, probably the induction of a systemic antibody (IgG) or T-cell responses through vaccination doesn't always prevent damage to the cells and we should focus on other components of the immune system to reach a higher level of protection. There is a good reason that intranasal application of respiratory vaccines is explored, you would like the immunity to be effective at the port of entry, the first place where virus replication will take place and damage will happen.

Clostridial vaccines are in many ways different, they contain the toxoids that induce the pathology, these vaccines are very effective, and immunity is long lasting, in contrast to the 'old' bacterin vaccines. Longitudinal studies on how long this immunity last exactly are lacking, but if we compare those to the human or equine tetanus vaccines, we can expect many years' protection after the initial completed series of vaccines and boosters (these are killed adjuvated vaccines, they require a booster). How many the 'X-way' clostridium vaccine should be required in the vaccination schedule depends on the specific situation of the farm, where the farm situated, history of disease, which animal group/age needs to be protected etc. When there are indications that a certain Clostridial species is causing clinical problems, that species need to be included. Boosting these vaccines yearly doesn't harm but is probably not always needed.

Provision of any 'non-core' vaccine is depending on farm-specific situations. Location, time of year, age of animal, immune status of those animals, availability of labour and funds, production goals or believes of farmer and many other factors should be considered. Some of those factors are easy to understand and incorporated in a vaccination strategy, others are less well understood or simply cannot be changed.

One of the most applied, non-core vaccines are the vaccines against scours. The same limited vaccine protection that was indicated for respiratory pathogens can be expected when GI tract pathogen vaccines (such as rota-, coronavirus) are used. Scours vaccines provide protection in a different way. Vaccinating the dam, to prevent diarrhea in the calf is complicated. Using this strategy variation can be expected, for example in the response of the vaccinated cow, the type and quantity of antibody that will be transferred to the colostrum, the uptake of that 'enriched' colostrum by the calf, the transfer to and circulation in the blood of those antibodies and lastly the amount of antibodies that ends up in the GI tract of that calf. Currently, we are performing a study that will evaluate all aspects of this vaccination strategy, identify what goes well, what needs improvement, and what is a reasonable expectation when animals (beef or dairy) are vaccinated. Timing in these cases is of the essence and not always easy to organize or predict. Many factors can influence the outcome: the 'booster' is sometimes forgotten or badly timed (it takes time to

make an antibody), the amount of colostrum ingested by the calf is insufficient or the pathogen level in the environment of the calf is too high etcetera. These are non-vaccine related factors, apart from the fact that most vaccines do not contain the most recent circulating virus strains.

The emerging epidemiological situation will determine if dairy farmers need to start vaccinating against for example leptospirosis. *Leptospira canicola, L. grippotyphosa, L. hardjo, L. icterohaemorrhagiae,* and *L. pomona* are incorporated in these vaccines. These strains are associated with abortion and infertility, but we haven't noticed a wide spread of these pathogens in the western Canadian cattle populations at this moment. They seem to slowly move northwards and are noticed sometimes when causing zoonotic infections. Especially with imports of animals from the south the risk of establishing these pathogens in the Canadian cattle herds is real. Generally, the immunity is relatively short lived, so frequent boosters are necessary, whereby vaccinations in the spring will provide the highest level of infection in the period of expected challenge (summer/autumn).

Despite all the opportunities and advances in vaccinology, we must always keep in mind that the success of a vaccine is dependent on 1. the quality of the vaccine, 2. the ability of the host to respond to the vaccination, 3. the willingness of the veterinarian and the farmer to apply the vaccine and 4. to do this in the correct way.
For 1 and 4 protocols and SOPs will be available to assist us, the producer of vaccines needs to prove that the product is what he/she promised, and the way the vaccine is delivered can be described in detail. The ability of the host to mount that needed immune response that turns a vaccination into an immunisation is surrounded with many variables. Sometimes due to controllable (stress, age, deworming, hygiene) or uncontrollable (genetics, weather) circumstances the immune response following vaccination can vary enormously. Another big known-unknown is the willingness of the farmers and veterinarians to use a certain vaccine. The human factor in the willingness to use a vaccine cannot be underestimated.

New Developments in Vaccines and Vaccination Strategies, what can be Expected in the Next 5-10 Years

Every intervention will have limitations, vaccination is no exception. Therefore, new strategies are being developed and different targets explored. Up till now following isolation of the pathogen, either killing, attenuation or utilizing a component of the pathogen was used to develop vaccines. Some of the pathogens are difficult to culture, others are very variable and therefore may require different vaccination approaches that are more adaptable. The high variability of certain pathogens, or the limited availability of vaccines drove the need for 'farm-specific' vaccines, derived from a pathogen that is known to circulate in a region or farm. Although a very attractive option, culture of these pathogens is required and only killed vaccines can be produced this way. An adjuvant is therefore required which can lead to side effects, and boosting with the same vaccine will

be needed for a robust, lasting immune response. Efficacy of this strategy is regularly questioned, and not without reason.

Outlook into the not-so-distant future:

A new trend is the so called 'platform' vaccines which are making an entrance in the veterinary world, adaptable backbones that can express a piece of the pathogen (the piece that induce an immune response) could be a new viable solution to the limitations of conventional vaccines and may be able to provide a more predictable immune response compared to the farm-specific vaccines. In human vaccinology this strategy is extensively used to provide quick updates for seasonal influenza vaccines. There is no reason to only restrict its use to these pathogens.

One of these platforms that is approved by the USDA is a baculovirus (=silkworm infecting virus) expression system which is used to produce adaptable vaccines (about 12 weeks from genome sequencing to vaccine). This system can produce many different pathogen proteins, and once a baculovirus+ foreign gene construct is created (and approved), it can be easily adapted to allow for a strain specific vaccine development. The result is a subunit vaccine which needs an adjuvant and requires a booster.

A versatile approach is the use of viral vectors, pox-, herpes-, and adenoviruses (these are all large DNA viruses) which are extensively explored whereby canarypox and fowlpox vectors are already used in veterinary vaccinology. Basically, a weakened large DNA virus that can accommodate foreign DNA can be used to express a gene of interest from another pathogen. Multiple foreign genes could be integrated in such a vector, so a multivalent vaccine may not be necessary anymore. Although it is a modified virus, it may be able to replicate, depending on the targeted host. For example, a canarypox vector doesn't replicate well in a mammalian host, but an adenovirus from a mammal might have that ability. Also, immune responses against the vector could have consequences for the possibility to boost with this same vector.

An example of a target that could make its entry soon in the cattle industry is the use of 'immunocastration' vaccines, based on gonadotropin-releasing hormone (GnRH). The release of this small protein that regulates the production of FSH and LH is the initial step in the hypothalamic–pituitary–gonadal axis, ultimately leading to the release of testosterone in male animals. The acceptance of surgical castration without anaesthesia by the public may diminish, especially when effective, non-surgical, pain free methods are available. These vaccines have made their entry in pig production, however, also here some getting used to (testicle containing boars are slaughtered and processed) will be required. Public acceptance of this technique will drive its future use.

Interesting strategies for vaccination against enzootic bovine leukosis are developed and tested in Argentina, many regulatory hurdles need to be taken before this promising approach also can be used in North America. In principle the cow will be infected with an

attenuated BLV provirus which is created by deleting genes dispensable for infectivity but required for efficient replication. Once infected with this vaccine virus, and the provirus is present in the cell, no super-infection with a BLV wildtype strain is possible. The vaccine virus is not excreted; therefore, transmission of this vaccine is not taking place. Another tool in the control of this virus.

New adjuvants for existing and new vaccines are explored. Apart from the widely used Alum salts, emulsions in various forms O/W, W/O, WOW and for example saponin molecules, new developments in Toll like receptor agonists or cytokines that can direct the immune response in specific ways are studied. With the ever-evolving knowledge about immunology, targeting the innate components of the immune system could provide a better response with less side effects.

Another trend that soon will make its entrance in cattle vaccination are biodegradable polymeric nanoparticles that can be constructed from organic or inorganic materials to mimic for example a virus. Molecules can be delivered that self-assemble in to 'empty' viruses (viral like particles), which do not contain genetic material, hence, cannot replicate. These are easier to produce and cheaper than conventional approaches.

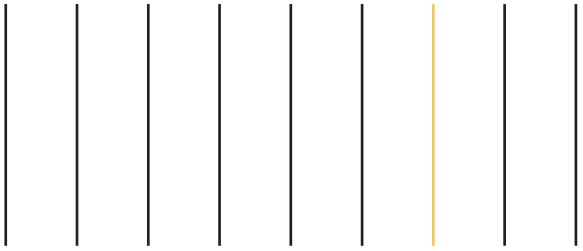
Nucleic acid-based platforms are by far the most versatile and have enormous potential. Both DNA or mRNA-based methods are currently developed and, in a few instances, already used in veterinary vaccinology. In swine production systems mRNA techniques are used against influenza- and rotavirus, and it will not stop there. This mRNA platform holds a lot of promise and there is substantial improvement expected in their efficacy, the duration of immunity and their practicality for use on farm. Despite the very mixed reviews by many experts and non-experts, this type of vaccine will create a way to quickly create a vaccine in case of outbreaks with new or emerging pathogens and enable the development of vaccines for difficult to culture pathogens. These mRNA vaccines generally include adaptations in the genomic material to ensure the longer persistence in the cell (several days). Normal mRNA molecules are very quickly degraded (within hours), they are only used once and recycled quickly thereafter. Also, these molecules may be recognized by the innate immune system, and cleared before they can produce the proteins that should trigger the adaptive immune response. To generate an even higher amount of protein, so called self-amplifying mRNA molecules are under development (saRNA) which should in theory provide a more robust stimulation of the adaptive immune system.

Use of DNA/mRNA vaccines will drive the use of needle free application devises (which is very good news for BLV control), but their use obviously doesn't have to be limited to these platforms. Most needle administered vaccines are delivered in the muscle although these tissues do not contain many cells that can process those antigens and initiate an immune response (such as dendritic cells). Delivery in, or just below the skin has many advantages, the targeting of specialized antigen presenting cells such as the dendritic cells (DC) could lead to a very efficient induction of immune responses. Dendritic cells are a class of 'professional antigen presenting cells'. When these cells are specifically targeted a higher

level of response can be expected: DCs and macrophages differ in their capacity to digest antigens. Macrophages endocytose antigens and rapidly digest them. In contrast, DCs sequester and preserve the captured antigen for later presentation. DCs initiate T cell immune response in the lymph nodes and spleen. Apart from these actions, through the excretion of cytokines and other chemicals to regulate the immune response. Targeting these cells could be very beneficial for the level and type of immune response that will be induced.

IDENTIFYING & MANAGING PREDATOR ATTACKS ON LIVESTOCK

DREW RICKETTS
BS, PHD



LARGE ANIMAL

Identifying and managing predator attacks on livestock

- Livestock predation statistics in the US and KS
- Evaluation of suspected predator attacks
- General characteristics of predator attacks
    - Coyotes
    - Dogs
    - Mountain Lion
    - Predatory/Scavenging Birds
- Commonly misidentified injuries
- Reporting procedure
- Connecting producers with assistance
- Managing livestock predation in KS
- Resources

KEEPING UP WITH ESTRUS SYNCHRONIZATION SYSTEMS FOR BEEF COWS & HEIFERS

SANDY JOHNSON
PHD

LARGE ANIMAL

Keeping up with estrus synchronization systems for beef cows and heifers

Sandy Johnson, PhD
sandyj@ksu.edu
June 2, 2024
Manhattan, KS

K-STATE Research and Extension

Outline

- Resources
- Sexed Semen
- Protocols
- Docility & Fertility
- Heterospermic semen packages
- Rapid rebreeding/resynchronization

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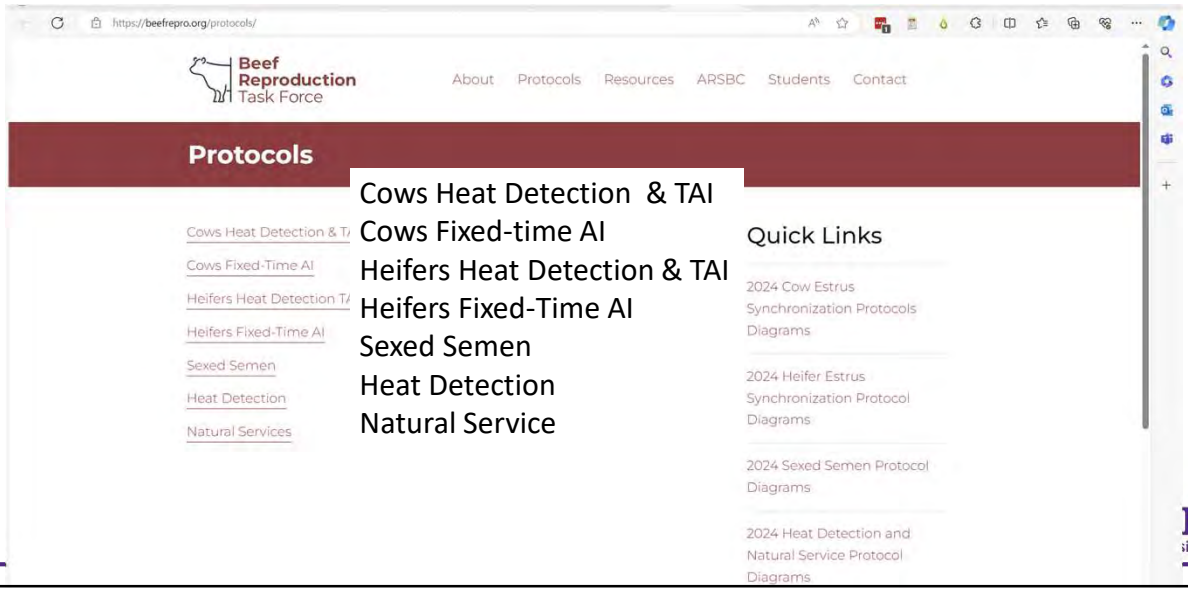
BeefRepro.org



Cows Heat Detection & TAI
Cows Fixed-time AI
Heifers Heat Detection & TAI
Heifers Fixed-Time AI
Sexed Semen
Heat Detection
Natural Service

Estrus Synchronization Planner

Free download at

https://beefrepro.org/resources/

Iowa Beef Center

Features

- Recommended systems for cows & heifers
- Select systems by type
  - Heat detect & AI systems
  - Heat detect & cleanup AI systems
  - Fixed-Timed AI Systems
- List of daily activities
- Generates Barn Calendar
- Cost per AI pregnancy
- Support materials

Estrus Synchronization Planner

Replace the example inputs (below) with your own.

Breed Type:	1=Bos taurus, 2=Bos indicus influence	1	
Semen Type:	1=Conventional, 2=Conventional & Sexed	1	
System Type:	1=Estrus AI, 2=Estrus AI + Clean-up AI, 3=Fixed-Time AI, 4=Split Time AI	3	
Synchronization Protocol:	*Select number from lists below.*	22	
Date to Start Breeding:	*Format date as "mm / dd / yyyy"*	9/15/2023	
Time of Day To Breed:	*Format time as "hh:mm AM or PM"*	8:00 AM	
GnRH:	1=Cystorelin, 2=Factrel, 3=Fertagyl, 4=OvaCyst, 5=GONAbreed	1	*2cc Cystorelin*
PG:	1= Estrumate, 2= EstroPLAN, 3= InSynch, 4=Lutalyse, 5= ProstaMate, 6=HiConc.Lutalyse, 7=Synchsure	4	*5cc Lutalyse*
Bull Turn In:	*Days after last AI*	14	
Gestation Length:	*Days*	281	

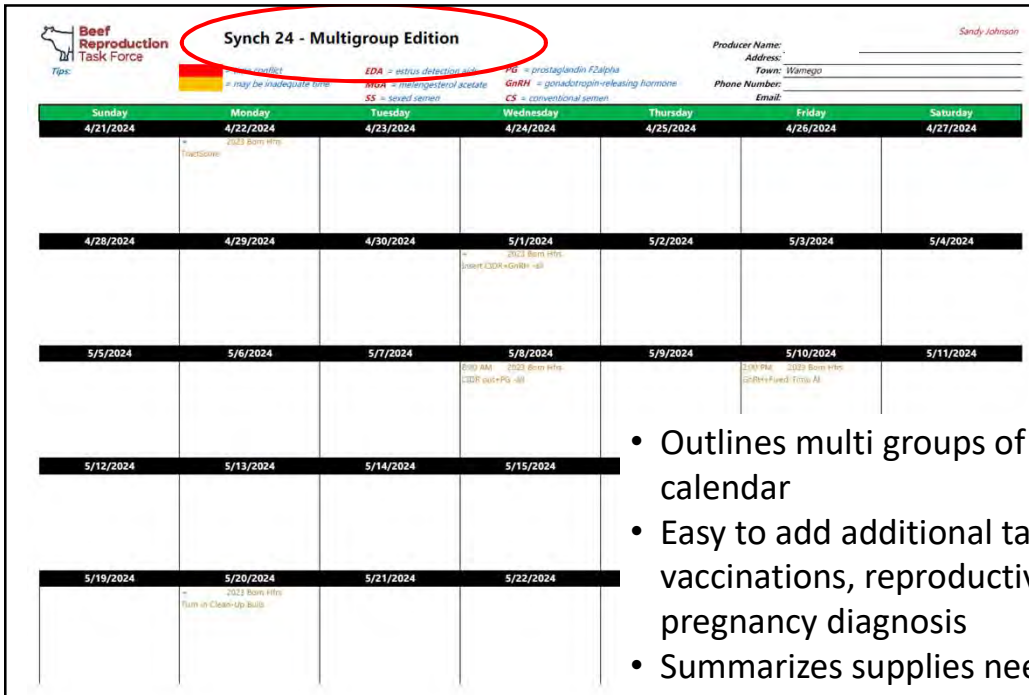
Calendar Printout Notes:

Output	
Expected Calving Date:	6/22/2024
Remove CIDR:	9/12/23 5:00 PM
Last PG Injection:	9/12/23 5:00 PM
Trips Through Chute	3
Head Worked per Hour (AI)	32
Group Size (head)	44

Cost Comparison	
Alternative System 1:	29
Alternative System 2:	39
Select number from lists below.	

Preferred Fixed-Time AI - Cow Protocols	*See Protocols*	**Preferred Fixed-Time AI - Heifer Protocols**
22 = 7 Day CO-Synch+CIDR -- Fixed-Time AI 63 +/-3		23 = 7 Day CO-Synch+CIDR -- Fixed-Time AI 54 +/- 2
29 = 5 Day CO-Synch+CIDR -- Fixed-Time AI 72 +/-2		27=MGA + PG -- Fixed-Time AI 72 +/-2
39=7 & 7 Synch -- Fixed-Time AI 66 +/- 2		32=14 Day CIDR+PG -- Fixed- Time AI 66 +/-2

Program | Planner Worksheet | Calendar | Print Out | Tips & Overview | Visual Protocols | notes



- Outlines multi groups of animals on one calendar
- Easy to add additional tasks – vaccinations, reproductive tract scores, pregnancy diagnosis
- Summarizes supplies needed

Democritus
c. 460 BC – c. 370 BC

Right testis produces males
Left testis produces females

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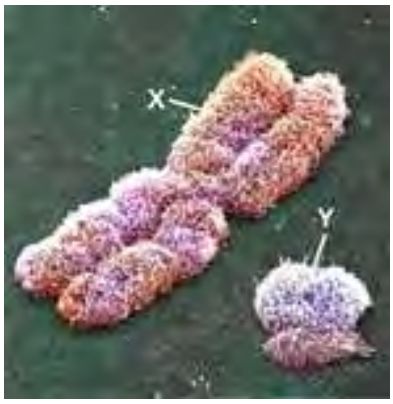

X – sperm have more DNA

Dye binds to DNA

Laser light causes dye to fluoresce

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Optimizing the use of sex-sorted semen

	Pregnancy rates		
Estrus	Conventional Semen @ 66 hr	Sex-sorted Semen @ 66 hr	Sex-sorted @ 66 hr with delay of non-estrous
Yes	**77%[a]**	**51%[b]**	**42%[b]**
No	**37%[c]**	**2%[d]**	**36%[c]**
Total	**56%**	**26%**	**39%**

- Treatment x estrous expression interaction ($P < 0.0001$)
- Pregnancy rates within a row with different superscripts are different ($P < 0.0001$)

Thomas et al., 2014

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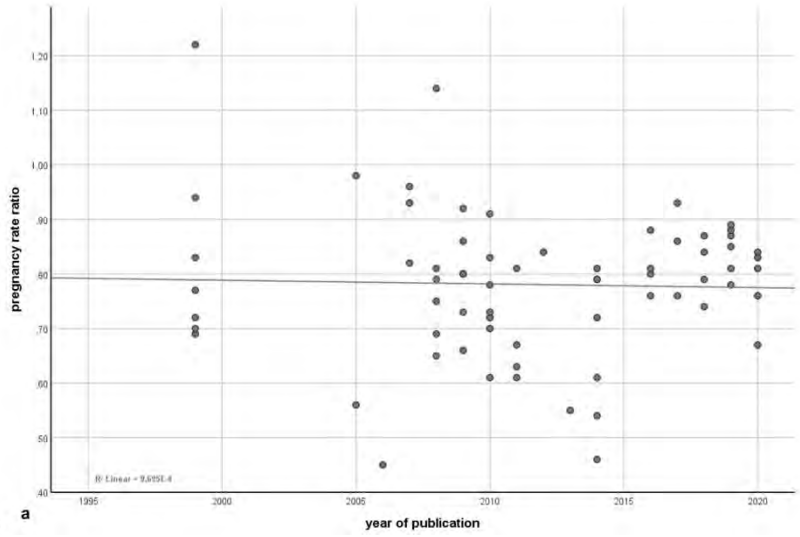
Variation in sexed semen results over time



Reese et al., 2021

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Beef Reproduction Task Force

SEXED SEMEN PROTOCOLS 2024

For additional synchronization protocols visit *BeefRepro.org*
Use of sexed semen can result in conception rates that are decreased compared to conventional semen used in the same situation

HEAT DETECTION

Sexed semen can be used on any female observed in estrus (heat) and following synchronization with any protocol on the Cow or Heifer Protocol Sheet.
For best results with sexed semen *perform AI 16 to 22 hr* after detecting female in estrus.
Use of estrus detection aids (EDA) is highly recommended.

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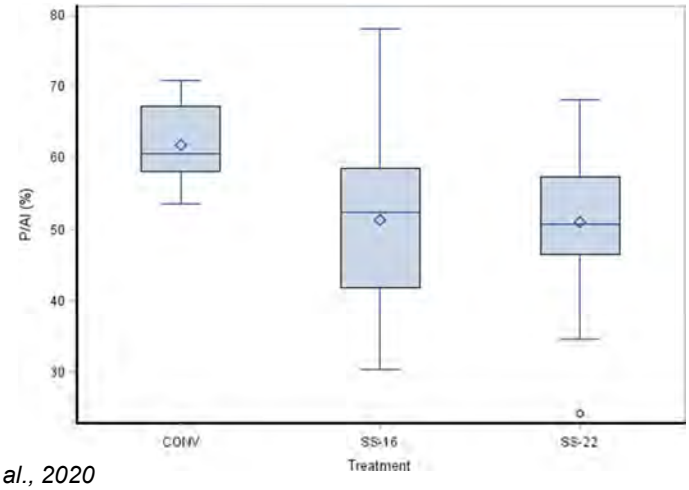
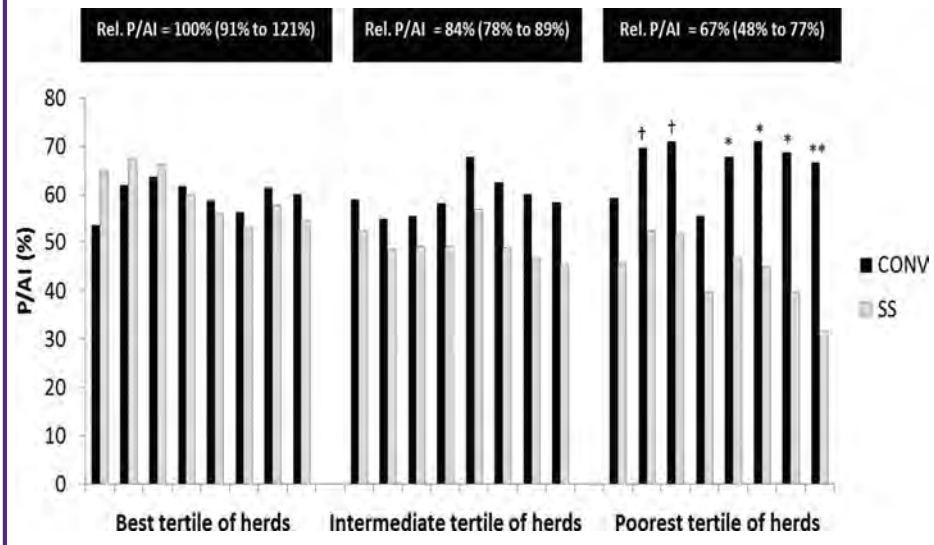
Variation in sexed semen



Drake et al., 2020
Journal of Dairy Science 2020 10312059-12068DOI: (10.3168/jds.2020-18847)

K-STATE Research and Extension



Drake et al., 2020
Journal of Dairy Science 2020 10312059-12068DOI: (10.3168/jds.2020-18847)

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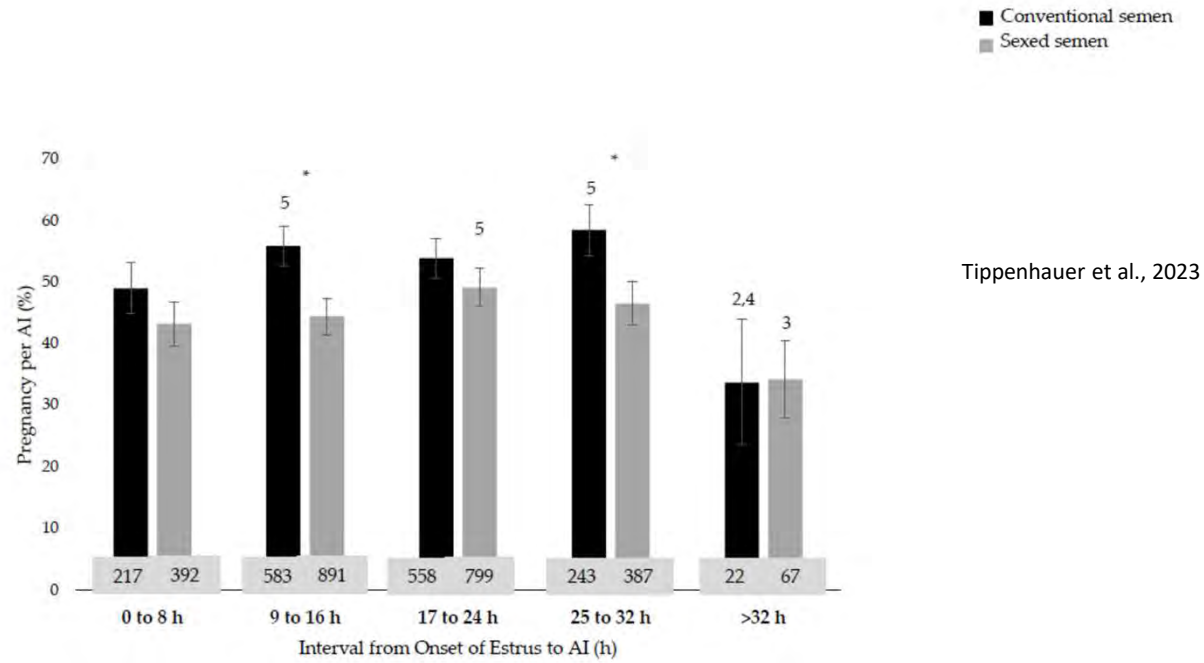
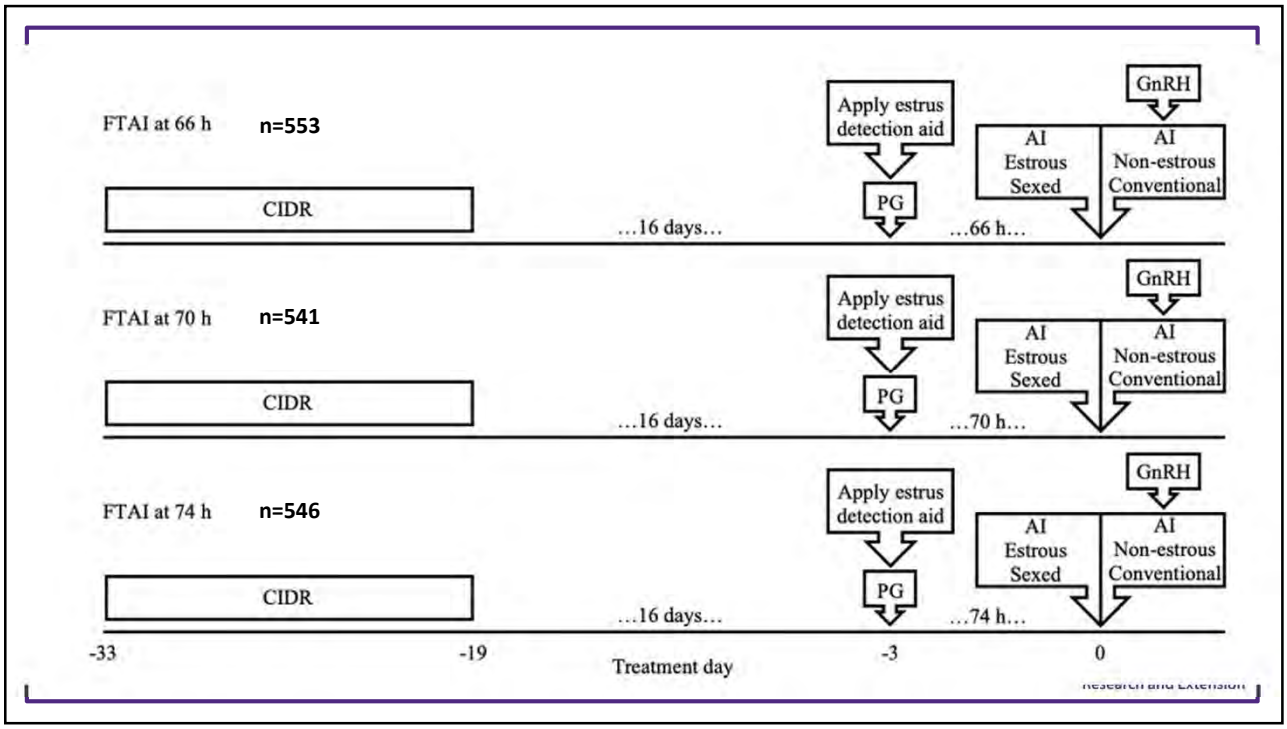
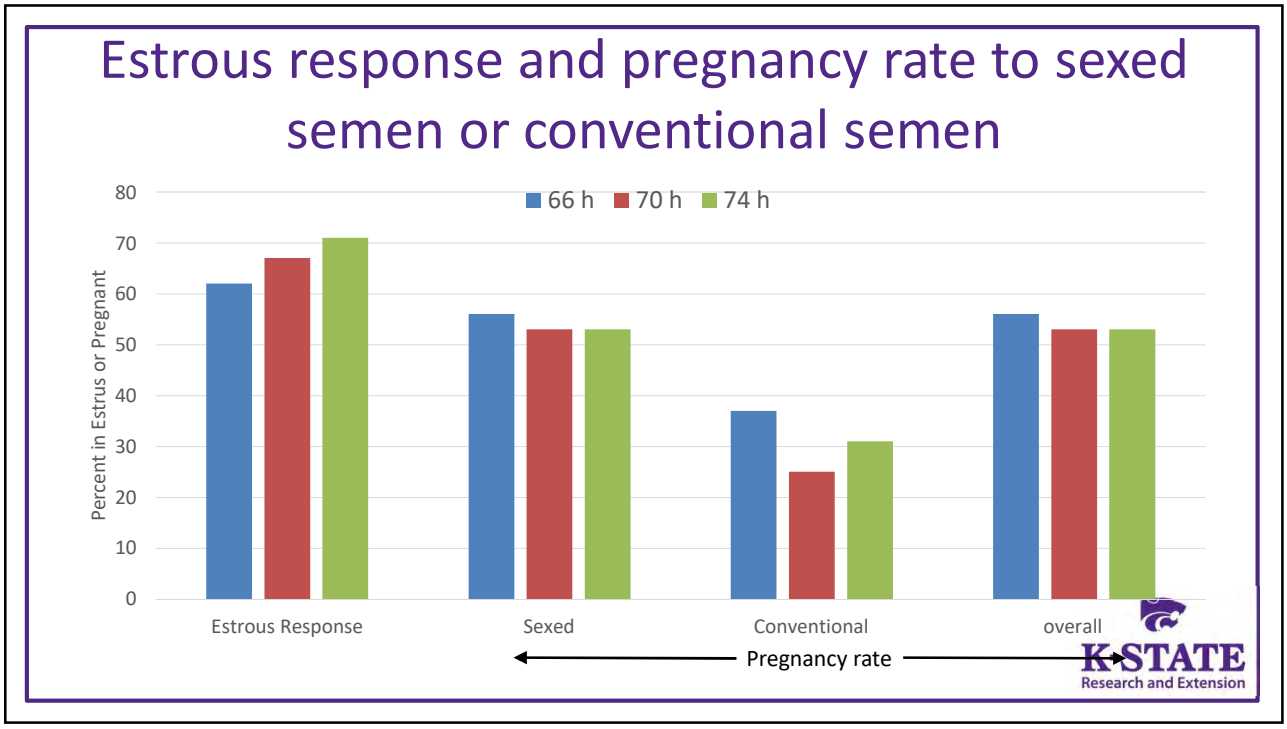
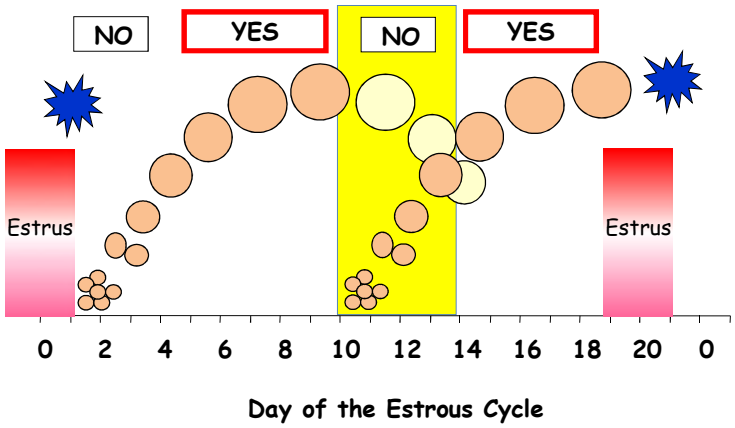


Tippenhauer et al., 2023


FTAI at 66 h
n=553
CIDR
…16 days…
Apply estrus detection aid
PG
…66 h…
AI Estrous Sexed
GnRH
AI Non-estrous Conventional
FTAI at 70 h
n=541
CIDR
…16 days…
Apply estrus detection aid
PG
…70 h…
AI Estrous Sexed
GnRH
AI Non-estrous Conventional
FTAI at 74 h
n=546
CIDR
…16 days…
Apply estrus detection aid
PG
…74 h…
AI Estrous Sexed
GnRH
AI Non-estrous Conventional
-33
-19
Treatment day
-3
0


Estrous response and pregnancy rate to sexed semen or conventional semen
66 h
70 h
74 h
Percent in Estrus or Pregnant
80
70
60
50
40
30
20
10
0
Estrous Response
Sexed
Conventional
overall
Pregnancy rate
K-STATE
Research and Extension

GnRH-Induced Ovulation During a Follicular Wave



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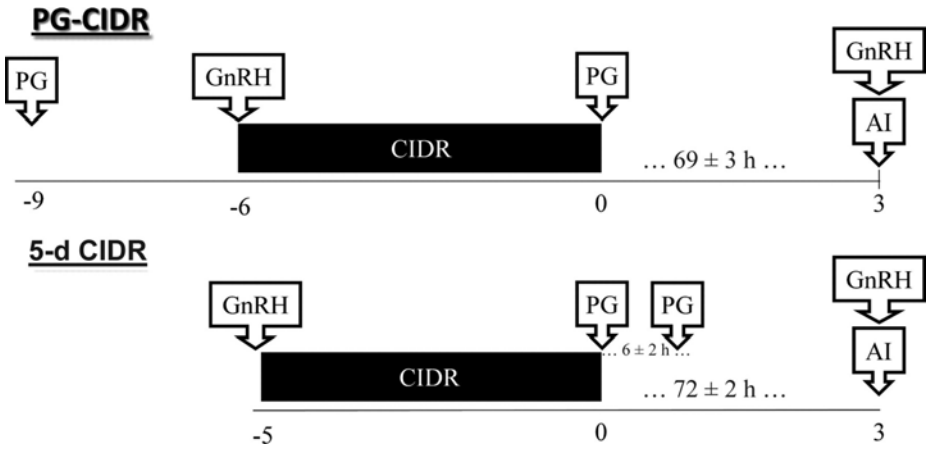
Pre-Synchronization



Perry et al., 2012

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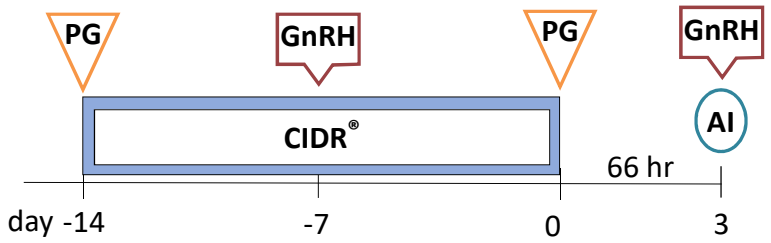
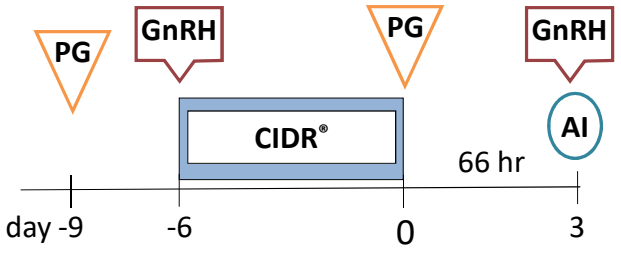
7&7 Synch



PG 6-day CIDR® & TAI



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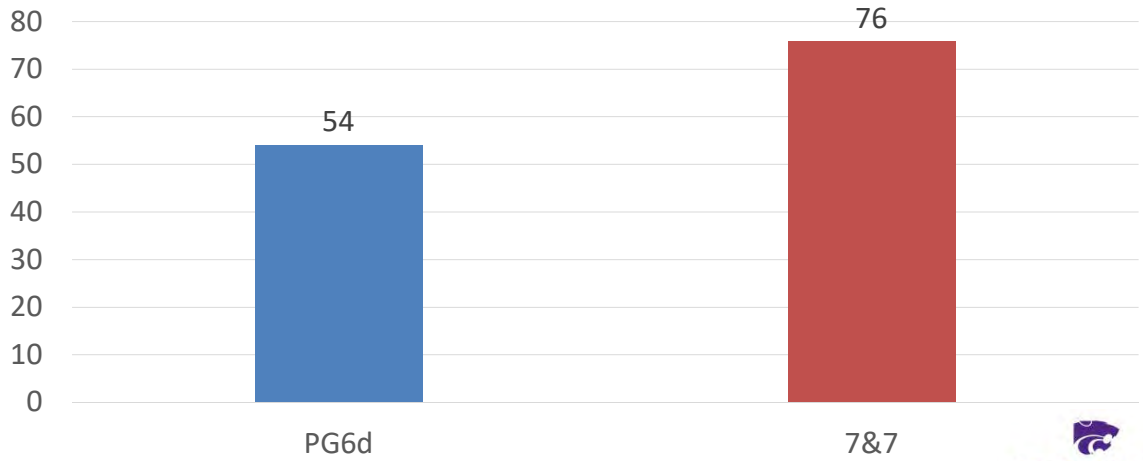
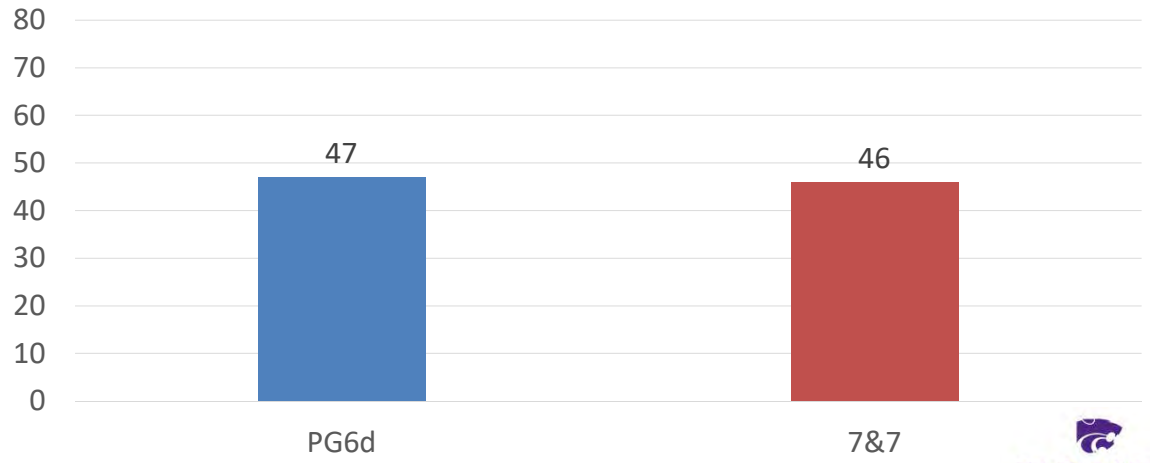
Estrous Response in Beef Cows



Ketchum et al., 2024

K-STATE Research and Extension

Pregnancy rate to fixed-timed AI

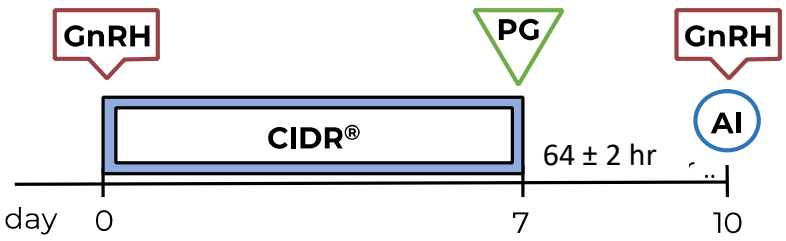


Ketchum et al., 2024

K-STATE Research and Extension

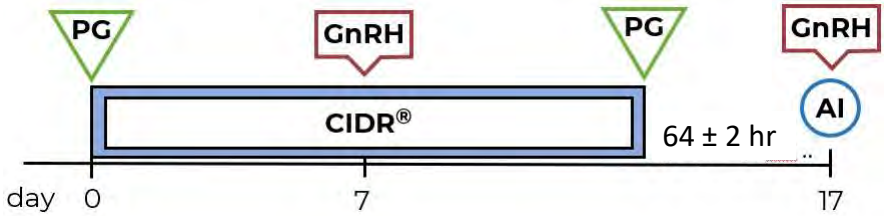
7-day CO-Synch+CIDR®

Pancini et al., 2022



n	BCS	DPP
606	5.5	81
610	5.5	80

7&7 Synch



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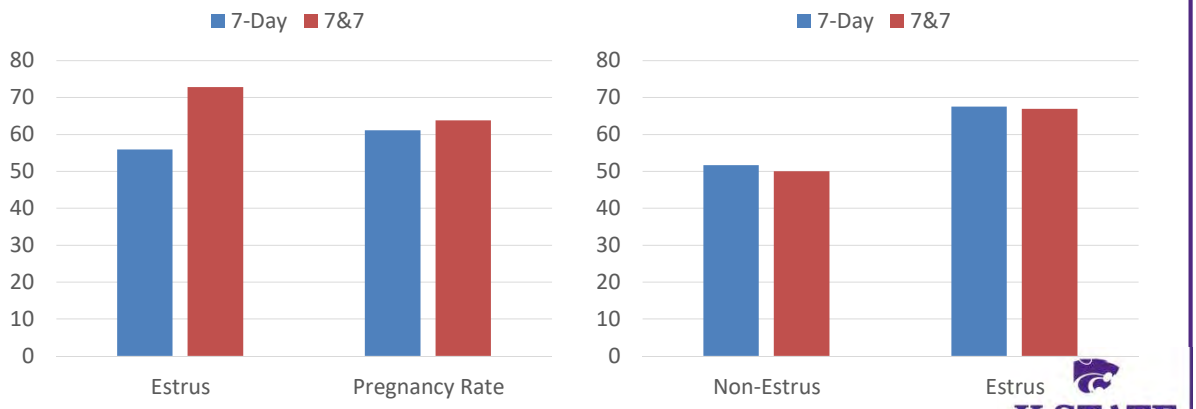
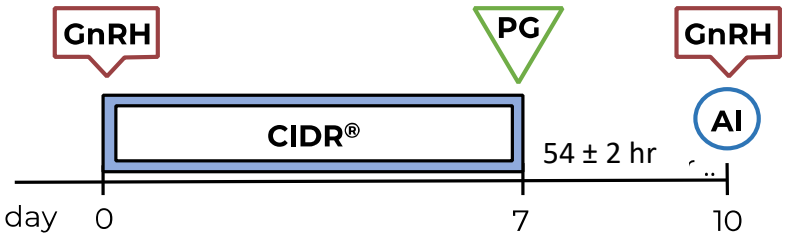
Beef Cows at Fixed-Time AI

*** Treatments differ P<0.01**

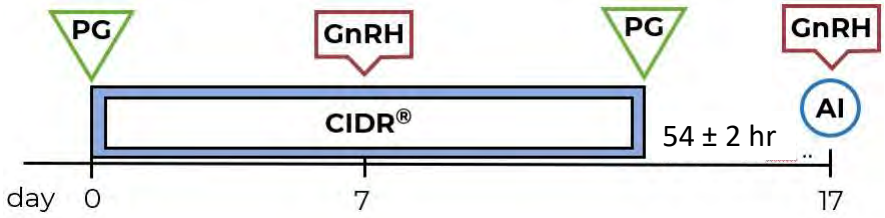


K-STATE Research and Extension

7-day CO-Synch+CIDR®



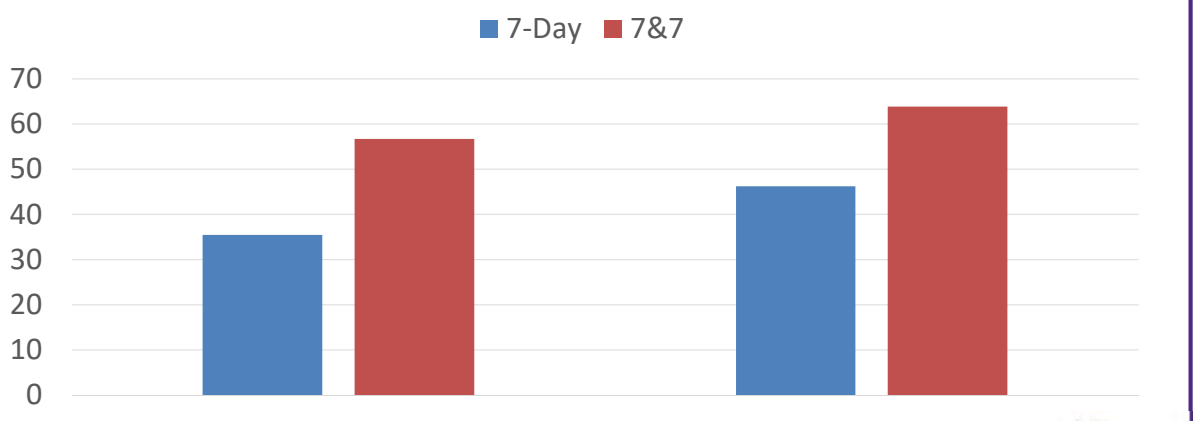
7&7 Synch



n
779
773

K-STATE Research and Extension

Beef Heifers at Fixed-Time AI



Trt P=0.001; Loc P<0.001

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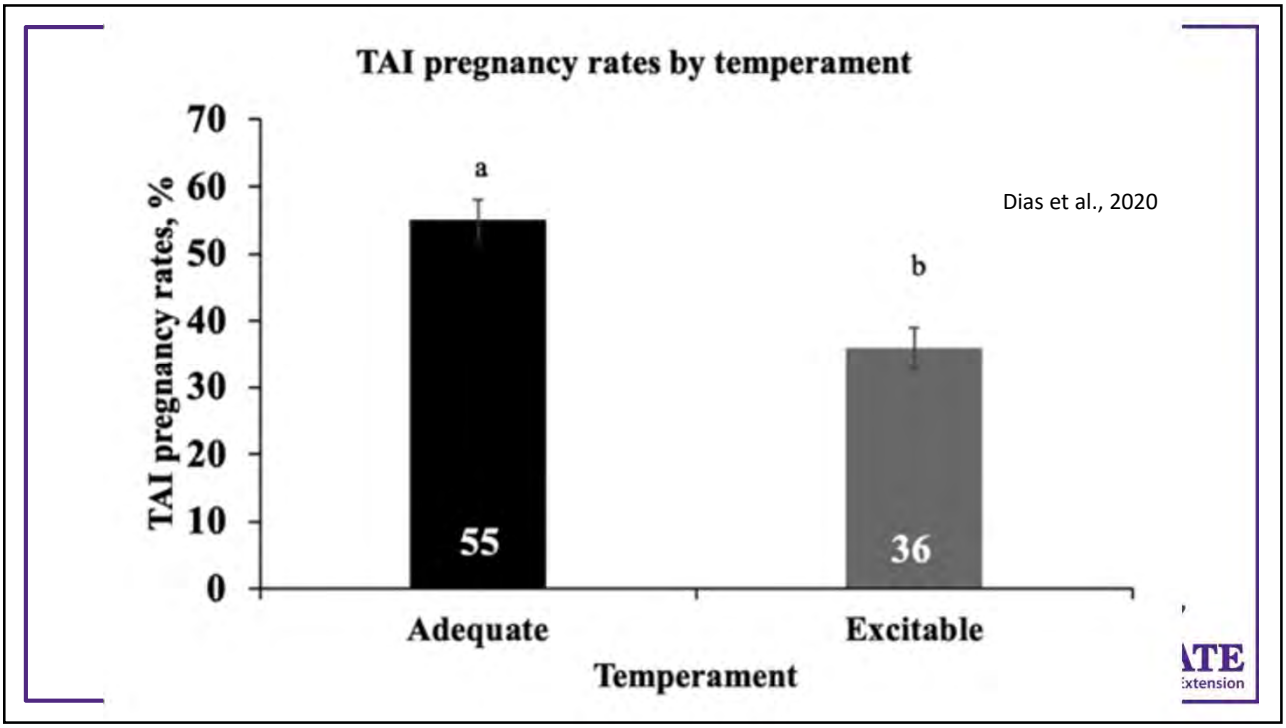
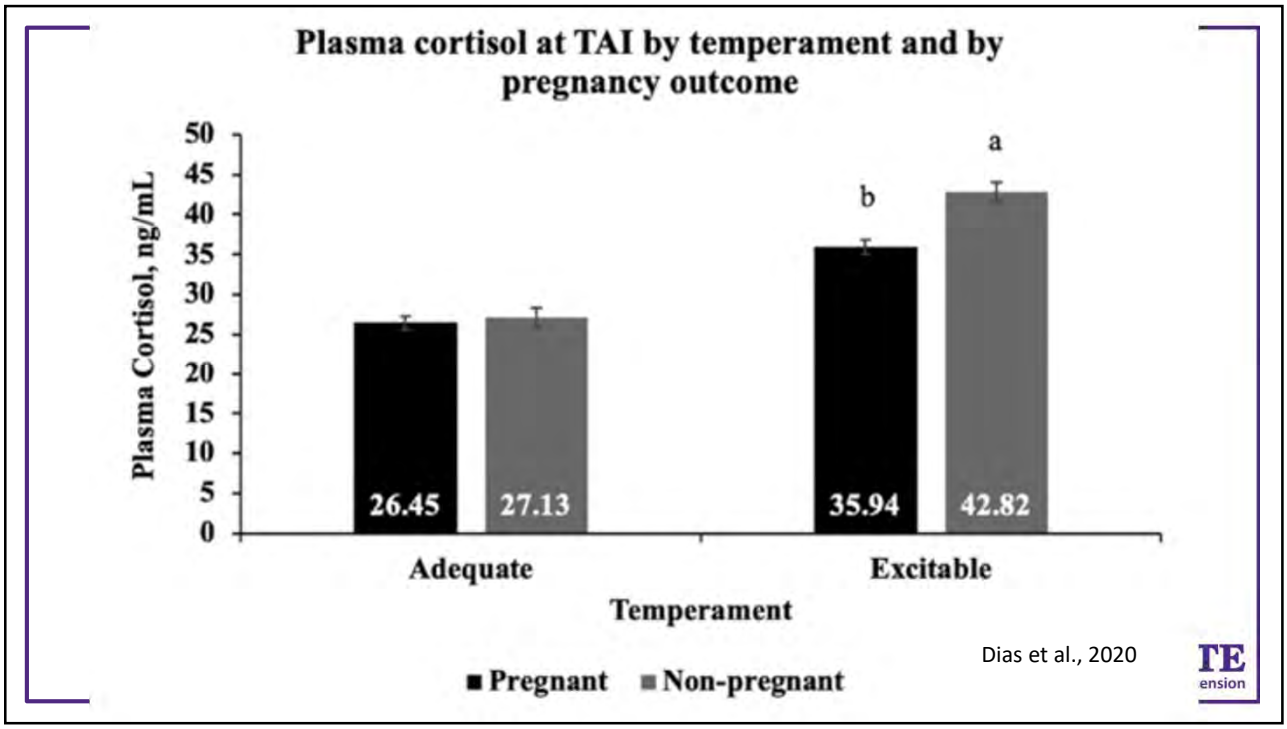
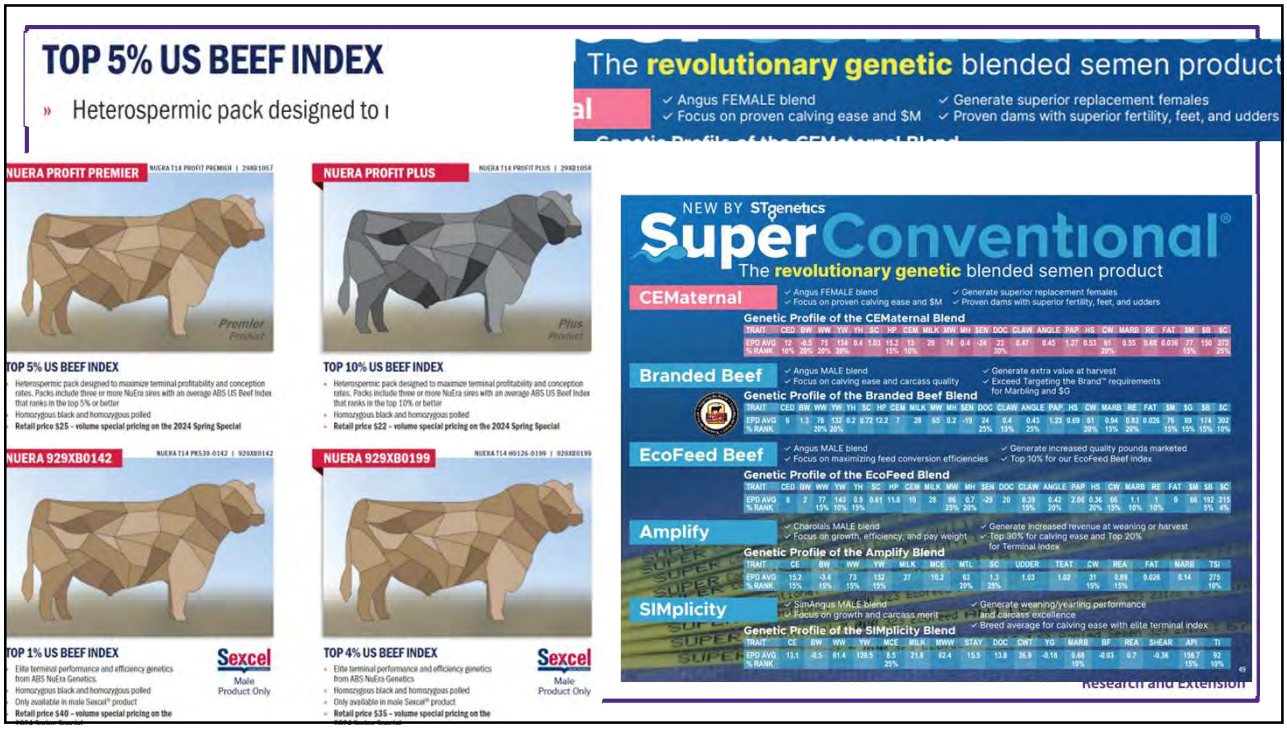


Dias et al., 2020


Plasma cortisol at TAI by temperament and by pregnancy outcome
Plasma Cortisol, ng/mL
50
45
40
35
30
25
20
15
10
5
0
26.45
27.13
35.94
42.82
a
b
Adequate
Excitable
Temperament
Pregnant
Non-pregnant
Dias et al., 2020
TE
ension


TOP 5% US BEEF INDEX
Heterospermic pack designed to
The revolutionary genetic blended semen product
Angus FEMALE blend
Focus on proven calving ease and $M
Generate superior replacement females
Proven dams with superior fertility, feet, and udders
NUERA PROFIT PREMIER
TOP 5% US BEEF INDEX
Retail price $25 - volume special pricing on the 2024 Spring Special
NUERA PROFIT PLUS
TOP 10% US BEEF INDEX
Retail price $22 - volume special pricing on the 2024 Spring Special
NUERA 929XB0142
TOP 1% US BEEF INDEX
Sexcel
Male Product Only
NUERA 929XB0199
TOP 4% US BEEF INDEX
Sexcel
Male Product Only
NEW BY STgenetics
SuperConventional
The revolutionary genetic blended semen product
CEMaternal
Genetic Profile of the CEMaternal Blend
Branded Beef
Genetic Profile of the Branded Beef Blend
EcoFeed Beef
Genetic Profile of the EcoFeed Blend
Amplify
Genetic Profile of the Amplify Blend
SIMplicity
Genetic Profile of the SIMplicity Blend
Research and Extension

Heterospermic semen packages

- 2 or more males mixed on equal # basis
- Disproportion of offspring from 1 male

- Takes advantage of differences in semen characteristics

K-STATE Research and Extension

Exploiting differences in semen

- Time to capacitation
- Longevity in tract
- Survival of freezing or thawing
- Freezing & thawing post sex-sorting

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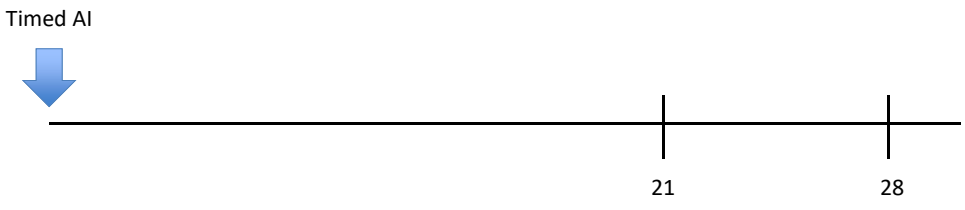
Effects of heterospermic insemination on conception rates of lactating dairy cows

DeJarnette et al., 2008

K-STATE Research and Extension

Resynchronization



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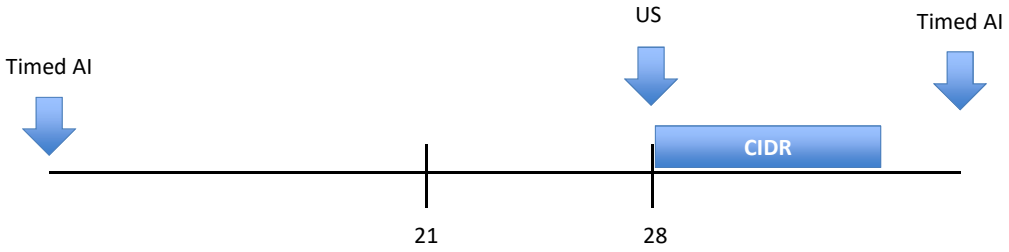
Resynchronization



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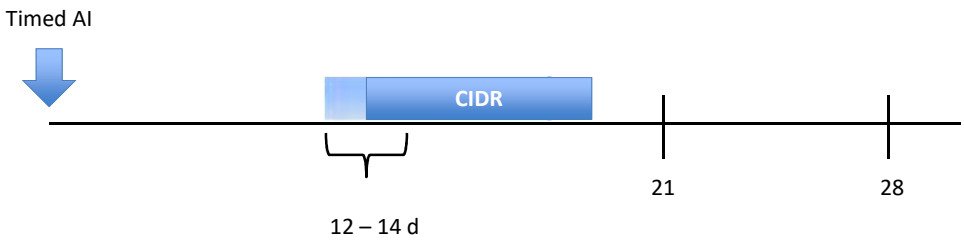
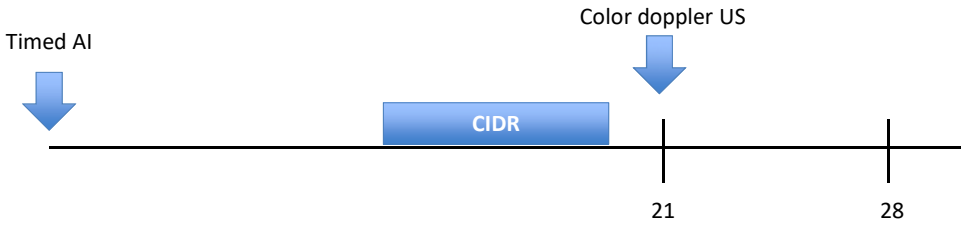


Resynchronization



K·STATE Research and Extension

True Positive

False Negative

Siqueira et al., 2013

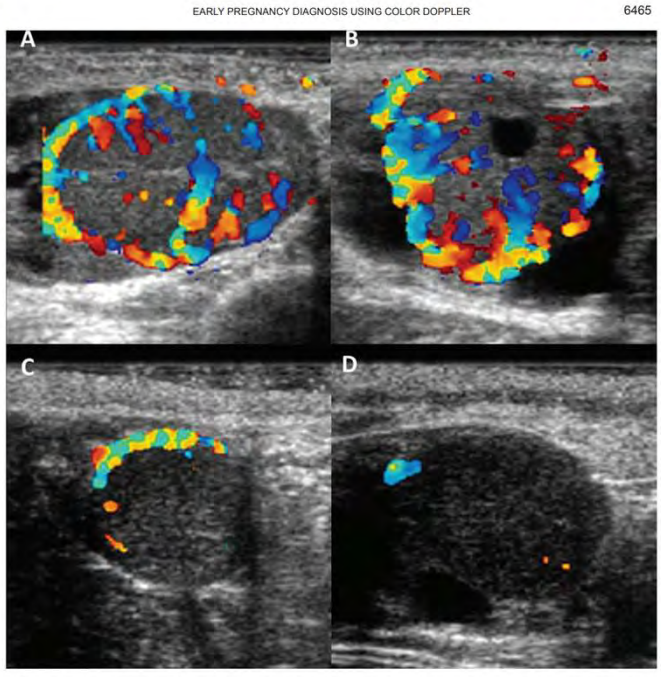


Figure 1. Doppler ultrasonographic images representing the 4 categories observed after pregnancy diagnosis using the corpus luteum blood flow at 20 d after AI (CLBF-d20) test: true positive (A), false positive (B), false negative (C), and true negative (D).

False Positive

True Negative

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Table 1
Studies comparing the use of Doppler ultrasonography to detect luteal function for diagnosis of pregnancy status between day 17 and day 22 and the B-mode ultrasonography for detection of embryo after day 28 of pregnancy in beef and dairy cows and heifers.

Authors	Type/breed	Parity order	Type of service	Moment of gestation, d	Animals, n	Conception rate, %	Accuracy, %	False positive, % (total)	False negative, % (total)	PPV, %	NPV, %
Utt et al., 2009	Beef (Crossbreed)	Cows	TET	17	50	46	60	30.6	9.4	54.4	73.3
				19			68.8	24.6	6.6	61.4	82.9
				21			71.4	21	7.6	64.8	81.5
Siqueira et al., 2013	Dairy (Holstein-Gir)	Parous cows	TAI	20	317	46.1	74.6	24.6	0.7	64.8	97.7
		Heifers			209	47.4	76.6	22.7	0.6	67.2	98
Pugliesi et al., 2014	Beef (Nelore)	Parous cows	TAI	20	111	37.8	91	9	0	80.8	100
Guimarães et al., 2015	Beef (Nelore)	Parous cows	TET	21	163	43.6	88.3	11.7	0	78.9	100
Scully et al., 2014[1]	Dairy (*Bos taurus*)	Parous cows	AI after estrus	18	80	52.8	68.1	20.9	11.1	66.6	70.3
				19	80	56.8	82.4	13.5	4.1	79.5	88
				20	90	42.2	77.7	17.3	4.9	70.8	87.8
				21	94	45.2	87.1	11.8	1.1	78.8	97.5
Pugliesi et al., 2018	Beef (Nelore)	Parous cows	TAI	22	246	49.6	94.7	5.3	0	90.4	100
		Heifers			231	41.6	90	10	0	80.5	100
Araíde et al., 2018	Beef (Nelore)	Parous cows	TET	22	221	35.5	83.8	16.2	0	68.8	100
Andrade et al., 2019	Beef (Nelore)	Heifers	TAI	21	113	–	87.8	12.2	0	77.3	100
Dalmaso de Melo et al., 2020	Beef (Nelore)	Parous cows	TAI	20	144	58	93	7	0	89	100
		Heifers			100	52	88.3	11.7	0	81.8	100
Wellert et al., 2020	Beef (Angus-cross)	Parous cows	TAI	21	84	–	–	–	0	89.4	100
		Heifers			25				0	75	100
Dubuc et al., 2020	Dairy (Holstein)	Parous cows	AI	21	1 632	22	62.1	37.5	0.4	52	98.1
Holton et al., 2022a	Beef (*Bos taurus*)	Parous cows	TAI	20	208	52.9	87	13	0	80	100
				22	209	52.6	92	8	0	85	100
Holton et al., 2022b	Beef (*Bos taurus*)	Heifers	TAI	20	183	–	90	10	0	86	100
				22		–	92	8	0	90	100
Madoz et al., 2022	Dairy (Holstein)	Parous cows	TAI	19/20	131	37.4	74.8	24.5	0.7	60	98.2
		Heifers									
Ferraz et al., 2022	Dairy (Holstein)	Parous cows	TAI	21	140	28.6	53	46.3	0.7	38	98
		Heifers			32	31.3	66	34	0	48	100

Abbreviations: PPV = positive predictive value; NPV = negative predictive value; TET = timed-embryo transfer; TAI = timed-artificial insemination; AI = artificial insemination.
[1] This study also considered the uterine echotexture and corpus luteum size as additional criteria to the luteal blood perfusion for diagnosis of non-pregnant animals.

Pugliesi et al., 2023

Research and Extension

Accuracy associated with color doppler imaging to detect CL blood flow / pregnancy

Accuracy	75	91	87	91	92	94
False Positive	24	9	12	9	8	6
False Negative	.5	0	0	0	0	0
Day			20	20	22	22
Type	Holstein	Nelore	Beef cows	Beef Heifers	Beef Cows	Beef Heifers
Year	2014	2013	2022	2022	2022	2022

Research and Extension

- Resources
- Sexed Semen
- Protocols
- Docility & Fertility
- Heterospermic semen packages
- Rapid rebreeding/resynchronization

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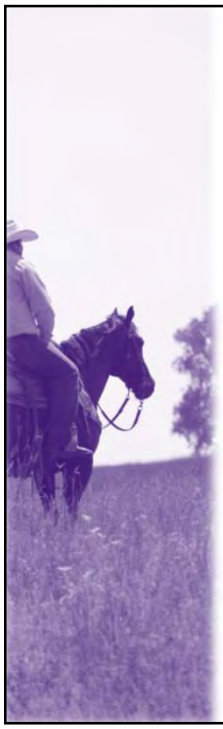
BeefRepro.org

K-STATE Research and Extension

HEIFER MANAGEMENT CONSIDERATIONS FOR FERTILITY & LONGEVITY

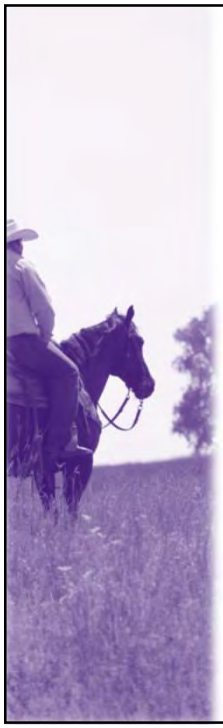
SANDY JOHNSON
PHD

LARGE ANIMAL



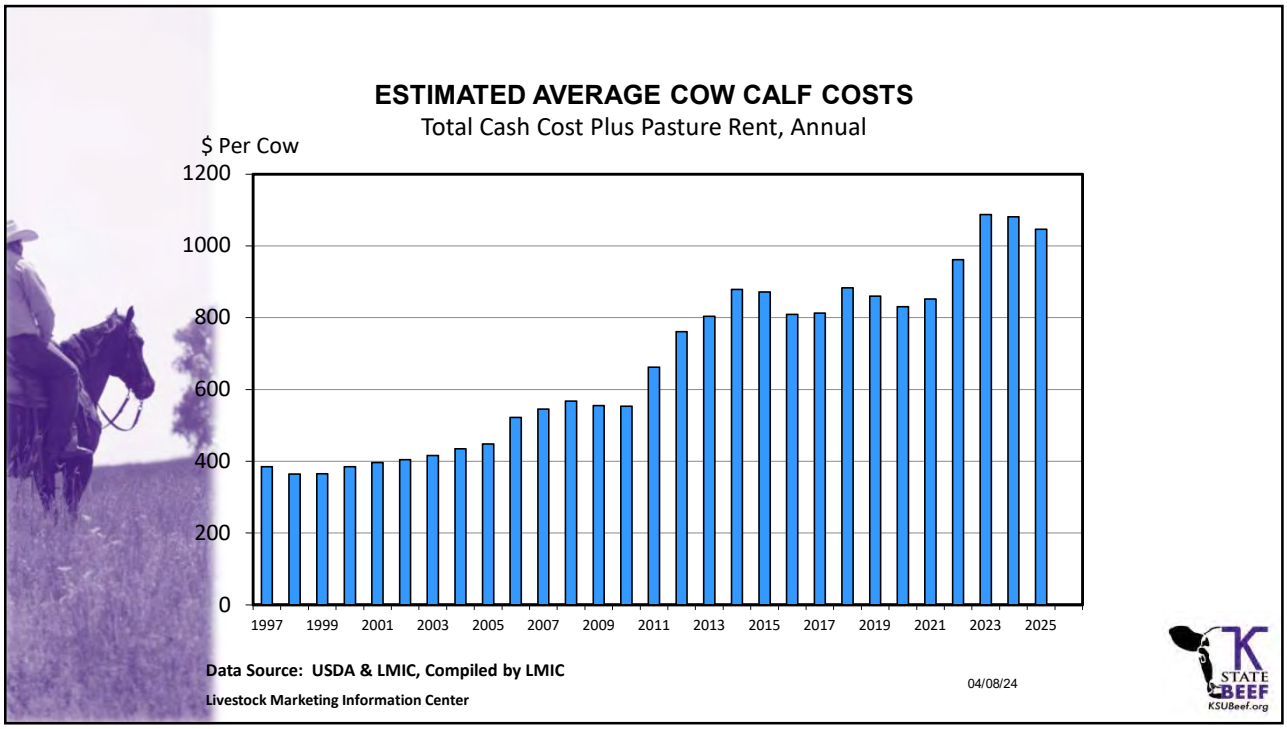
Heifer Management Considerations for Longevity and Fertility

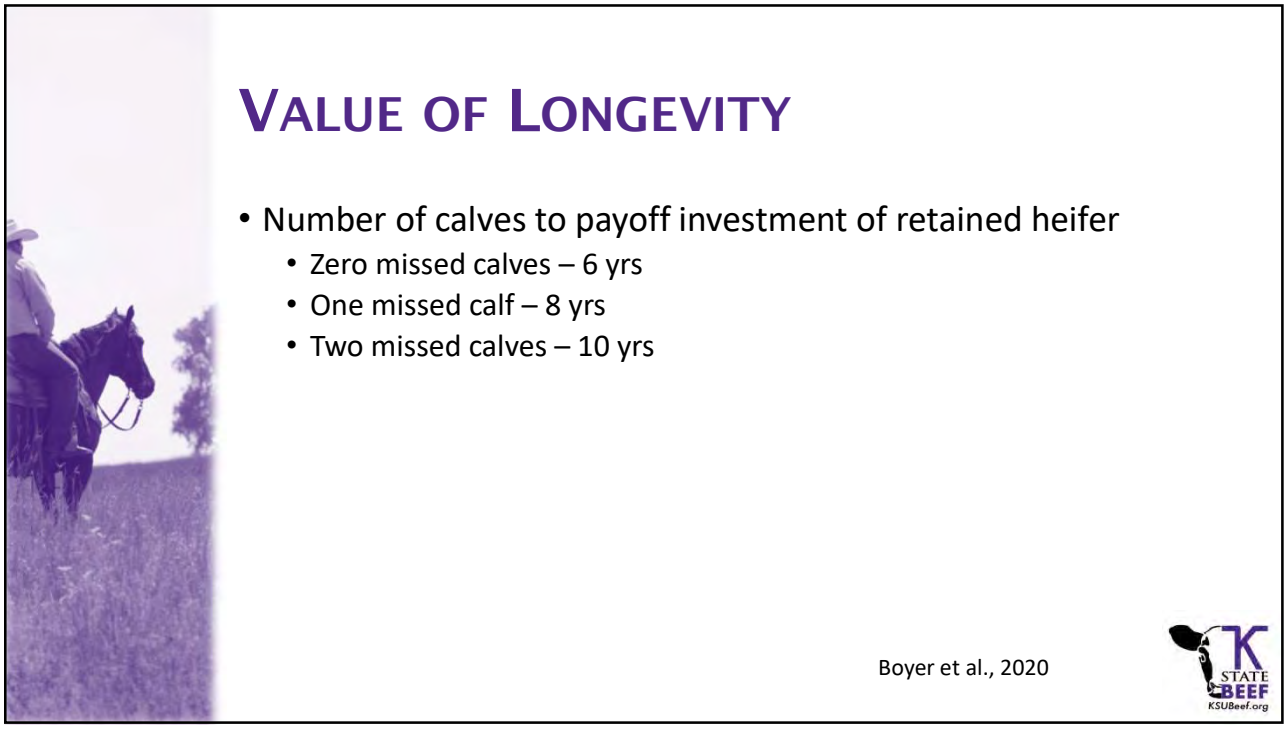
Sandy Johnson, PhD
sandyj@ksu.edu
June 2, 2024
Manhattan, KS



Considerations

- Production System
- Marketing Endpoint
- Production Goals

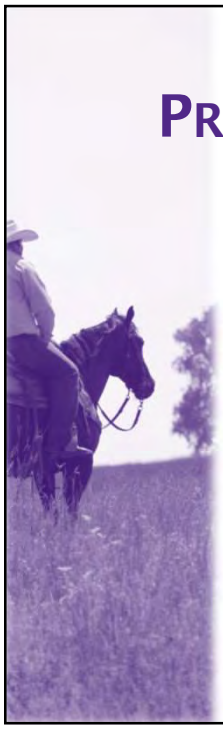




Value of Longevity

- Number of calves to payoff investment of retained heifer
  - Zero missed calves – 6 yrs
  - One missed calf – 8 yrs
  - Two missed calves – 10 yrs

Boyer et al., 2020

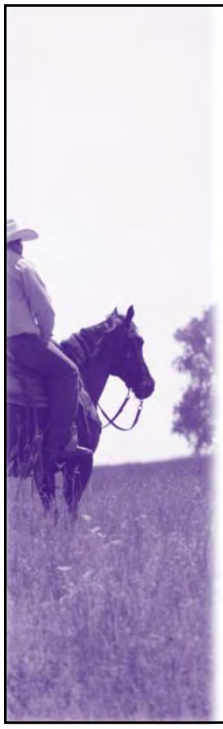


Productive Years & Cow Depreciation

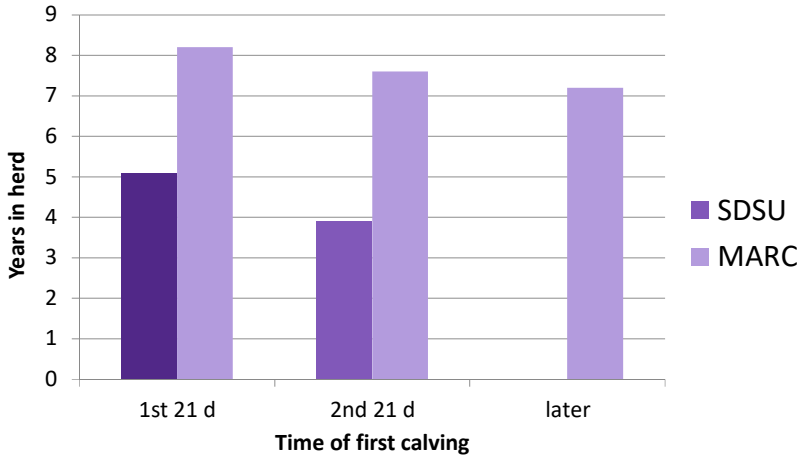
- Value of replacement entering herd
- Sale value when she exits
- Number of years in production

$2000 - $1000 = $1000

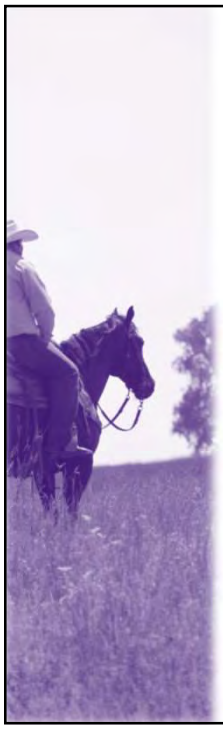
4 yrs - $250/yr
5 yrs - $200/yr
6 yrs - $167/yr
8 yr - $125/yr



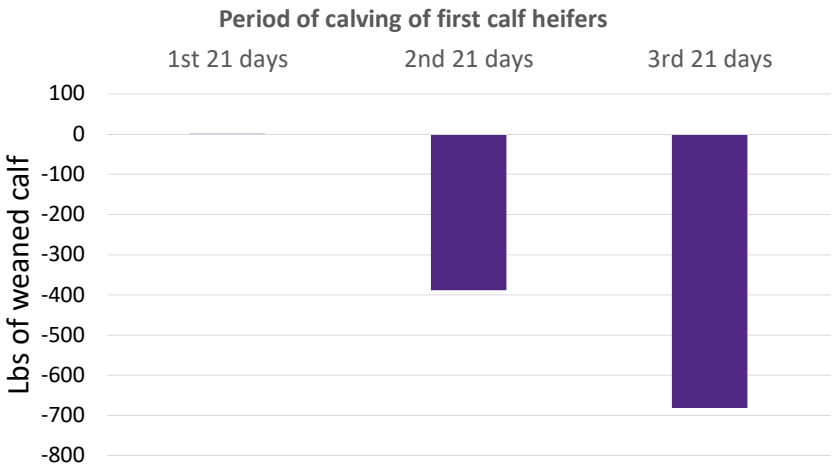
Longevity in herd based on calving date as heifer



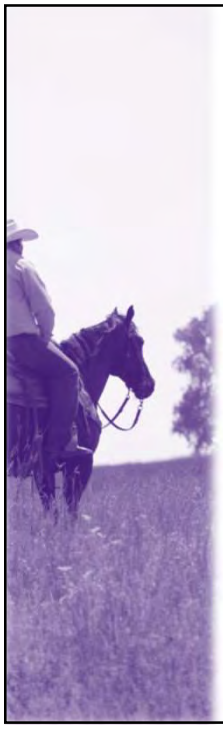
Cushman et al., 2013



Lifetime weaning weight disadvantage of heifers that conceive later in the first breeding season



Cushman et al., 2013



Opportunity

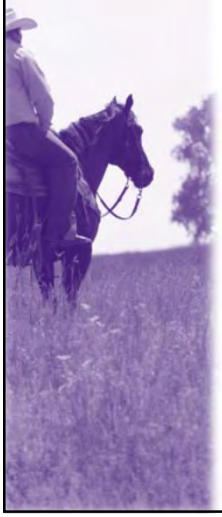
- Leave bull in longer
- Stage pregnancies to identify those that conceive early

- Market opens and late breds

Postweaning Systems

Cost
Performance

Shifts in heifer development



THEN

- Emphasis on puberty
- Target weight 60 – 65%
- Cheap grain
- Feedlot system

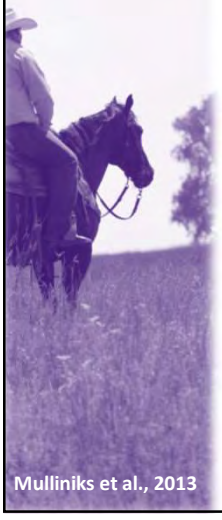
NOW

- Puberty less of an issue - heifers becoming pregnant on the cow
- Higher production costs - lower target weight reduce development costs
- Open yearling heifers profitable

Summer gains followed the reverse order of winter gains. (Lemenager et al., 1980)

- Heifers wintered with no supplement exhibited compensatory gains on summer grass when compared to heifers wintered with supplemental feed. Joubert (1954), Zimmerman *et al.* (1958) and Short and Bellows (1971)

	Ground Ear Corn lbs /hd/day winter phase		
	0	2.7	5.4
Winter ADG	.07	.48	.77
Summer ADG	1.72	1.50	1.32



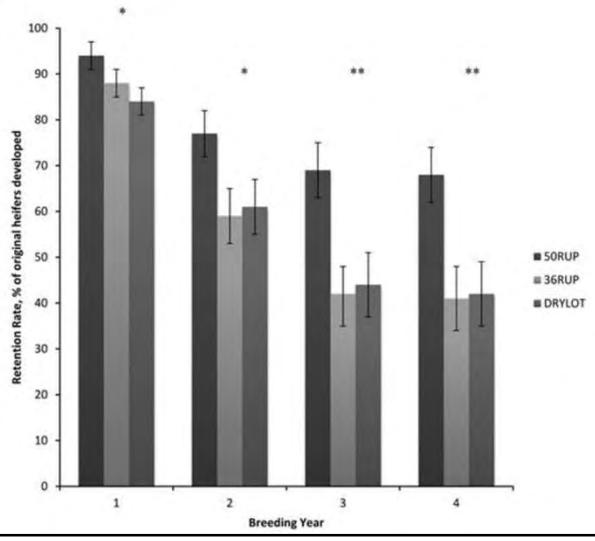
Growth & reproductive performance of heifers grazing native dormant range with protein supplement or in drylot

Item	36RUP	50RUP	Drylot	P-Value
BW				
Weaning	491	493	493	0.97
Breeding	607	607	693	<0.01
ADG				
Initial to breeding	0.59	0.57	1.52	<0.01
Breeding to preg	1.87	1.76	1.34	<0.01
% mature BW	51	51	58	<0.01
Pregnancy rate, %	88	94	84	0.10
Calving date	66	65	63	0.89
Net Return/Heifer developed	$256.03	$268.86	$168.85	

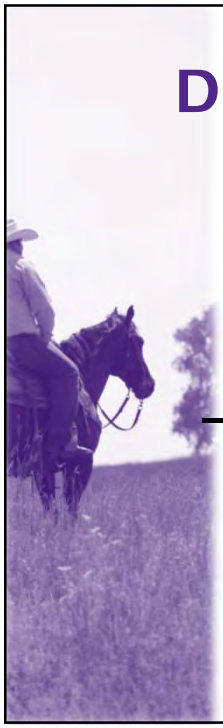
Mulliniks et al., 2013

Retention of heifers fed 2 types of protein supplementation or fed in drylot

Figure 1.



Mulliniks et al., 2013



Design

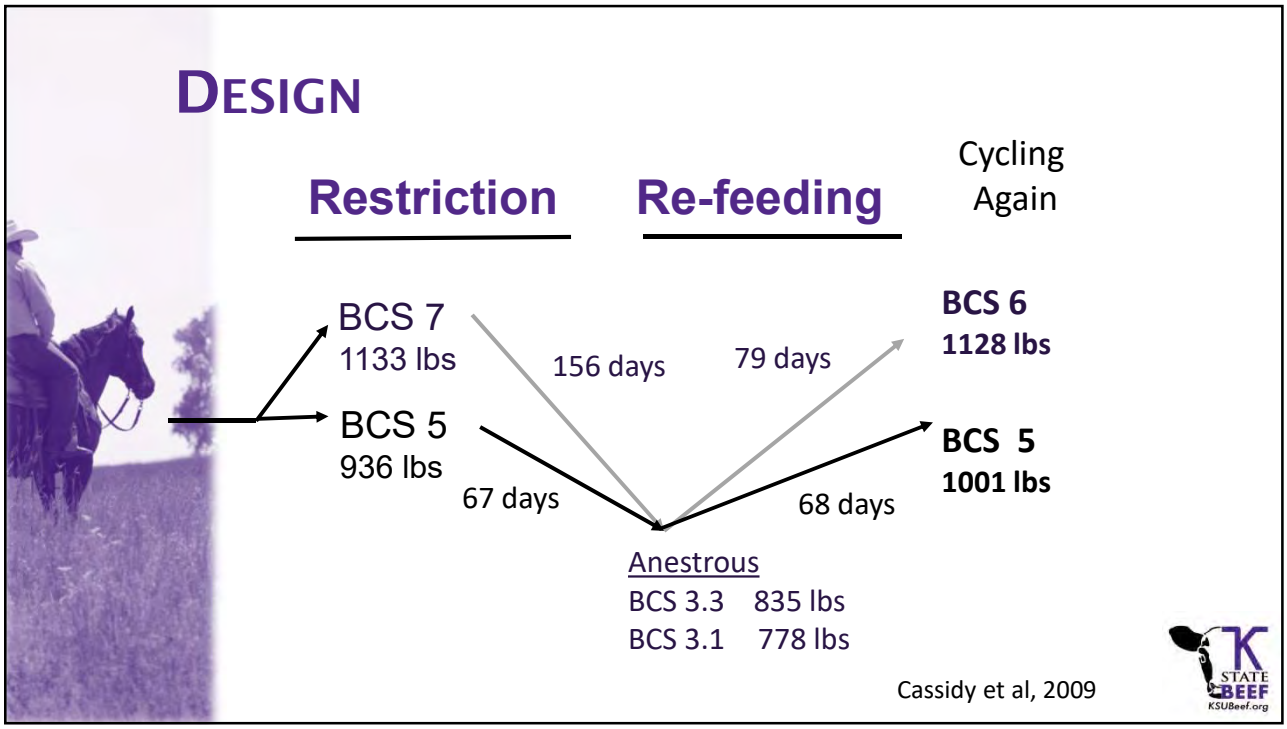
Restriction **Re-feeding**

BCS 7

BCS 5

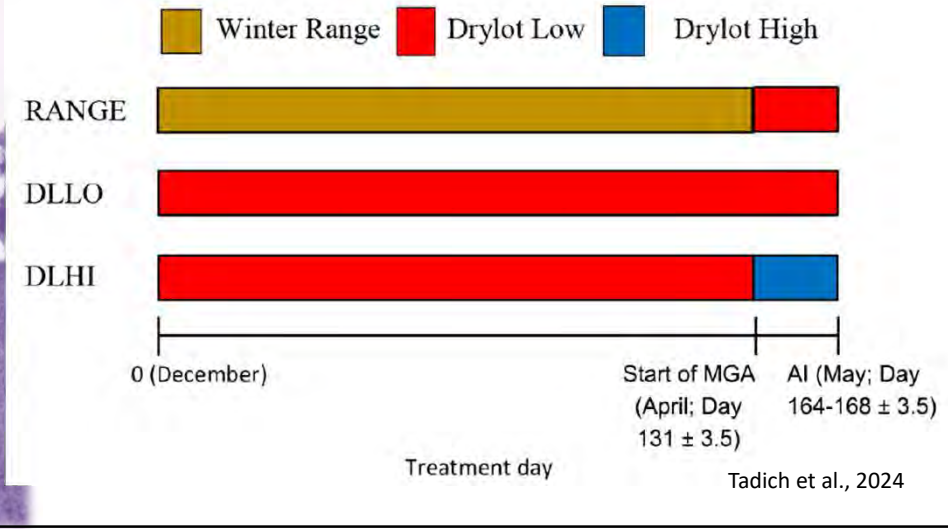
Cassady et al 2009

Design



Cassidy et al, 2009

Effects of short-term nutritional increase before AI



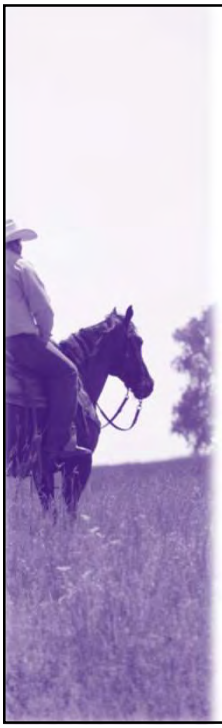
Tadich et al., 2024

Effect of nutritional increase on reproductive performance during a 33-d synchronization treatment period in beef heifers

Item	Range	DLLO	DLHI
n	3	3	3
Initial BW	482	480	482
Development ADG	0.57[b]	1.41[a]	1.41[a]
Prebreeding BW	686[b]	799[a]	825[a]
% Mature BW	57[b]	66[a]	68[a]
Breeding ADG	1.52[b]	0.84[a]	0.77[a]
Final Preg BW	909[b]	972[a]	986[a]

[a,b] Means differ P <0.02

Tadich et al., 2024



Effect of nutritional increase on reproductive performance during a 33-d synchronization treatment period in beef heifers

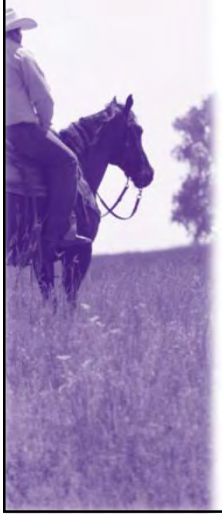
Item	Range	DLLO	DLHI	P-value
n	3	3	3	
Cycling (P4), %	14	62	24	0.16
Detection of estrus, %	70	93	89	0.07
AI Pregnancy Rate, %	49	63	69	0.34
Final Pregnancy Rate, %	84	95	93	0.09
Calving Rate, %	77	85	93	0.11
Calved in first 21 d, %	42	41	55	0.23

Tadich et al., 2024

How big at first breeding?

% Mature BW	Yearling Preg Rate	2-yr-old Preg Rate	3-, 4-, 5-yr-old Preg Rate	Calve first 21 d
70	85	92	ND	65
65	85	90	ND	65
60	83	87	ND	76
55	80	82	ND	77
50	73	75	ND	76

Crouch et al., 2024



How much should they weigh?

65% of mature weight

- Tried and true
- At one time needed to obtain puberty
- Good early fertility

1800 x .65 = 1170
1400 x .65 = 910
1200 x .65 = 780

55% of mature weight

- Decrease heifer development cost
- Develop grazing skills
- Show compensatory gain on summer grass
- Riskier

1800 x .55 = 990
1400 x .55 = 770
1200 x .55 = 660

Reproductive Tract Scores (RTS)

RTS	Uterine horns	Ovarian Measurement (mm) Length	Height	Width	Ovarian Structures	Description
1	Immature < 20 mm diameter No tone	15	10	8	No palpable follicles	Infantile
2	20-25 mm diameter No tone	18	12	10	8 mm follicles	Prepubertal
3	20-25 mm diameter Slight tone	22	15	10	8-10 mm follicles	Peripubertal
4	30 mm diameter good tone	30	16	12	> 10 mm follicles CL possible	Cycling
5	> 30 mm diameter	> 32	20	15	CL present	Cycling

Adapted from Anderson et al., 1991

Table 1. Distribution of animals by ovarian score (OS) and uterine score (US) in Experiment 1.

		Ovarian score[1]				
		2	3	4	5	Total
Uterine Score[2]	2	0.6 % (3/469)	2.1 % (10/469)	0.0 % (0/469)	0.0 % (0/469)	2.8 % (13/469)
	3	1.5 % (7/469)	26.4 % (124/369)	1.1 % (5/469)	3.0 % (14/469)	32.0 % (150/469)
	4	0.0 % (0/469)	1.7 % (8/469)	4.1 % (19/469)	12.0 % (56/469)	17.7 % (83/469)
	5	0.4 % (0/469)	12.0 % (56/469)	0.4 % (2/469)	35.2 % (165/469)	47.6 % (223/469)
	Total	2.1 % (10/469)	42.2 % (198/469)	5.5 % (26/469)	50.1 % (235/469)	

Smith et al., 2022

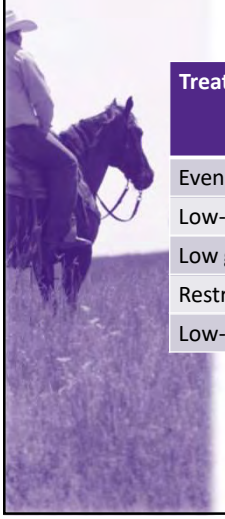
Table 6. Pregnancy outcomes of heifers by reproductive tract score[1] (RTS) in Experiment 2.

	Conceived to first AI service		Failed to become pregnant	
RTS[1]	%	Proportion	%	Proportion
2–3	60.9%[a]	14/23	17.4%[a,b]	4/23
3–2	62.2%[a]	23/37	13.5%[a,b]	5/37
3–3	68.9%[b]	2511/3643	14.4%[a,b]	526/3643
3–4	69.1%[b,c]	1215/1759	12.2%[a]	213/1749
4–3	66.7%[b,c]	1100/1649	13.7%[b]	226/1649
4–4	69.6%[c]	9938/14,274	13.9%[b]	1990/14,274

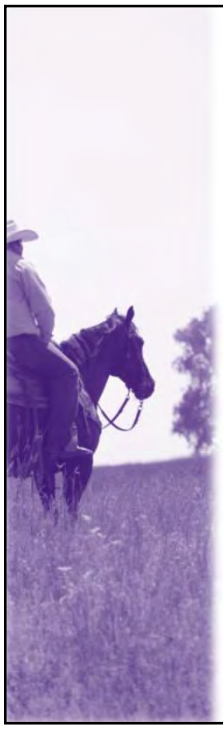
Smith et al., 2022

KSUbeef.org

Timing of Weight Gain

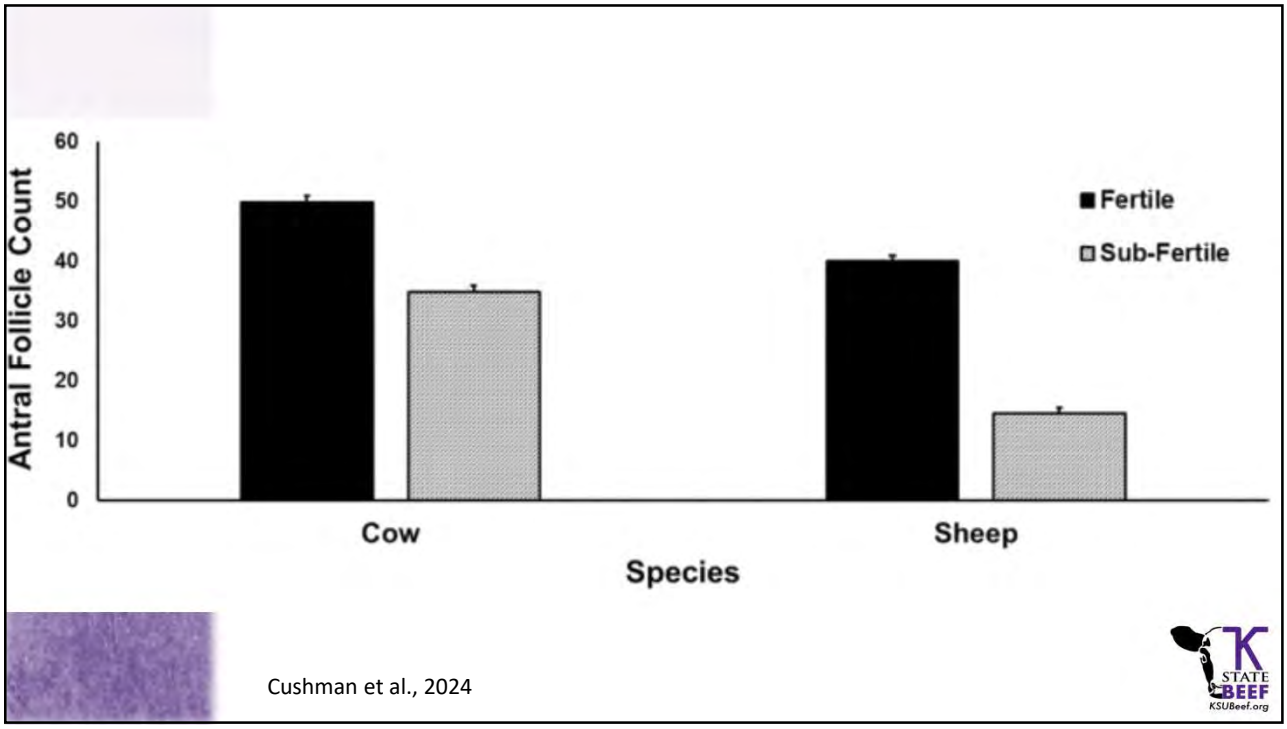


Treatment	Age at puberty	Heifer pregnancy rate	Mean calving date	2nd year pregnancy rate	Reference
Even gain vs Late gain	INCR	NS	_	_	Lynch et al., 1977
Low-High vs High	_	NS	NS	NS	Freetly et al., 2001
Low gain vs High gain	DECR	NS	NS	NS	Funston & Deutscher, 2004
Restricted vs Control	INCR	NS	_	_	Roberts et al., 2009
Low-High vs Constant	NS	NS	NS	_	Rosasco et al., 2017

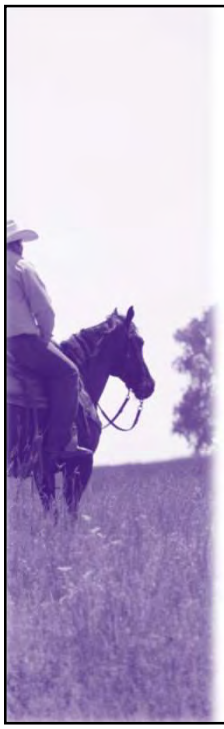


Ovarian Reserve

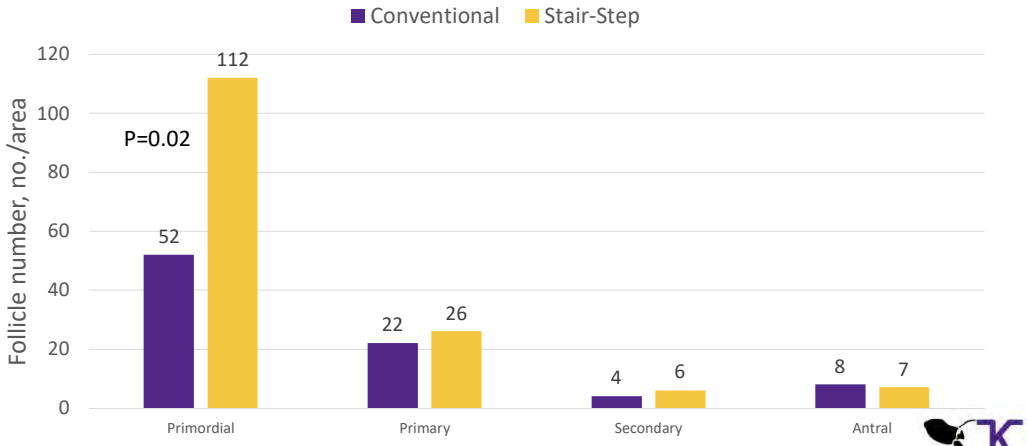
- Germ cells – number increases to mid-gestion then declines
- Positive association between antral follicle count and reproductive tract size



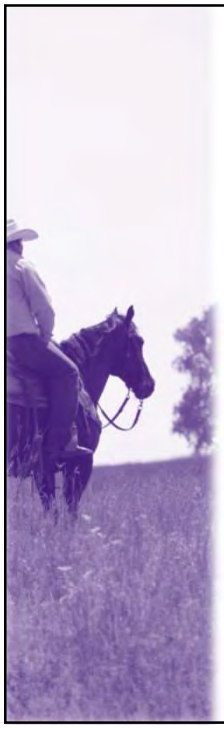
Cushman et al., 2024



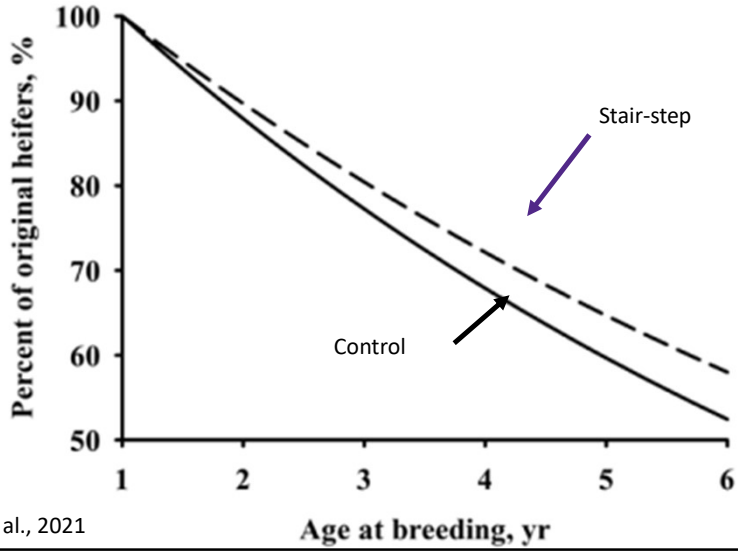
Control vs Stair-Step development diet and ovarian Histology



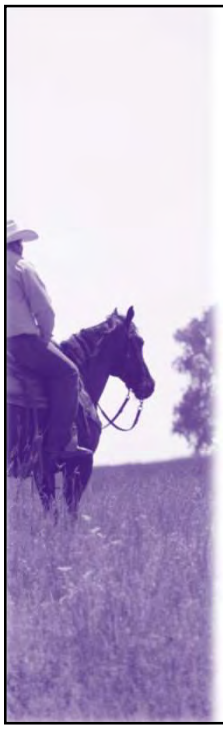
Freetly et al., 2014



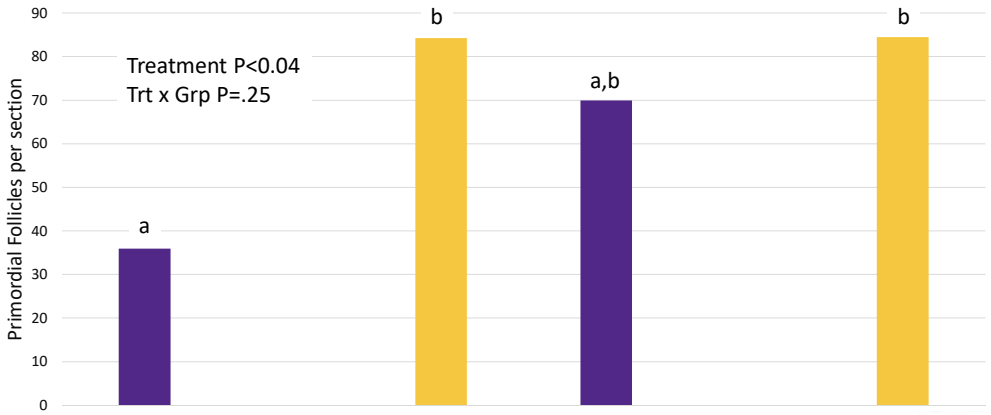
Retention of Cows Raised on Different Postweaning Growth Patterns



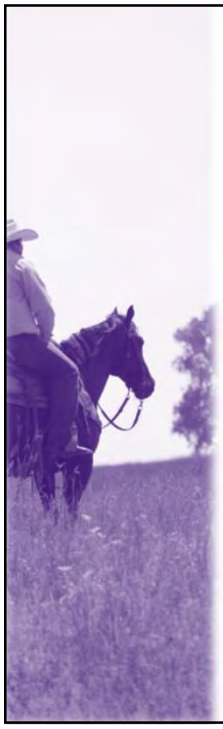
Freetly et al., 2021



Effect of Nutritional Programming on the Ovarian Reserve



Rosasco et al., 2020

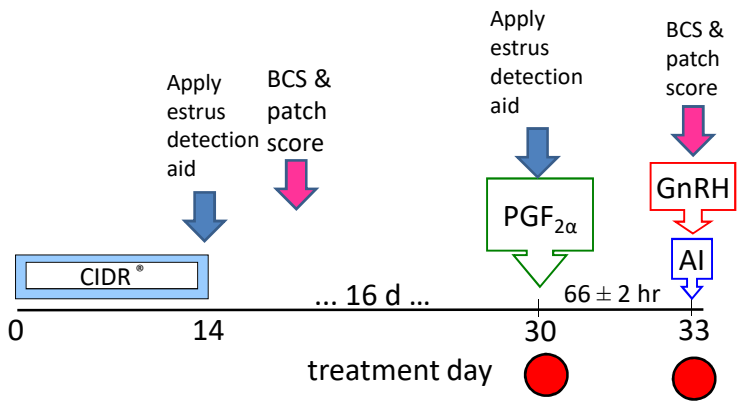


Implications

- Need to weigh heifers and balance diets

Objective to compare fertility in two systems

Year	Trt starting date	Days before CIDR	n
2017	3/16	-7	169
2018	3/7	-17	86
2019	3/24	0	97
2020	3/23	0	130



Serum sample

AI Preg Check 53 – 64 d after FTAI

2017 – 10 days to bull turnout
2018, 2019, 2020 – AI only

K-STATE Research and Extension

Nutrient content of triticale

Start of grazing

Year	CP	ADF	NDF
2017	28.8	22.5	45.2
2018	20.2	24.4	47.1
2019	29.3	20.8	41.4
2020	27.8	21.3	39.6

at AI

Year	CP	ADF	NDF
2017	22.1	28.8	47.8
2018	17.8	32	56.2
2019	15.0	34.1	55.3
2020	12.6	28.5	52.2

Drylot ration composition, % DM

	2017	2018	2019	2020
Forage Sorghum Hay	69.1		41.4	48.4
CRP Hay		32.2		
Triticale Silage		47.0	33.7	
DDG	21	10.7	12.8	
WDG				26.8
Corn	9.9	10.1	12.1	23.3
CP	11.7	10.7	10.6	14.1
Ne_m Mcal/lb	.70	.57	.69	.78
Ne_g Mcal/lb	.43	.31	.41	.49

K-STATE Research and Extension

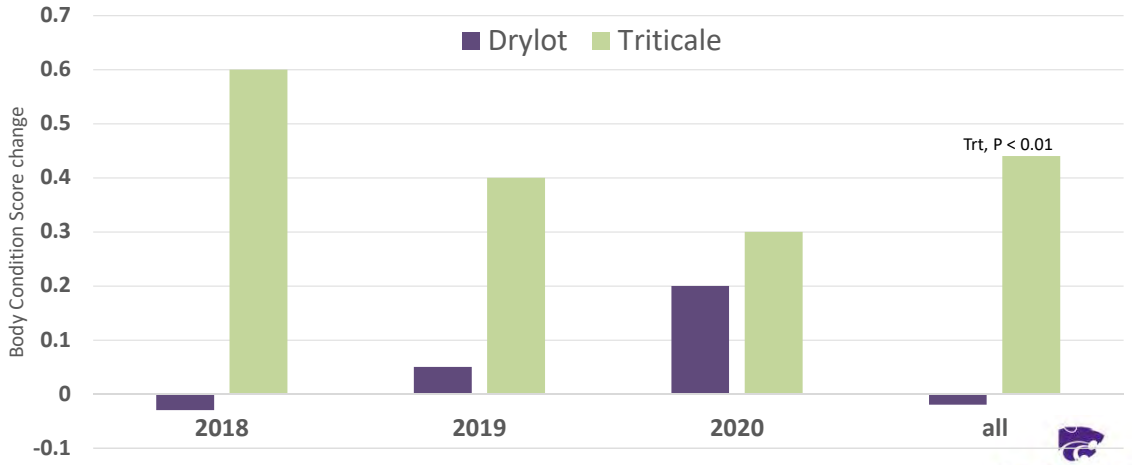
Weight gain during treatments

	Starting Weight		Ending Weight		ADG	
Year	Drylot	Triticale	Drylot	Triticale	Drylot	Triticale
2017	773	781	823	837	1.23	1.41
2018	813	797	887	888	1.48	1.82
2019	775	778	856	898	2.53	3.75
2020	662	656	766	755	3.15	3.02
all	757	752	834	**845**	2.16	**2.51**
Age		**		**		NS
Year		**		**		**
Trt		NS		*		*
Trt x Y		NS		*		*

* P<0.05; ** P<0.001; NS = no statistical difference

K-STATE Research and Extension

Body condition change from CIDR removal to AI



Trt, P<0.01
Trt x Yr, P<0.01

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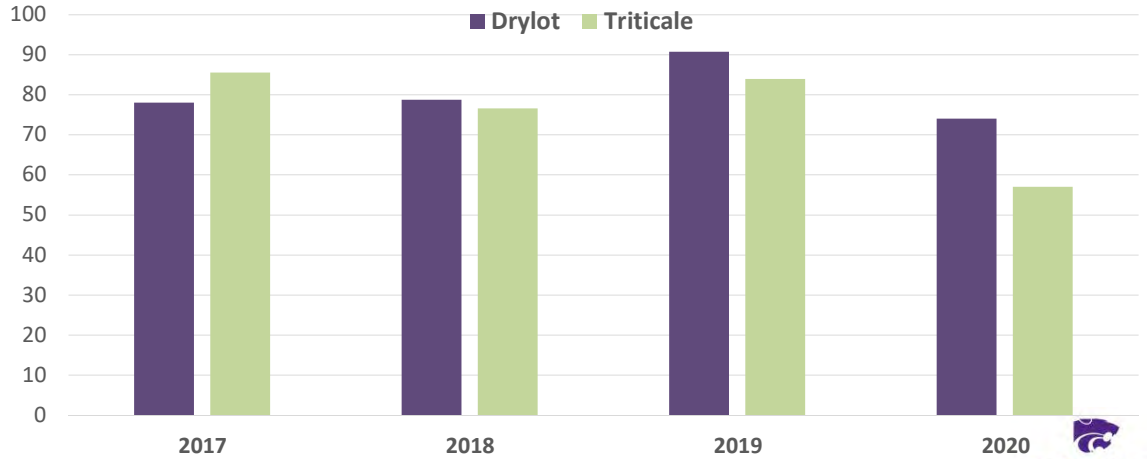
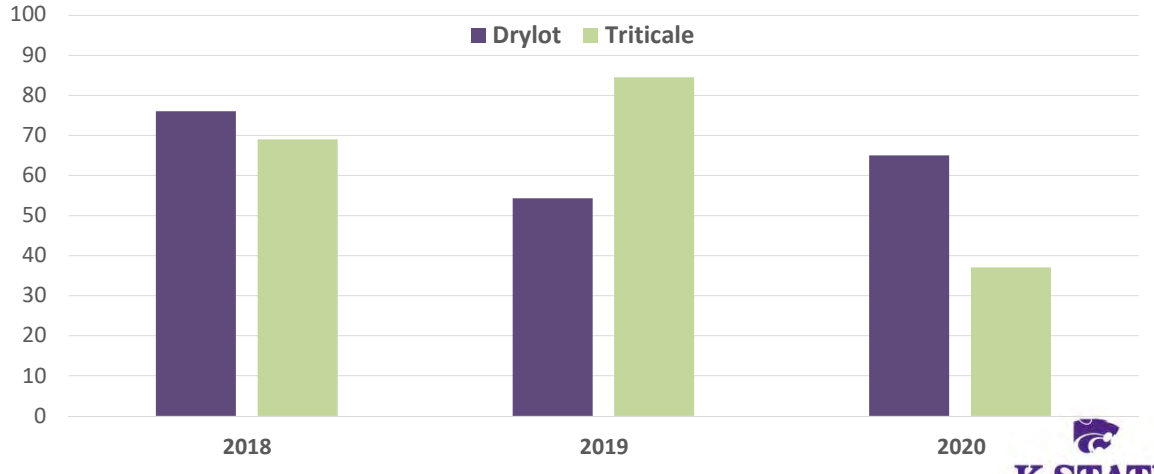
Proportion of heifers with patch scores of ≥ 3, 5 to 6 days after CIDR removal



Age P=.29, Year P=0.002, Treatment P=0.29, Treatment x Year P=0.14

K-STATE Research and Extension

Proportion of heifers with patch scores of ≥3 at AI.



Age P=.36, Year P=0.001, Treatment P=0.98, Treatment x Year P<.001

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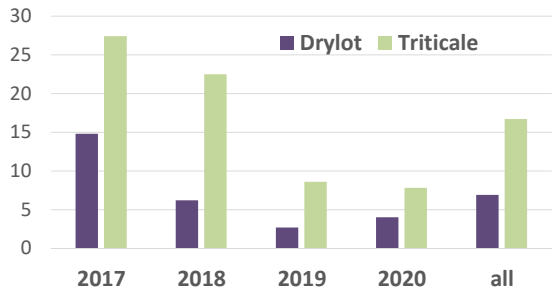
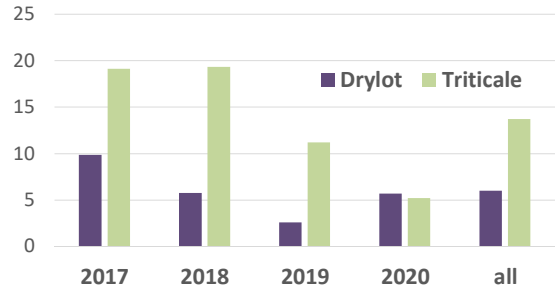
Blood Urea Nitrogen (mg/dL)

At PGF



Age	NS
Year,	**
Treatment	**
Trt x Year	**

At Insemination



Age	*
Year,	**
Treatment	**
Trt x Year	**

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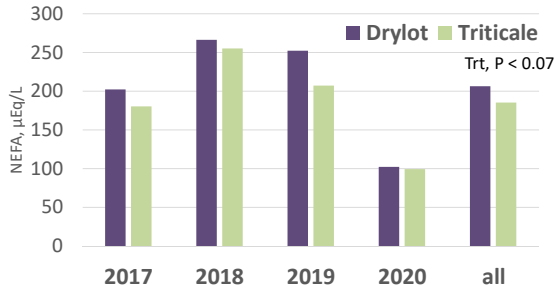
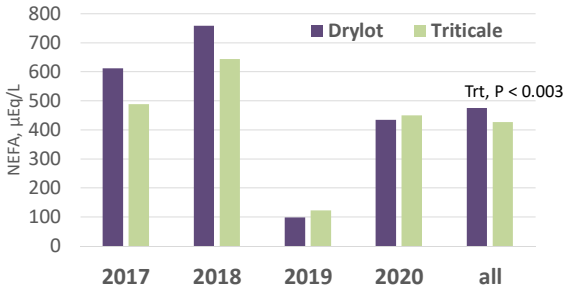
Concentrations of NEFA

At PGF



Year P<0.001, Treatment P=0.07, Treatment x Year P=0.21

At Insemination



Year P<0.001, Treatment P=0.003, Treatment x Year P<0.001

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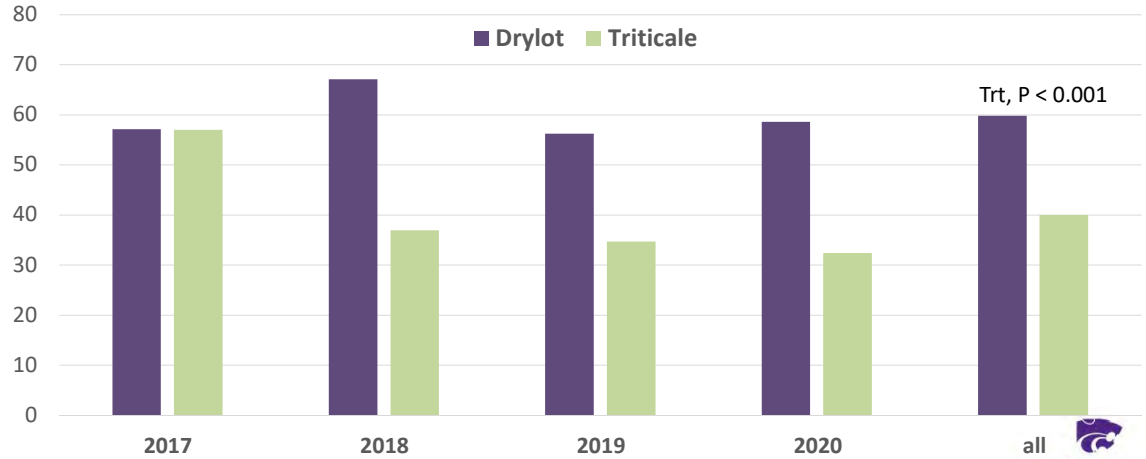
AI Pregnancy rate



Year P=.16; Treatment P<0.001, Treatment x Year P=0.05

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Summary

- Costs and longevity
- Early bred heifers have huge advantage / late bred heifers are high risk
- Pattern of gain
  - Cost
  - Some restriction seems beneficial in longevity
  - Target development wt may depend on risk tolerance
- Lush vs drylot System

K-STATE Research and Extension

Sandy Johnson, PhD
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www.KSUBeef.org

THE FINER POINTS OF DIAGNOSTIC INVESTIGATIONS OF COMMON FOOD ANIMAL TOXINS

SCOTT FRITZ
DVM, DABVT

LARGE ANIMAL

The Finer Points of Diagnostic Investigations of Common Food Animal Toxins

Scott Fritz DVM, DABVT
Clinical Assistant Professor of Toxicology
Kansas State University College of Veterinary Medicine

Diagnostic investigations involving food animals can be a frustrating endeavor. Often, the presenting complaint directed towards veterinarians is "found dead." Lack of clinical signs makes rapid identification of the affected organ system impossible. Furthermore, once an animal expires, the clock is already ticking regarding diagnostic sample quality. Issues that arise during the summer months are even more complicated by lack of daily observation and high ambient temperatures that can ruin a carcass in a couple of hours. This presentation addresses common challenges veterinarians can expect to encounter when working through these frustrating and sometimes catastrophic cases. The outline below should help remind readers of the discussion points from the presentation. It is important to recognize diagnostic medicine is always changing and many laboratories do not employ a veterinary toxicologist as part of the diagnostic team, much toxicology testing is done on a referral basis. As such, each situation should be approached independently and frequent communication with a diagnostic laboratory offers the best chance at a definitive diagnosis.

Nitrate/Nitrite

- Ideal diagnosis:
    - Brown discoloration of tissues
    - Elevated nitrate concentrations in ocular fluid
    - Elevated nitrate concentrations in the source
    - Confirmation of exposure to a source
- Challenges
    - The source may be totally consumed
    - The source may change prior to sampling
    - There may be a hot spot that cows consumed
    - Endogenous production of intraocular nitrate by postmortem bacteria
        - Assays cannot differentiate

Non-Protein Nitrogen

- Ideal Diagnosis
    - Response to therapy in sublethally-exposed cohorts
    - Basic rumen pH (>8)
    - Elevated ammonia in ocular fluid
    - Elevated NPN concentrations in the source
    - Evidence of accidental consumption
    - Clinical signs in minutes to hours after consumption of elevated NPN in the diet
- Challenges
    - Rumen pH reverts to normal over time

    - Ammonia is volatile so it disappears quickly
    - The source can change

Neurotoxins that can cause laminar cortical necrosis

- Ideal Diagnosis
    - Histologic diagnosis
    - Toxic concentrations of lead in liver/kidney or,
    - Toxic concentrations of sodium in the brain or,
    - Toxic concentrations of sulfur in the total diet including drinking water
- Challenges
    - Delayed sampling can ruin brain lesions
    - Death may be too rapid for brain lesions to develop
    - If only fixed brain is submitted, sodium quantification is not useful
    - Feed and water sulfur sources are additive, all need analyzed

Ionophores

- Ideal Diagnosis
    - Compatible timeline of events – off feed event, transient diarrhea
    - Histologic evidence of myocardial degeneration and necrosis
    - Toxic concentrations in feed
- Challenges
    - Often mis-diagnosed as respiratory disease early
    - Delay in clinical effects can be over a week after the over dose
    - Offending feed is often gone

Abortive toxins

- Ideal Diagnosis
    - Lack of identification of infectious etiologies
    - Proof of exposure in the dam
    - Clinical signs in the dam for certain toxins
    - Detection of compound in various fetal or maternal samples – toxin dependent
- Challenges
    - Robust rule outs of infectious causes
    - Abortion could be secondary issue
    - A test does not exist for everything

Plants

- Ideal Diagnosis
    - Confirmation of possible exposure
    - Compatible clinical signs, gross and/or histologic lesions
    - Confirmation of ingestion
- Challenges
    - When a plant is eaten it no longer exists

- Lack of daily observation on pasture
- Delay in sampling can limit histologic examination
- Delay in clinical signs can be months for certain plants
- Unknown toxic principle
- Lack of tests for toxic principle in others

BEEF CATTLE MINERAL NUTRITION

STEVE ENSLEY
BS,DVM, MS, PHD

ROBERT LARSON
DVM, PHD, DACT, DACVPM (EPIDEMIOLOGY), ACAN

LARGE ANIMAL

Beef Cattle Nutrition:
Mineral Nutrition

Dr. Bob Larson
Dr. Steve Ensley

Macrominerals

- Salt (NaCl) is the mineral needed consistently and in the largest amount (1 to 2 oz. daily)

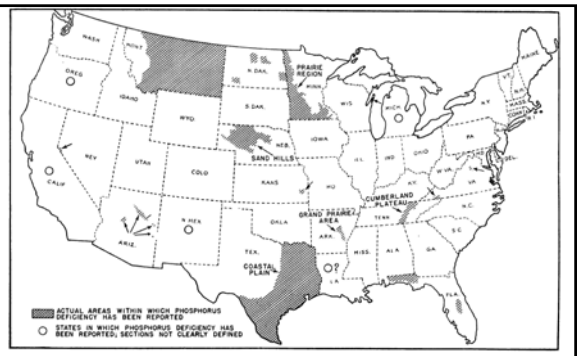
Macrominerals

- Calcium is usually supplied in adequate amounts in forage. Higher requirement during lactation

Macrominerals

- Phosphorus is deficient in some areas of U.S. and during some production phases (lactation)

  Maturity of forage affects supplementation needs



Phosphorus deficient soils (and plants) tended to be most problematic in the Northern Great Plains and Midsouth. States with low soil P include the Coastal Plain of Texas (southeastern 1/3 of Texas), Montana, Western Minnesota, portions of North Dakota, the Sandhills of Nebraska, and isolated areas in other states (particularly mid-south)

Macrominerals

- Magnesium supplementation is required when grazing wheat pasture or lush early-growth cool season grasses

  (↑potassium & ↓ magnesium)

Macrominerals

- Potassium is rarely required with a forage-based diet

  Deficiency reported with badly weathered hay

Microminerals

- Six trace minerals potentially deficient in forage-based diets:

Copper	Cobalt
Iodine	Selenium
Zinc	Manganese

Microminerals

- Copper
  - Molybdenum and sulfur levels impact copper utilization
  - Iron can also impact copper utilization

Copper Functions

- Formation of hemoglobin
- Incorporation into ceruloplasmin
- Iron movement
- Protection from oxidation
- Involvement in immune response (maybe not ruminants)

Copper Functions cont.

- Co-factor in many enzyme systems
  - Prostaglandin synthesis
  - Collagen and elastin synthsis
  - Conversion of L-tyrosin to melanin

Signs of Copper Deficiency

- Reduced fertility (male and female)
- Increased risk of retained placenta
- Increased risk of abomasal ulcers
- Iron anemia
- Ataxia and dummy calves
- Foot problems (cracks, abscesses, etc.)

Signs of Copper Deficiency

- Grey hair coat
- Poor performance / wt. gain
- Impaired immune response - poor response to vaccination
  (maybe not ruminants)

Copper Antagonists

- Molybdenum
  - Usually associated with alkaline soil
  - Legumes accumulate more than grasses
  - Cu:Mo ratio should be >6:1
    - Borderline ratio of 2-3:1
    - Toxic levels <2:1

Copper Antagonists

- Sulfate
  - Sulfates and molybdenum both needed to form thiomolybdates
- Iron
- Other minerals: phosphorus, zinc, lead, calcium, cobalt, mercury, selenium, tin, silver, tungsten...

Copper Antagonists

- Others
  - Protein
  - Estrogen
  - Nitrates

Microminerals

- Cobalt
  - Not much is known ?

Microminerals

- Iodine
  - Deficiencies seldom reported ?
- Selenium
  - Deficiency and toxicity reported (activity tied to Vit E)
  - Areas in Kansas

Microminerals

- Zinc

  Reported to be most commonly deficient in forage

- Manganese

  Occasionally deficient – calf defects
  Very poorly available from forage

Suspected trace mineral problems should be investigated not as a single element problem, but as an imbalance of several or all minerals

Factors in Mineral Imbalances

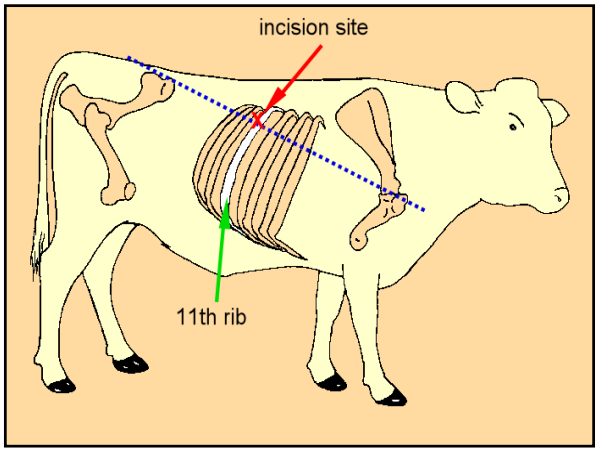
- Breed
- Growth rate
- Milk production
- Feed source
- Water source
- Crop / feed production practices

Microminerals

- If a trace mineral deficiency is suspected - a thorough diagnostic work-up is required

  Feed and water analysis

  Liver and serum sampling



Microminerals

- Supplementation Baseline

  Use sources available to animal
  (i.e. Copper oxide is poorly available)

Micromineral Forms

- Chelated minerals

Mineral chelated to 2 amino acids
Industry definition is not uniform
Much more expensive
Theory has some logic
Scientific data to support is lacking

Microminerals

- Supplementation Baseline

Provide 50% of NRC recommended levels of Cu, Zn, Co, Se, and I

Provide 100% of NRC recommended level of Mn

Interesting Case:

Too many open cows…..

KANSAS STATE UNIVERSITY

Introduction: First Contact

August 2020: Phone call from veterinarian in North Dakota. A client is concerned about poor fertility and thinks the problem may be copper deficiency. The veterinarian wants to know if I can help her interpret the mineral analysis.

Larson: Yes, I'll be glad to help, and I am going to pull Dr. Steve Ensley into the discussion as well. Please send me the lab reports and a history of the situation.

From: [redacted]
Sent: Thursday, August 13, 2020 11:02 AM
To: Robert Larson <rlarson@vet.k-state.edu>
Subject: [redacted] liver biopsy mineral analysis

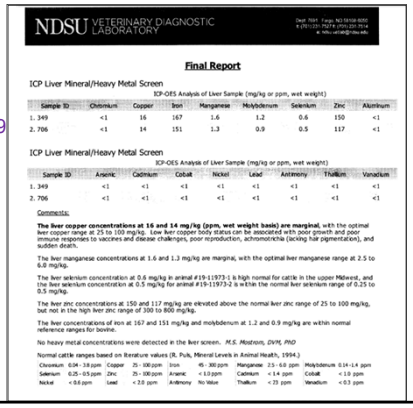
Dr. Larson,
Attached are a water sample report, mineral records for the biggest group of cattle (a high percentage of these are first and second calf heifers), the tag from the mineral they are feeding and also the liver biopsy report done on 11-22-19, and the recent liver biopsy results.

[redacted] is a "silver bullet" kind of client. He thinks copper deficiency is the issue with his cattle's low fertility rates and he decided to test these calves without necessarily our input but the feed company advised him to. He has commercial calves by Berlin, ND which is SE corner. That soil is alkaline high typically. What Dr. Hall from Utah had advised was to liver biopsy the cows at PG and then formulate a ration at that time to correct their imbalance. Richard wants to help his calves from this year now and also get an idea of a plan/know if what he was feeding was inadequate.

They are not feeding any DDG's.
Please let me know if you need any other information. Thank you.
[redacted]

Introduction: First Contact

Liver samples from 2 calves taken 11/18/2019



NDSU VETERINARY DIAGNOSTIC LABORATORY

Final Report

ICP Liver Mineral/Heavy Metal Screen

ICP-OES Analysis of Liver Sample (mg/kg or ppm, wet weight)

Sample ID	Chromium	Copper	Iron	Manganese	Molybdenum	Selenium	Zinc	Aluminum
1. 349	<1	16	167	1.6	1.2	0.6	150	<1
2. 706	<1	14	151	1.3	0.9	0.5	117	<1

ICP Liver Mineral/Heavy Metal Screen

ICP-OES Analysis of Liver Sample (mg/kg or ppm, wet weight)

Sample ID	Arsenic	Cadmium	Cobalt	Nickel	Lead	Antimony	Thallium	Vanadium
1. 349	<1	<1	<1	<1	<1	<1	<1	<1
2. 706	<1	<1	<1	<1	<1	<1	<1	<1

Comments:

The liver copper concentrations at 16 and 14 mg/kg (ppm, wet weight basis) are marginal, with the optimal liver copper range at 25 to 100 mg/kg. Low liver copper body status can be associated with poor growth and poor immune responses to vaccines and disease challenges, poor reproduction, achromotrichia (lacking hair pigmentation), and sudden death.

The liver manganese concentrations at 1.6 and 1.3 mg/kg are marginal, with the optimal liver manganese range at 2.5 to 6.0 mg/kg.

The liver selenium concentration at 0.6 mg/kg in animal #19-11973-1 is high normal for cattle in the upper Midwest, and the liver selenium concentration at 0.5 mg/kg for animal #19-11973-2 is within the normal liver selenium range of 0.25 to 0.5 mg/kg.

The liver zinc concentrations at 150 and 117 mg/kg are elevated above the normal liver zinc range of 25 to 100 mg/kg, but not in the high liver zinc range of 300 to 800 mg/kg.

The liver concentrations of iron at 167 and 151 mg/kg and molybdenum at 1.2 and 0.9 mg/kg are within normal reference ranges for bovine.

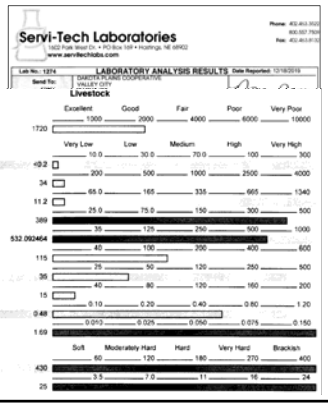
No heavy metal concentrations were detected in the liver screen. *M.S. Mostrom, DVM, PhD*

Normal cattle ranges based on literature values (R. Puls, Mineral Levels in Animal Health, 1994.)

Chromium	0.04 - 3.8 ppm	Copper	25 - 100 ppm	Iron	45 - 300 ppm	Manganese	2.5 - 6.0 ppm	Molybdenum	0.14 -1.4 ppm
Selenium	0.25 - 0.5 ppm	Zinc	25 - 100 ppm	Arsenic	< 1.0 ppm	Cadmium	< 1.4 ppm	Cobalt	< 1.0 ppm
Nickel	< 0.6 ppm	Lead	< 2.0 ppm	Antimony	No Value	Thallium	< 25 ppm	Vanadium	< 0.3 ppm

Introduction: First Contact

Water sample taken 12/16/2019



Servi-Tech Laboratories
1902 Fole West Dr. • PO Box 169 • Hastings, NE 68902
www.servitechlabs.com

Lab No. 1274 — LABORATORY ANALYSIS RESULTS — Date Reported: 12/18/2019
Send To: DAKOTA PLAINS COOPERATIVE, VALLEY CITY

Livestock

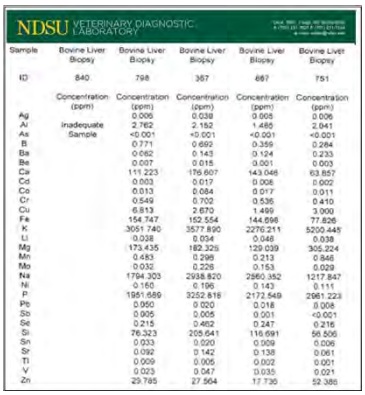
		Excellent	Good	Fair	Poor	Very Poor
Total Dissolved Solids (Calc) (TDS), mg/L	1720	1000	2000	4000	6000	10000

		Very Low	Low	Medium	High	Very High
Nitrate+Nitrite Nitrogen (NO3+NO2-N), mg/L	<0.2	10.0	30.0	70.0	100	300
Sulfate (SO4), mg/L	34	200	500	1000	2500	4000
Total Sulfur (S), mg/L	11.2	65.0	165	335	665	1340
Total Sodium (Na), mg/L	389	25.0	75.0	150	300	500
Chloride (Cl), mg/L	532.092464	35	125	250	500	1000
Total Calcium (Ca), mg/L	115	40	100	200	400	600
Total Magnesium (Mg), mg/L	35	25	50	120	250	500
Total Potassium (K), mg/L	15	40	80	120	160	200
Total Iron (Fe), mg/L	0.48	0.10	0.20	0.40	0.80	1.20
Total Manganese (Mn), mg/L	1.69	0.013	0.025	0.050	0.075	0.150

		Soft	Moderately Hard	Hard	Very Hard	Brackish
Hardness (CaCO3), mg/L	430	60	120	180	270	400
Hardness (CaCO3), grains/gal	25	3.5	7.0	11	16	24

Introduction: First Contact

Liver samples from 5 calves taken 07/29/2020



NDSU VETERINARY DIAGNOSTIC LABORATORY

Sample	Bovine Liver Biopsy	Bovine Liver Biopsy	Bovine Liver Biopsy	Bovine Liver Biopsy	Bovine Liver Biopsy
ID	840	798	367	667	751
	Concentration (ppm)	Concentration (ppm)	Concentration (ppm)	Concentration (ppm)	Concentration (ppm)
Ag		0.005	0.038	0.005	0.006
Al	Inadequate Sample	2.762	2.152	1.485	2.041
As		<0.001	<0.001	<0.001	<0.001
B		0.771	0.669	0.359	0.264
Ba		0.062	0.143	0.124	0.233
Be		0.007	0.015	0.001	0.003
Ca		111.223	176.607	143.046	63.857
Cd		0.003	0.017	0.008	0.002
Co		0.013	0.084	0.017	0.011
Cr		0.549	0.702	0.535	0.410
Cu		6.813	2.670	1.499	3.000
Fe		154.747	152.554	144.698	77.826
K		3051.740	3577.890	2276.211	5200.445
Li		0.038	0.034	0.048	0.038
Mg		173.435	182.325	129.039	305.224
Mn		0.483	0.296	0.213	0.846
Mo		0.032	0.228	0.153	0.029
Na		1794.303	2938.820	2860.352	1217.847
Ni		0.160	0.196	0.143	0.111
P		1951.589	3232.818	2172.549	2961.223
Pb		0.050	0.020	0.018	0.008
Sb		0.005	0.005	0.001	<0.001
Se		0.215	0.462	0.247	0.216
Si		76.323	205.641	116.691	56.506
Sn		0.033	0.020	0.009	0.006
Sr		0.092	0.142	0.138	0.061
Ti		0.009	0.005	0.002	0.001
V		0.023	0.047	0.035	0.021
Zn		29.785	27.564	17.736	52.386

Introduction: First Contact

Current mineral supplement

DLF BRDR 8P MINERAL
FOR BEEF CATTLE
CAUTION: USE ONLY AS DIRECTED
GUARANTEED ANALYSIS

4671984-106

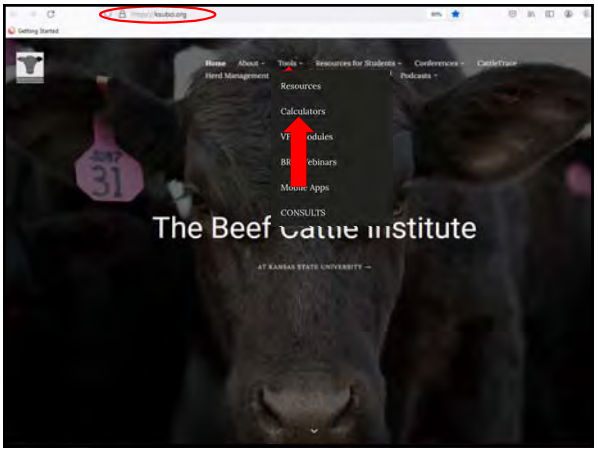
Calcium (CA) (MIN)	11.0000 %
Calcium (CA) (MAX)	13.0000 %
Phosphorus (P) (MIN)	8.0000 %
Salt (NACL) (MIN)	11.0000 %
Salt (NACL) (MAX)	13.0000 %
Magnesium (MG) (MIN)	2.0000 %
Potassium (K) (MIN)	1.0000 %
Manganese (MN) (MIN)	2500.0000 PPM
Copper (CU) (MIN)	2000.0000 PPM
Iodine (I) (MIN)	110.0000 PPM
Selenium (SE) (MIN)	30.0000 PPM
Zinc (ZN) (MIN)	6000.0000 PPM
Vitamin A (MIN)	200000.00 IU/LB
Vitamin D-3 (MIN)	12000.00 IU/LB
Vitamin E (MIN)	250.00 IU/LB

Next Steps:

What do you think?

What would you say/ask?

What would you do?



Put

Evaluating Mineral Feed Tags for Beef Cows
College of Veterinary Medicine
Kansas State University

er

Mineral			Will supply to total diet		Percentage of NRC Recommendation	NRC Reccommendation
Calcium	12.0	%	0.101	%	37%	0.27%
Phosphorus	8.0	%	0.067	%	29%	0.23%
Salt	11.0	%	0.093	%	132%	0.07%
Magnesium	2.0	%	0.017	%	17%	0.10%
Potassium	1.0	%	0.008	%	1%	0.65%
Cobalt	0	ppm	0.000	ppm	0%	0.10
Copper	2000	ppm	8.917	ppm	89%	10.00
Iodine	110	ppm	0.777	ppm	155%	0.50
Manganese	2500	ppm	9.590	ppm	24%	40.00
Selenium	30	ppm	0.115	ppm	115%	0.10
Zinc	6000	ppm	36.343	ppm	121%	30.00
vit A	200,000	IU/lb	43,750	IU	95%	46,091
vit D	12,000	IU/lb	2,625	IU	81%	3,250
vit E	250	IU/lb	55	IU	9%	591
thiamine	0	IU/lb	0	IU		

Zinc (Zl
Vitamin
Vitamin
Vitamin

PM
/LB
/LB
/LB

Next Steps:

- Bulls need attention – both pre-breeding BSE and continual monitoring during the breeding season.
- Need to address “momentum” by calving heifers ahead of cows and making sure BCS is good going into breeding
- I realize that the producer is concerned that trace mineral issues are contributing to the poor reproductive success. Although I don’t have the complete history and I have not been on the property, based on the patterns provided by the history, if a copper deficiency problem exists – I doubt that it is the primary problem.
- Continue to monitor liver copper and other minerals – key time points are near the start of the breeding season & at weaning.
- Try to locate some higher-copper and higher-manganese supplements (without raising selenium) - if commercial supplements aren’t available, we can help calculate a custom mineral mix.

Another Case:

Producer concerned that cows are not cycling…..

KANSAS STATE UNIVERSITY

Producer is very concerned about nutrition

Producer is very concerned about nutrition – particularly mineral nutrition as a cause of his poor reproductive efficiency.

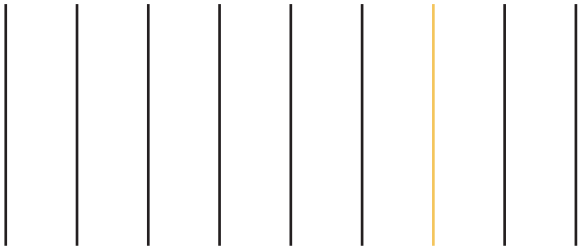
The 3 Minerals Have Important Differences

Jan 2018 2015-2016 May 2017

	Rangeland Pro Breeder Min 8 Availa 4	Rangeland Pro Breeder	Custom Ruffage Mate (8 oz /d)
Calcium	36%	36%	34%
Phosphorus	28%	32%	16%
Salt	191%	0%	58%
Magnesium	16%	2%	56%
Potassium	1%	7%	3%
Cobalt	187%	461%	38%
Copper	47%	22%	23%
Iodine	82%	59%	68%
Manganese	17%	10%	14%
Selenium	100%	100%	46%
Zinc	62%	31%	71%
Vitamin A	183%	183%	78%
Vitamin D	972%	972%	111%
Vitamin E	18%	18%	6%

DIAGNOSING REPRODUCTIVE FAILURE IN BEEF HERDS & / OR ASSESSING REPRODUCTIVE PERFORMANCE IN BEEF HERDS

TERRY ENGELKEN
DVM, MS



LARGE ANIMAL

INVESTIGATING THE CAUSES OF REPRODUCTIVE FAILURE IN BEEF HERDS

Terry J. Engelken, DVM MS
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College of Veterinary Medicine
Iowa State University
Ames, IA 50011

Abstract

Reproductive efficiency is still the most important output factor affecting profitability of the cow/calf enterprise. While reproductive performance can be negatively impacted by many factors, infectious disease often plays a pivotal role in those situations where suboptimal reproductive performance is demonstrated. Early embryonic death, late-term abortion, delayed conception and "weak calf syndrome" may all be manifestations of a disease outbreak. Regardless of the cause, the end result is that the operation will have fewer pounds of calf to market after weaning. The practitioner plays a central role in understanding the relationship of infectious agents with the risk of exposure and the timing of gestational losses in order to utilize the proper diagnostics and determine why reproductive losses are occurring. Then herd health protocols can be built to minimize these losses in the future.

Utilization of Diagnostics

The diagnosis of reproductive failure may be one of the most frustrating undertakings for the practitioner and producer. In cases of abortion, an etiologic diagnosis is made less than 50% of the time.[1] Adequate information, including a herd history, with complete to a diagnostic laboratory is the most important step in a obtaining a definitive diagnosis.[2] Since diagnostic laboratories have different capabilities in handling an abortion case, it is prudent have a close working relationship with our diagnostic lab. Consultations with the pathologist are sometimes necessary to develop a systematic approach to working up reproduction losses. Submission of the appropriate tissue and body fluid samples will increase the chances of obtaining useful information. A review of the common pathogens, their timing of gestational loss, and commonly used testing modalities has been published.[3] However, the practitioner must keep in mind that there are many *noninfectious* causes of reproductive loss in cattle.

Serology has long been used as a diagnostic tool to define disease response in the individual animal and the movement of an organism within a population. When dealing with cases of abortion, serology must be interpreted with great caution.[3] Single serum samples have little to no value when diagnosing abortions as it is difficult to differentiate titers that arise from vaccination or natural exposure. However, a lack of titer may serve to rule out certain diseases. Paired sera may have limited value as well. Many of the bacterial and viral pathogens that cause abortion may infect the fetus or placenta long before the abortive event occurs. This lag time between infection and abortion may prevent the practitioner from detecting the rising or falling titers associated with the initial infection. This leads to the collection of two "convalescent" serum samples that will fail to detect the increase in antibody titer, if it indeed occurred. This is especially true when only affected females are sampled at the time when the abortion is noted. Overall, the time of seroconversion is dependent on the exposure of the agent and the amount of immunity established prior to the breeding and throughout gestation. Paired sera are much more useful when it is used as part of a complete diagnostic work-up that includes samples from the placenta, fetus, and fetal fluids.

Serologic profiling is one option to optimize the use of serologic testing. The basis of serologic profiling is analyzing titers from affected/aborted and nonaffected dams over the same time period.[2] It is

unclear how many samples are needed, but some suggest that the same number of affected and nonaffected animals, preferably at same stage of gestation and age, is adequate.[4] In herds with chronic gestational losses serum may be collected and frozen from a statistically relevant number of cows for future testing as needed. These frozen samples may be collected as the females are processed prior to breeding and/or at the time of pregnancy examination. Then, as fetal loss is detected, banked serum samples can be submitted along with acute and convalescent samples to provide a clearer serologic picture of the affected animals and their normal cohorts. This should give a more complete picture of when seroconversion occurred and what pathogens were involved.

In some cases, all samples will have elevated titers due to endemic infections of specific agents. For example, in herds endemically infected with bovine viral diarrhea virus (BVDV), all animals may have seroconverted yet have no clinical evidence of etiologic diagnosis of abortions.[5] In some instances, fetal/precolostral serology may be beneficial. Fetuses must be immunocompetent for specific agents (*Toxoplasma gondii*/*Neospora caninum*/BVDV/infectious bovine rhinotracheitis/*Brucella*) to produce serologic evidence in fetal fluids.[1,2,6] However, in some cases of premature placental separation, maternal serum antibodies may "leak" into the fetal circulation and give a false positive result on serology. While serology may be valuable in certain circumstances, the interpretation of these results should be carefully assessed as it relates to the entire diagnostic work-up and clinical signs in the herd.

Vaccination Protocol Design

While vaccines represent an important tool in protecting reproductive performance, they tend to be somewhat underutilized in beef herds.[3] When designing protocols to immunize the beef breeding herd against reproductive pathogens, there are several other important factors to consider. The potential at-risk level of the herd should be considered not only from the entry of potential pathogens, but also from the standpoint of the current disease level in the resident herd, different management groups on the ranch, breeding animal movement, and the potential side effects of the immunizing agents. While complete protection against every pathogen in every individual is not realistic, the goal would be to minimize the number of susceptible animals in the population. This should prevent epidemic outbreaks of reproductive disease as well as the establishment of chronic endemic losses in the cow herd.

While veterinarians and producers often think of individual vaccination protocols for different management groups on the ranch, vaccination programs should be viewed as a *continuum*. For example, if producers are developing their own replacement heifers, the suckling calf vaccination program should be viewed beyond the summer grazing season and fall weaning events. This vaccination program should be constructed to consider the probability that these young heifer calves will join the replacement pool, become pregnant, and eventually become a productive member of the mature herd. The suckling calf protocol should be designed to prepare the calf for post-weaning disease challenges and increase the calf's response to subsequent reproductive vaccination. Research has clearly shown that calves vaccinated at an early age will mount a cell-mediated immune response that will enhance the calf's ability to respond to subsequent vaccination or disease challenge. This approach will maximize protection against reproductive pathogens and minimize the potential for any negative vaccine side effects associated with the pre-breeding vaccination of seronegative females. These side effects may include multifocal areas of ovarian necrosis, hemorrhage and inflammatory cell infiltrate in the ovary, as well as the development of cysts in the corpus luteum. These lesions are transitory in nature, but can result in decreased reproductive performance in the short term.

Other factors to consider in vaccine selection include fetal protection and duration of immunity.[3] Recent advances in vaccine technology and diagnostic testing have allowed vaccine manufacturers to document the ability of their products to prevent disease organisms from spreading to the placenta and fetus following maternal infection. Challenge studies using virulent BVDV, infectious bovine rhinotracheitis (IBR), and *Leptospira borgpetersenii* (serovar *hardjo*) have shown that fetal protection against pregnancy wastage, BVDV persistent infection (PI), and leptospiral renal colonization and urine

shedding is possible following vaccination. While both modified-live viral and killed virus vaccines have demonstrated fetal protection against BVDV, typically modified-live vaccines provide better protection and a longer duration of immunity. Studies have also shown that this protection can last for 1 year or longer following vaccination of animals of various ages. The concepts of fetal protection and duration of immunity are especially important for beef operations as they are more likely to come in contact with adjacent herds and may only be handled for vaccination once per year.

Before constructing any vaccination program for a cow/ calf operation, the potential risk for exposure of the herd to a particular pathogen through herd additions or herd contact with clinical or inapparent carriers of a pathogen should be evaluated. The epidemiological terms "open," "closed," and "modified open" have been used to describe the potential risk level of a given herd.[3] When assessing the need for vaccination, factors such as risk-level management, the magnitude and etiology of previous reproductive losses, herd working patterns and animal management, and the producer's long-term goals should all be considered. Once this information is collected and evaluated, recommendations concerning the use of specific vaccine antigens, the type of vaccine needed, and the frequency of vaccination can be constructed to fit within the confines of the total ranch management plan.

Summary

The diagnosis of reproductive losses in beef herds can be frustrating. This is due in part to our inability to collect needed samples in a timely fashion, the immunological response of cattle to the pathogen, and the multitude of noninfectious causes of abortion. Whenever possible, diagnostic information should be combined biosecurity practices and vaccination to prevent reproductive loss. The goal of the immunization program should be to increase the level of collective herd immunity by minimizing the number of animals that are susceptible to reproductive disease. This will prevent not only epizootic outbreaks of pregnancy wastage, but should also control chronic endemic disease. The end result is that the practitioner can provide the client with cost-effective vaccine options to help insure optimum reproductive performance.

References:

1. Anderson, M. (2007). Infectious causes of bovine abortion during mid- to late-gestation. *Theriogenology* 68:474-486.

2. Holler, L. (2012). Ruminant abortion diagnostics. *Vet Clinics North Am: Food Animal Pract* 28:407-418.

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6. Iowa State University College of Veterinary Medicine, Veterinary Diagnostic Laboratory. Abortion serology. Available at http:// vetmed.iastate.edu/diagnostic-lab/user-guide/pathology# abortion_serology.

MANAGING DIGITAL DERMATITIS IN FEEDYARDS

TERRY ENGELKEN
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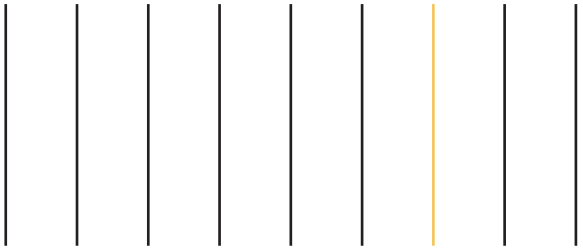

LARGE ANIMAL

Managing Digital Dermatitis in Feedyards

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Introduction

Reports of "hairy heel warts" or digital dermatitis (DD) have been described in cattle beginning in the early- to mid-1970's. While first described in dairy cows, more recent reports have centered on the development of DD in beef cattle operations. Over the years, this disease has been referred to as interdigital dermatitis, interdigital papillomas, Mortellaro's disease, and strawberry foot. While called by different names, the hallmark of this disease include well defined lesions on the heel that show erosions and ulceration, protruding wart-like structures, skin bearing thickened and elongated hairs, and the skin at the border of the lesion is thickened. Due to the location and relative lack of swelling, it is relatively easy to differentiate DD from other causes of lameness.

Cause of the Disease

A type of bacteria called *Treponema* have classically been blamed for the disease. This is based on consistent testing results that grew or identified the bacteria in the lesions. *Treponema* is a group of bacteria and should not be considered as one organism. There are at least five different *Treponema* organisms that are consistently isolated from DD lesions, but many others have also been identified. However, it is becoming more evident that DD lesions should be considered more like a "complex" that involves multiple species of bacteria, the immune response at the skin level, and environmental conditions.

Recent work done in dairy cattle at the College of Veterinary Medicine at Iowa State has shown that the bacterial population changes as the lesions move from early to late (chronic). DD lesions were sampled and profiled for bacterial DNA in order to determine differences in the populations of bacteria as these lesions aged. There were at least 11 different bacterial *families* represented in these lesions and the combination of bacteria changed dramatically as these lesions aged. While there was no indication of involvement of viruses or fungi, it is very clear that focusing on a single bacterium (such as *Treponema*) will not solve the puzzle of DD lesion development. Work at ISU CVM in feedlot cattle has shown very similar bacterial changes as dairy cattle.

The development of DD lesions in feedyards has not been well defined. It is common for lameness to be exhibited close to the normal reimplant date (90-120 DOF) but that can be highly variable. Lesions tend to worsen the last 60-90 DOF. Calves that have been through backgrounding programs / yards may arrive at the feedyard with active lesions. Other factors such as comingling, breed or genetics, history of feeding dairy animals, or size of the cattle may also have an impact of the prevalence of DD. It is believed that any factor that negatively impacts the integrity of the skin on the animal's heel can increase the likelihood of lesion development. Extremely wet pen conditions, excess manure buildup, exposed concrete edges, or rough surfaces can trigger pen outbreaks of this disease. Once established in the pen environment, it is difficult to eliminate the disease from the facility.

Treatment and Prevention

According to the American Association of Bovine Practitioners, a range of topical antibiotics are effective, but all are extra-label uses in the United States and require veterinary oversight. In individual cases, the lesion should be cleaned and dried and the antibiotic applied with a dressing or topical spray.

For topical spray treatments, oxytetracycline (mixed at 10–25mg/ml) or lincomycin (mixed at 1–8mg/ml) are effective when mixed with water in a 2–4 gallon hand sprayer and applied once or twice daily for 5–7 days. Alternatively, the prepared solution of oxytetracycline or lincomycin can be soaked into a gauze swab and wrapped on the lesion. Injectable antibiotics may be indicated for severe lesions, especially those on the dorsal aspect of the claw, but they are secondary to topical treatments and should not be used alone. In severe cases, a pain relief may also be indicated.

A range of different products are effective including copper sulfate (5%), zinc sulfate (5–10%), formalin (2–5%), and commercial chemicals containing quaternary ammonium compounds, organic acids, and other disinfectants. Recently, several new products which serve to activate copper sulfate have been released which allow lower concentrations (2%) to be used. It is essential that the volume of the foot bath is known so that the correct amount of chemical may be used to provide the appropriate final concentration. The volume in gallons may be calculated from the formula; length × width × depth (in inches) divided by 231. The University of Wisconsin has a "footbath calculator" available online that will match footbath dimensions with the proper amount of needed additives. Foot baths should be at least 8 feet long and 5 inches deep to ensure that enough contact is made between the chemical and the lesions. There are various options and locations that will work in a feedyard setting. The key is to put these in high traffic areas where the calves must walk through them with enough access to make recharging the bath easier. Minimizing the amount of manure on the feet will decrease the organic material tracked into the footbath and make the solution last longer.

Footbath frequency and solution selected will vary depending upon the cattle handling facilities, safety of the people working the cattle, and the percent of the pen that is affected.

Prevention still centers around the pen environment and avoiding negative impacts on the heel area of the calves. We have found that decreasing the moisture in the pen by more aggressive cleaning and decreasing animal density can be helpful. Scraping outdoor lots to remove manure and smooth out frozen hoof prints should improve foot health. Close observation of the feet of newly arrived cattle and recording their source can potentially identify problem sets of calves at arrival. Running cattle through a footbath at arrival should also be considered if active lesions are suspected in new cattle.

Future Needs and Direction

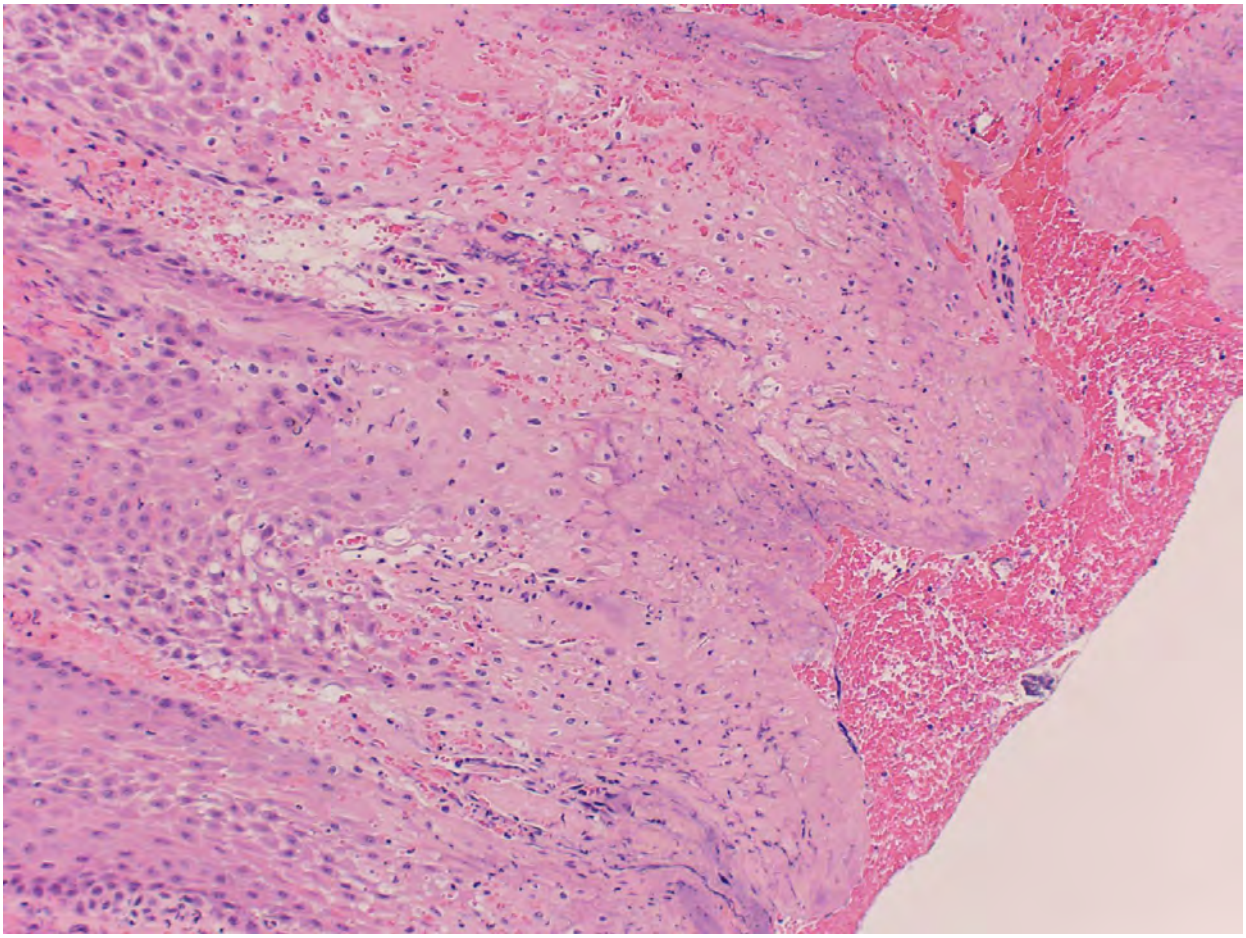
DD lesions are produced like many other disease complexes. There is some combination of how the organisms survive in the feedyard environment, animal factors affecting immunity and lesion development, and the various interactions among the bacteria found in clinical cases. Intervention strategies need to be developed that go beyond the routine use of footbaths containing caustic chemicals. Routine footbaths require increased labor, ingredient costs, and moving the cattle out of their routine. This can result in a decrease in feed intake that may last for several days. Since there seems to be no consensus as to how often cattle should be run through the footbath or the ideal ingredient mix, it would appear that there is widespread dissatisfaction with this option.

One option that we intend to explore at ISU is the ability to change the pen environment using litter treatments utilized by the poultry industry. This might be applicable for use in "indoor" pens such as monoslopes or hoop structures. These products would be periodically spread on the bedding pack and the feed pad in front of the bunks. The intent is to dry the pen out, decrease the pH of the pack and dramatically reduce the number of bacteria present. This should result in decreased active lesions and a reduction in trips through the footbath. Obviously, the cost and labor associated with this practice would have to be weighed against footbath use.

The development of a vaccine for DD would seem to be difficult. This is a polybacterial complex with populations that shift over time. Previous attempts to infer protection using *Treponema spp.* have not been successful. *Treponema* does not penetrate intact healthy skin and is more abundant in the more

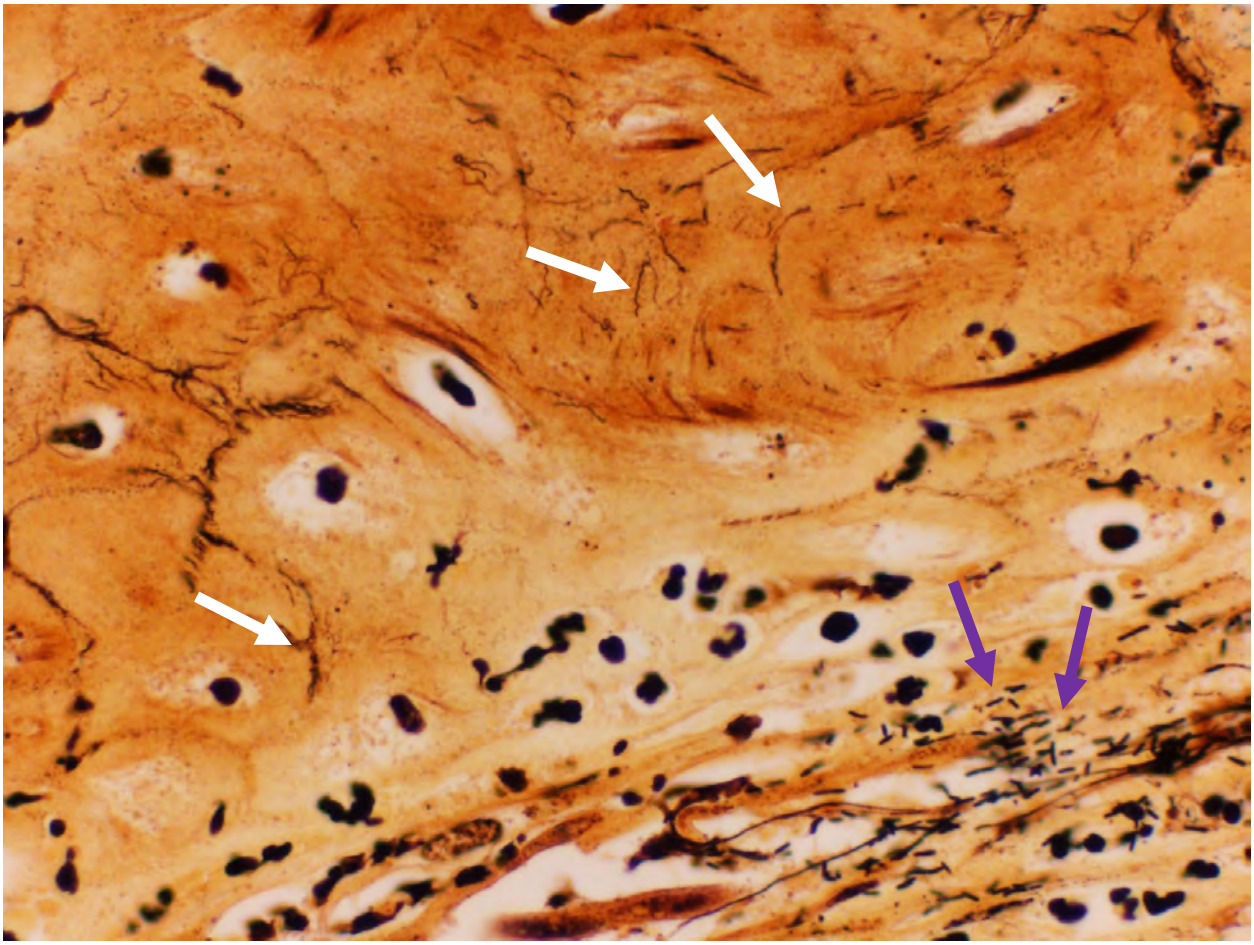
chronic lesions. It would seem logical to target organisms found in the early lesions since they are there when the problem is initiated. As with other vaccines that target bacterial diseases, it is almost certain that a minimum of two doses would be required. Finally, the economics of any potential vaccine would have to weighed against the current use of footbaths.

Figure 1. Histopathology slide (magnified 1000X) using H&E staining of a digital dermatitis lesion ("hairy heel wart").



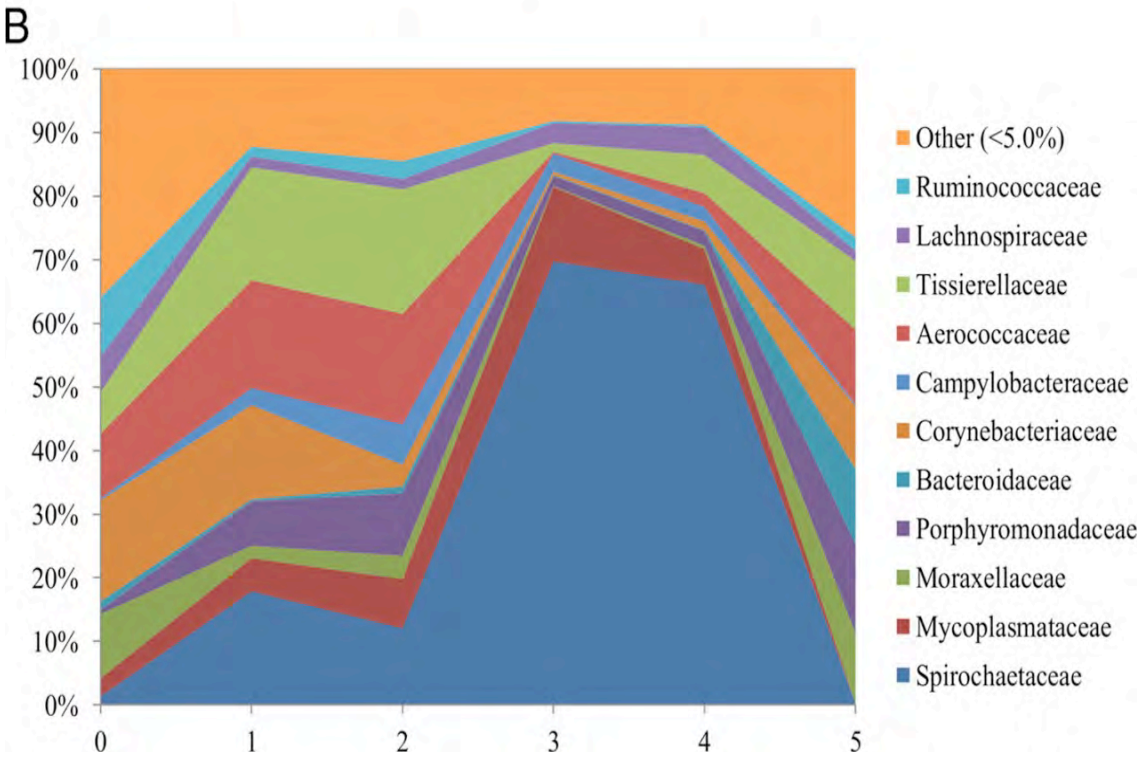
Histopathology report: Sections examined consist primarily of frond-shaped layers of keratin, with ballooning degeneration of epithelial cells and clusters of pyogranulomatous inflammatory cell populations. Surfaces are densely colonized by mixed bacterial flora including cocci, coccobacilli, bacilli, and delicate spirochetes. Localized areas of ballooning degeneration exhibit large numbers of delicate spirochetes in deeper cell layers. Lesions are consistent with infectious pustular pododermatitis ("hairy heel warts").

Figure 2. Histopathology slide (magnified 1000X) using Warthin-Starry staining of a digital dermatitis lesion ("hairy heel wart"). This staining is used to identify the presence of Spirochete bacteria.

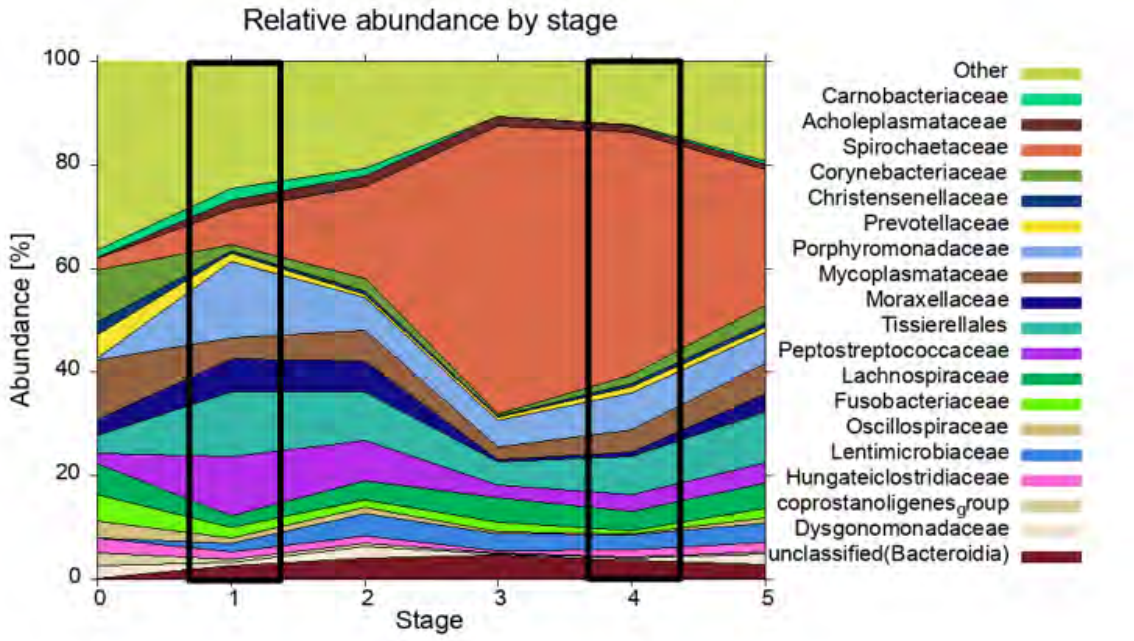


Multiple long dark colonies of Spirochetes are present in the slide (white arrows) in the deeper tissues. There are also multiple rod-shaped bacteria of a different type at the bottom of the slide (purple arrow).

Figure 3. Relative abundance, by stage, of bacterial families that represent at least 5% of the bacterial reads acquired. (Top = Dairy Cattle; Bottom = Feedlot Cattle)



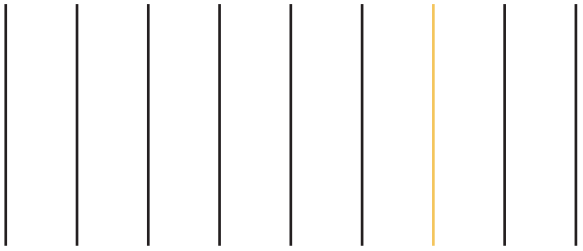
Krull AC et al. *Infection and Immunity* 82(8):3359-3373 (2104)



Engelken 2022 (*unpublished data*)

MANAGING HEALTH IN CONFINED COW/CALF OPERATIONS

TERRY ENGELKEN
DVM, MS



LARGE ANIMAL

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A range of different products are effective including copper sulfate (5%), zinc sulfate (5–10%), formalin (2–5%), and commercial chemicals containing quaternary ammonium compounds, organic acids, and other disinfectants. Recently, several new products which serve to activate copper sulfate have been released which allow lower concentrations (2%) to be used. It is essential that the volume of the foot bath is known so that the correct amount of chemical may be used to provide the appropriate final concentration. The volume in gallons may be calculated from the formula; length × width × depth (in inches) divided by 231. The University of Wisconsin has a "footbath calculator" available online that will match footbath dimensions with the proper amount of needed additives. Foot baths should be at least 8 feet long and 5 inches deep to ensure that enough contact is made between the chemical and the lesions. There are various options and locations that will work in a feedyard setting. The key is to put these in high traffic areas where the calves must walk through them with enough access to make recharging the bath easier. Minimizing the amount of manure on the feet will decrease the organic material tracked into the footbath and make the solution last longer.

Footbath frequency and solution selected will vary depending upon the cattle handling facilities, safety of the people working the cattle, and the percent of the pen that is affected.

Prevention still centers around the pen environment and avoiding negative impacts on the heel area of the calves. We have found that decreasing the moisture in the pen by more aggressive cleaning and decreasing animal density can be helpful. Scraping outdoor lots to remove manure and smooth out frozen hoof prints should improve foot health. Close observation of the feet of newly arrived cattle and recording their source can potentially identify problem sets of calves at arrival. Running cattle through a footbath at arrival should also be considered if active lesions are suspected in new cattle.

Future Needs and Direction

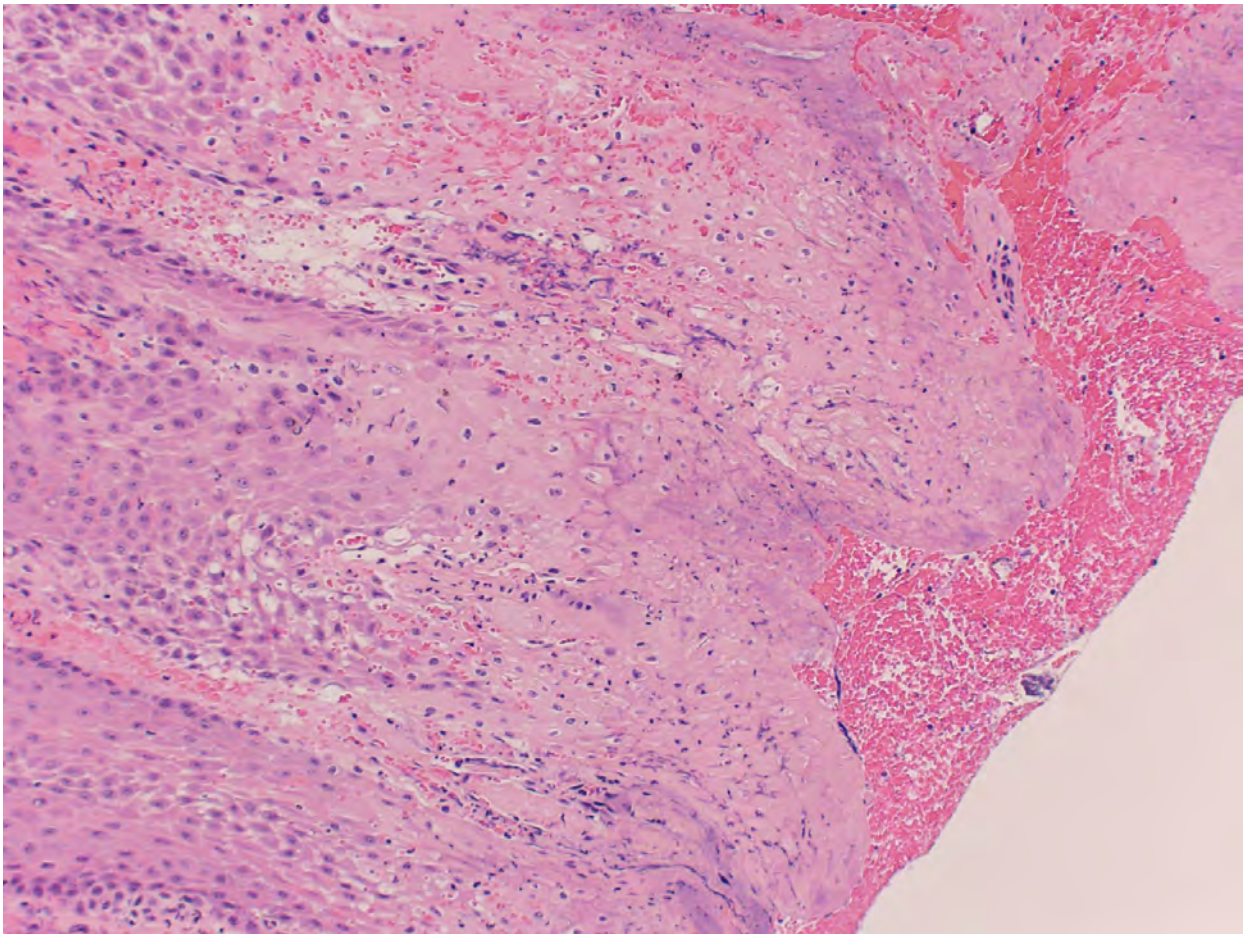
DD lesions are produced like many other disease complexes. There is some combination of how the organisms survive in the feedyard environment, animal factors affecting immunity and lesion development, and the various interactions among the bacteria found in clinical cases. Intervention strategies need to be developed that go beyond the routine use of footbaths containing caustic chemicals. Routine footbaths require increased labor, ingredient costs, and moving the cattle out of their routine. This can result in a decrease in feed intake that may last for several days. Since there seems to be no consensus as to how often cattle should be run through the footbath or the ideal ingredient mix, it would appear that there is widespread dissatisfaction with this option.

One option that we intend to explore at ISU is the ability to change the pen environment using litter treatments utilized by the poultry industry. This might be applicable for use in "indoor" pens such as monoslopes or hoop structures. These products would be periodically spread on the bedding pack and the feed pad in front of the bunks. The intent is to dry the pen out, decrease the pH of the pack and dramatically reduce the number of bacteria present. This should result in decreased active lesions and a reduction in trips through the footbath. Obviously, the cost and labor associated with this practice would have to be weighed against footbath use.

The development of a vaccine for DD would seem to be difficult. This is a polybacterial complex with populations that shift over time. Previous attempts to infer protection using *Treponema spp.* have not been successful. *Treponema* does not penetrate intact healthy skin and is more abundant in the more

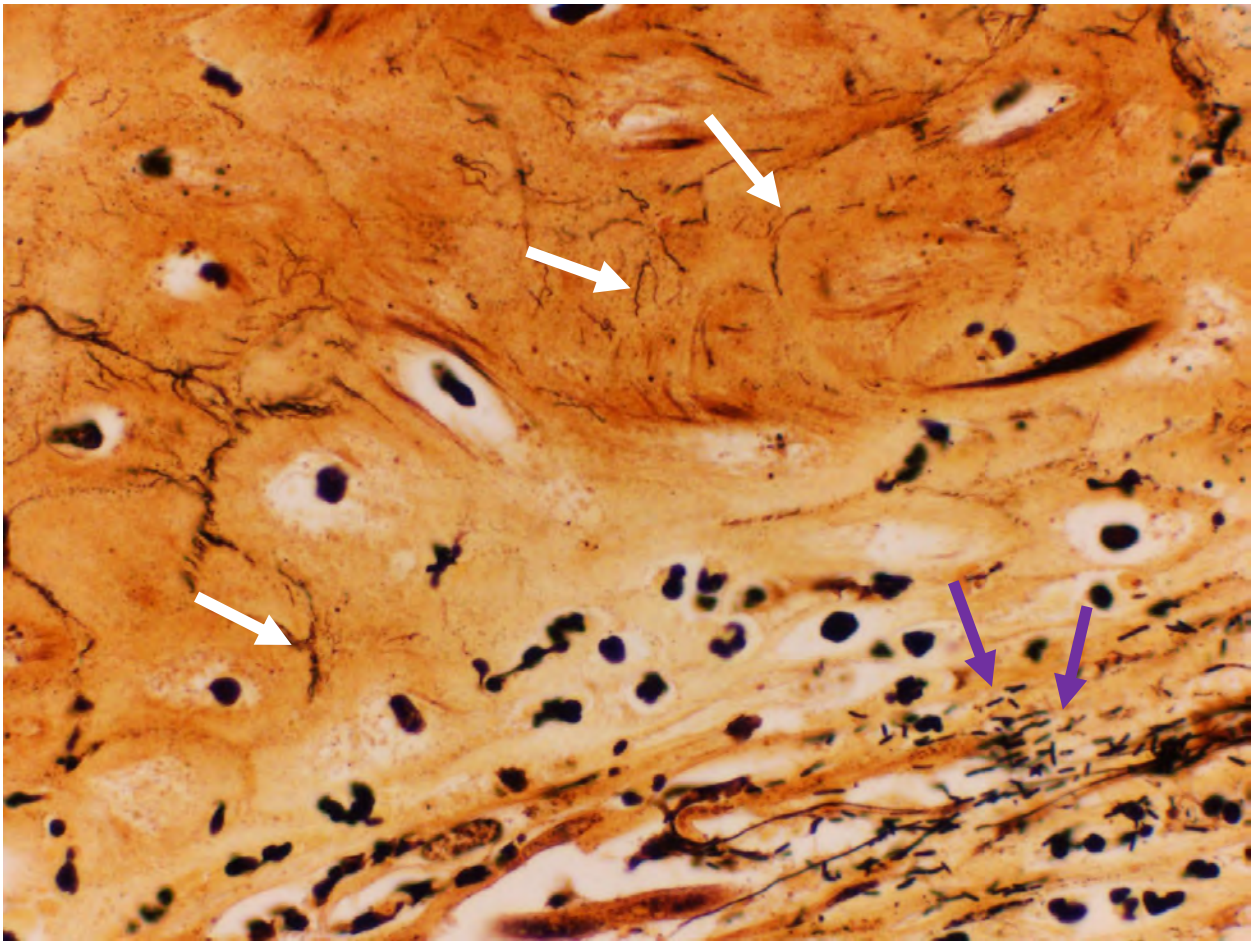
chronic lesions. It would seem logical to target organisms found in the early lesions since they are there when the problem is initiated. As with other vaccines that target bacterial diseases, it is almost certain that a minimum of two doses would be required. Finally, the economics of any potential vaccine would have to weighed against the current use of footbaths.

Figure 1. Histopathology slide (magnified 1000X) using H&E staining of a digital dermatitis lesion ("hairy heel wart").



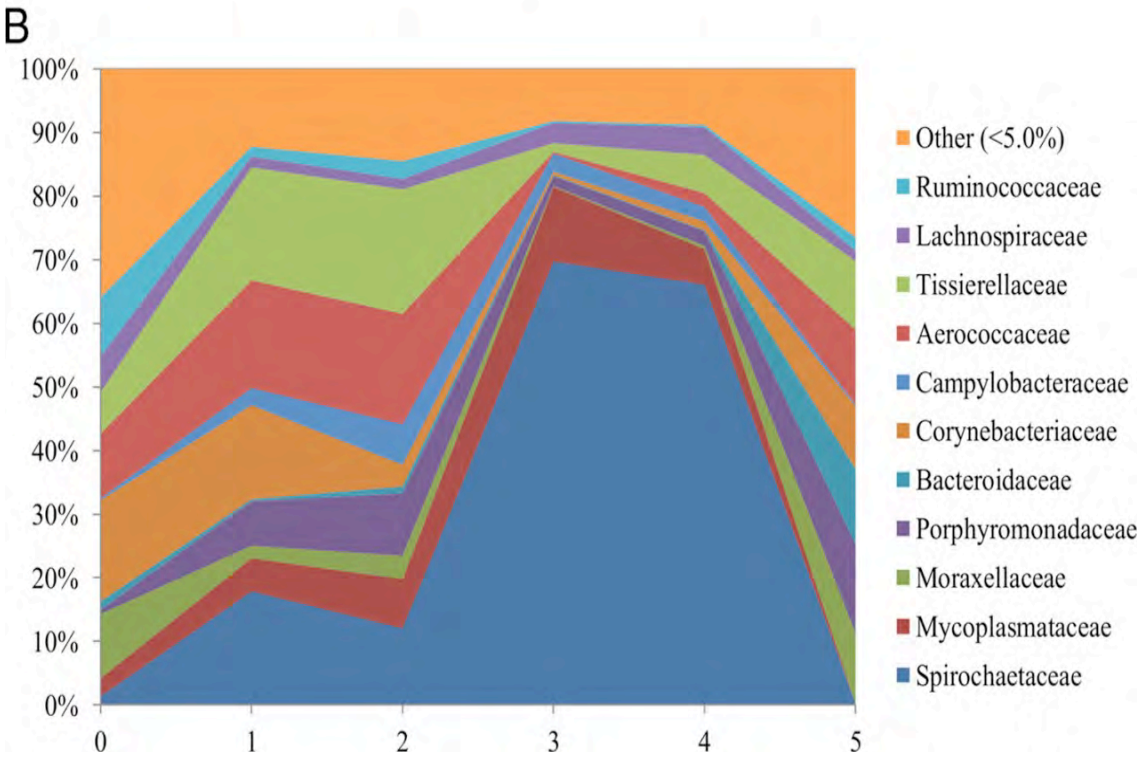
Histopathology report: Sections examined consist primarily of frond-shaped layers of keratin, with ballooning degeneration of epithelial cells and clusters of pyogranulomatous inflammatory cell populations. Surfaces are densely colonized by mixed bacterial flora including cocci, coccobacilli, bacilli, and delicate spirochetes. Localized areas of ballooning degeneration exhibit large numbers of delicate spirochetes in deeper cell layers. Lesions are consistent with infectious pustular pododermatitis ("hairy heel warts").

Figure 2. Histopathology slide (magnified 1000X) using Warthin-Starry staining of a digital dermatitis lesion ("hairy heel wart"). This staining is used to identify the presence of Spirochete bacteria.

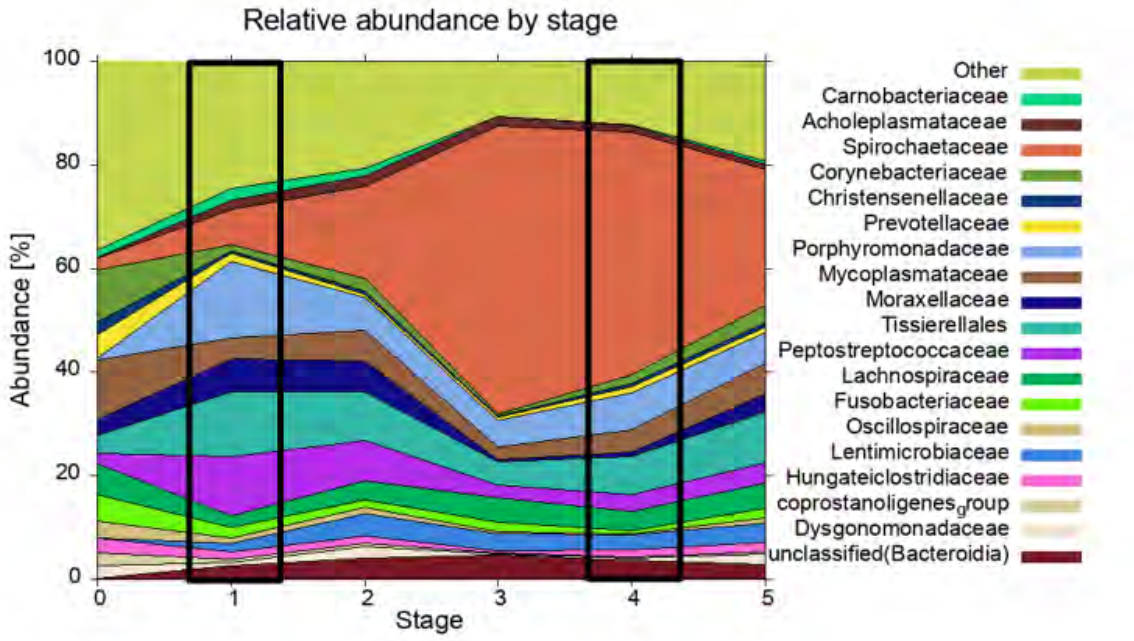


Multiple long dark colonies of Spirochetes are present in the slide (white arrows) in the deeper tissues. There are also multiple rod-shaped bacteria of a different type at the bottom of the slide (purple arrow).

Figure 3. Relative abundance, by stage, of bacterial families that represent at least 5% of the bacterial reads acquired. (Top = Dairy Cattle; Bottom = Feedlot Cattle)



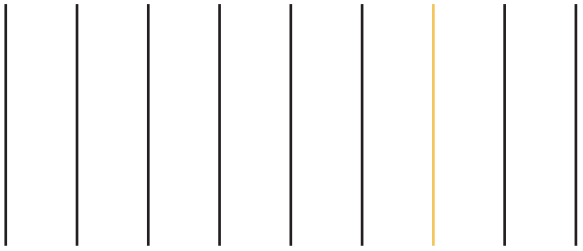
Krull AC et al. *Infection and Immunity* 82(8):3359-3373 (2104)



Engelken 2022 (*unpublished data*)

INFECTIOUS BOVINE KERATOCONJUNCTIVITIS – WHAT WE KNOW & DON'T KNOW ABOUT "PINKEYE"

TERRY ENGELKEN
DVM, MS



LARGE ANIMAL

Infectious Bovine Keratoconjunctivitis – What we Know and Don't Know about "Pinkeye"

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Iowa State University
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Infectious Bovine Keratoconjunctivitis (IBK) or "pinkeye" is the most common ocular disease of cattle, worldwide. Colorado researchers have identified IBK as the 3rd most important health concern among producers, falling behind only respiratory disease and flies. Clinical signs include excess lacrimation, photophobia and blepharospasm. This is followed by corneal edema and if the infection continues, ulceration. It is common for these corneal ulcers to heal with evidence of scarring in the center of the eye. Eye irritation from face flies, plant awns, or extreme UV light may damage the cornea and make it more susceptible to bacterial invasion. Economic losses associated with this disease include treatment costs, labor, decreased weaning weight, and discounts associated with blind calves. According to the 2017 NAHMS Cow-Calf Survey, nearly 20% of all cattle are vaccinated against IBK on an annual basis.

The exact etiology of IBK is currently being debated. There are multiple studies that have identified different potential bacterial pathogens and their associated clinical presentation. *Moraxella bovis* infection alone has been shown to cause the disease both experimentally and in field outbreaks. There are various other organisms isolated from both active lesions and normal eyes such as *Mycoplasma bovoculi, Moraxella bovoculi, Mycoplasma bovis,* and *Branhamella spp.* An Iowa State study using *M. bovoculi* inoculation alone on scarified corneas could not produce lesions of IBK. Researchers suspect that there is synergism between various organisms to produce IBK lesions, but the exact mechanism remains elusive. Multiple studies have shown the importance of maintaining the normal microbiome of the eye. Disruptions in the normal bacterial population of one eye may cause similar changes in the opposite eye as well and lead to bilateral disease.

Ocular immunity involves tear film, the mucosal epithelium, and diffuse lymphoid tissue. Tear film provides a physical barrier and contains secretory IgA. The epithelium produces antimicrobial proteins and dendritic cells while the lymphoid tissue around the eye secretes IgA and IgG. On the other hand, invading bacteria need two components to produce disease: Cytotoxin (RTX) and a pilus. The cytotoxin causes lysis of the corneal epithelium and migrating neutrophils and lymphocytes. This is the primary driver of corneal ulceration. The antigens of cytotoxin are highly conserved across different bacterial strains and animals will develop antibodies following cases of IBK. The invading bacteria need pili in order to adhere to the corneal surface and produce disease. There are seven distinct serogroups (A-G) and unfortunately, heterologous protection across the different subgroups does not occur. It has also been shown that adding too many of these pilus antigens to a single vaccine has a "dilution" effect that will prevent adequate immune response to any of the serogroups. This obviously complicates serogroup selection for vaccine production.

Studies have looked at the ability of serum antibody to protect against cases of IBK. An ELISA was developed to measure IgG specific antibody to the type IV pilus protein of *M. bovis*. These studies compared the relative antibody levels between calves that developed clinical IBK versus those that did not. While calves resistant to IBK had numerically higher ELISA titers, the differences were not significant. ISU data showed that antibody levels increase over the course of the summer (May through October), but there was no significant difference in titers between calves with clinical IBK and normal calves at any time point. Nebraska researchers showed that ELISA titers may increase following

vaccination with an autogenous or commercial product compared to sham vaccinated controls. However, there was no significant difference in disease outcomes between these three groups.

Vaccination against the organisms implicated in IBK has been controversial at best and ineffective at worst. Multiple studies using either autogenous or commercially available vaccines have failed to produce significantly less disease in well-designed studies. Factors such as strain selection, causal organisms present, and insufficient time between vaccination and pathogen colonization of the eye will all impact apparent vaccine efficacy. These results should not be totally surprising as we struggle to define the basic pathophysiology of how microbiome insult, pathogen interaction, and ocular immune system stimulation interact to produce disease. A better understanding of how the immune system of the calf interacts with ocular pathogens will be required to enhance vaccine efficacy.

Suggested Reading:

Ruminant Ophthalmology. Veterinary Clinics of North America: Food Animal Practice 37(2):237-380, July 2021. O'Connor AM, editor.

Hille MM, Spangler ML, Clawson ML et al. A five year randomized controlled trial to assess the efficacy and antibody responses to a commercial and autogenous vaccine for the prevention of infectious bovine keratoconjunctivitis. *Vaccines* 2022, 10, 916 https://doi.org/10.3390/vaccines10060916.

Angelos JA, Agulto RL, Mandzyuk B et al. Randomized controlled field trial to assess the efficacy of an intranasal *Moraxella bovis* cytotoxin vaccine against naturally occurring infectious bovine keratoconjunctivitis. *Vaccine: X* 15 (2023) 1003778 https://doi.org/ 10.1016/j.jvacx.2023.100378.

Garcia MD, Matukumalli L, Wheeler TL et al. Markers on bovine chromosome 20 associated with carcass quality and composition traits and incidence of contracting infectious bovine keratoconjunctivitis. *Animal Biotechnology* 21:188-202 (2010) https://doi.org/10.1080/10495398.2010.495012.

BALANCING BEEF COW DIETS: INTRODUCTION TO BRANDS

ROBERT LARSON
DVM, PHD, DACT, DACVPM (EPIDEMIOLOGY), ACAN

LARGE ANIMAL

BRANDS
Beef Ration and Nutrition Decisions Software

Balancing Beef Cow Diets: Introduction to BRANDS

Dr. Bob Larson

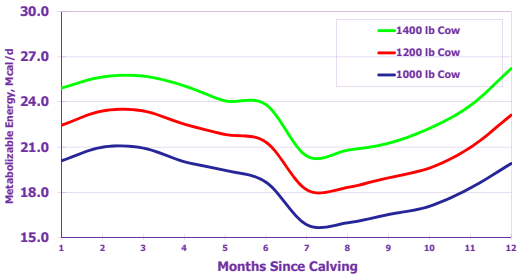
Step 1: Determine Diet Requirements

Energy and Protein Requirements

- **Requirements for Breeding Cattle are Dependent Upon:**
  - Body Size
  - Stage of Pregnancy
  - Body Condition and Compensation
  - Milking Ability
  - Breed Type

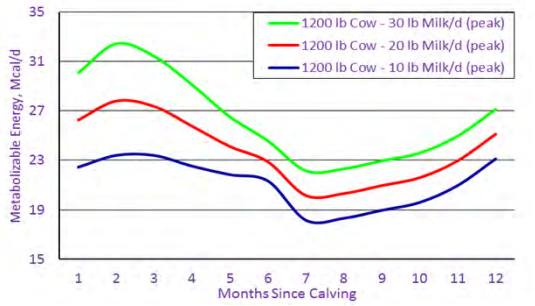
Animal Nutrient Requirements

Effect of Cow Size



Animal Nutrient Requirements

Effect of Milk Production Level and Pregnancy



Nutrient Requirements of Beef Cows

Dependent on Stage of Pregnancy

Nutrient	Period 1 80 days (post-calving)	Period 2 125 days (pregnant & lactating)	Period 3 110 days (mid-gestation)	Period 4 50 days (pre-calving)
TDN (lbs/day)	16.0	12.7	10.5	12.3
NEm Mcal/day	16.4	13.4	10.1	11.3
Protein (lbs/day)	2.5	2.1	1.5	1.8
Calcium (gm/day)	33	27	17	25
Phosphorus (gm/day)	25	22	17	20
Vitamin A (IU/day)	39,000	36,000	25,000	27,000

*NRC requirements plus 10%

Step 1: Determine Diet Requirements

Other Considerations:

- Temperature / Moisture
- Hair coat
- Acclimation
- Ionophores

Interpreting BCS of Beef Cows

Assumptions:

- BCS is a good estimate of fat stores, but not a good indication of current energy partitioning
- Low BCS at calving and the start of breeding is associated with poor reproductive efficiency
  - Low BCS at calving is associated with prolonged postpartum anestrus
  - Declining (or steady) BCS at breeding is associated with poor fertility
- Low BCS at calving of the dam is associated with increased neonatal calf health risk
- Extremely high BCS is an economic waste
- A range of BCS is compatible with optimum production based on timing within production cycle

BCS Association with Beef Cattle Reproduction

- Several publications document unfavorable measurements of reproductive success in thin cattle compared to cattle characterized as having moderate to good body condition
  - Longer interval to resumption of fertile cycles
  - Lower percentage pregnant
  - Lighter weaning weights of calves

Interpreting BCS of Beef Cows

My Current Thoughts:

- BCS is easily obtained (low technical, labor, and financial investment) and is appropriate for mature cow/bull evaluation
- BCS can be repeatable within and somewhat consistent between evaluators with training
- BCS at calving serves as a proxy for length of postpartum anestrus
- BCS change from calving to breeding serves as a proxy for onset of fertile cycles (PPA) and fertility

Interpreting BCS of Beef Cows

My Current Thoughts:

- BCS collected prior to calving, prior to breeding, and at the time of pregnancy diagnosis (mid-gestation) provide different information – but all three data collection times are important
- Good BCS prior to calving and breeding is indicative of good management from several perspectives (forage management, supplementation strategy, matching cows to the environment, access to feed, health, etc.)
- Manage cows and heifers as groups (populations) so that a herd-specific minimum percentage have BCS ≥ 5
- Individual cow BCS should be interpreted in relation to the group's BCS distribution

Relationship Between Body Condition and Reproductive Efficiency

Calving BCS	Number of Days to Resume Fertile Cycles
3	88.5
4	69.7
5	50.5
6	51.7

Adapted from Houghton 1990

Relationship Between Body Condition and Reproductive Efficiency

- Thin cows require special management during late pregnancy and early postpartum periods to maintain reproductive performance (Houghton et al., 1990)

BCS at Time of Examination	BCS Needed at Calving	Total Wt. Gain Needed	Days to Onset of Calving	Daily Weight Gain Needed (lbs./d)
Very Thin (3)	5	194 lbs.	60 (2 mo)	3.2
Thin (4)	5	97 lbs.	60 (2 mo)	1.6
Moderate (5-6)	5	0 lbs.	60 (2 mo)	0
Very Thin (3)	5	194 lbs.	90 (3 mo)	2.1
Thin (4)	5	97 lbs.	90 (3 mo)	1.0
Moderate (5-6)	5	0 lbs.	90 (3 mo)	0
Very Thin (3)	5	194 lbs.	120 (4 mo)	1.6
Thin (4)	5	97 lbs.	120 (4 mo)	0.8
Moderate (5-6)	5	0 lbs.	120 (4 mo)	0

Step 1: Determine Diet Requirements
Step 2: Estimate Forage Intake

Forage Quality	Percent Body Weight Intake
Excellent	3-3.8%
Average	2-2.5%
Crop residues	1.8-2.0%
Below average	1.8-2.0%
Extremely poor quality	1.4-1.8%

2%$^{+}$/– 0.5 (1.5-2.5%)

Step 1: Determine Diet Requirements
Step 2: Estimate Forage Intake
Step 3: Determine Forage Contribution

Requirement – Forage Contribution = Supplement Needed

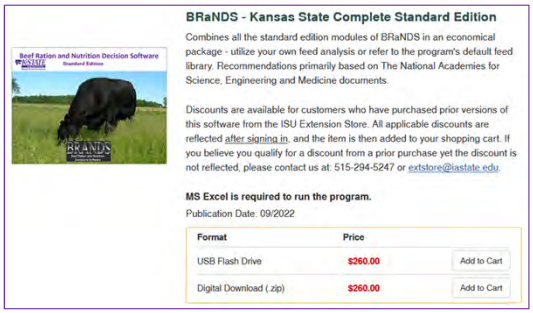
How energy-dense does the supplement need to be?

- Corn → CGF/DDG/SH/WM → high quality forage

https://www.iowabeefcenter.org/brands.html

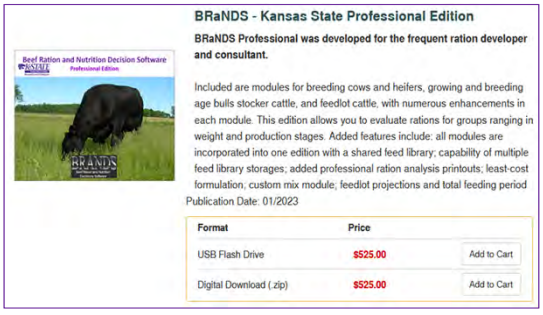
Kansas Standard Cow & Heifer Edition

https://store.extension.iastate.edu/product/14899



Kansas State Professional Edition

https://store.extension.iastate.edu/product/12888



Using BRANDS

Producer Setup

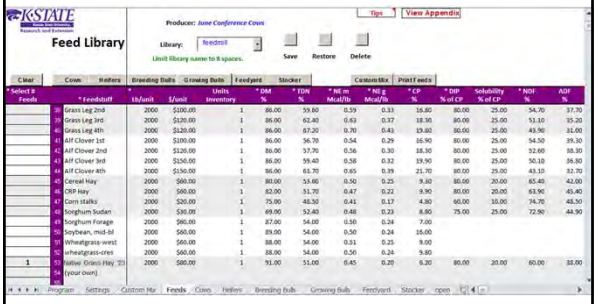
- Open Brands Software
- Enable MACROS
- Go to "Settings" Tab
- Click on New Producer, Enter producer Info, Click Save Button

Using BRANDS

Feeds Selection and Customization

- Go to Feeds Library - click on Feeds tab
- Enter Native Grass Hay (2023) manually

Dry Matter, %	91%
TDN	51%
NEm, mcal/lb.	0.45
NEg, mcal/lb.	0.20
Crude Protein, %	6.2%
DIP, % of CP	80%
Sol. Of CP, % of CP	20%
NDF, %	60%
ADF, %	38%
eNDF, % of NDF	90%
Calcium, %	0.81
Phosphorus, %	0.18



Using BRANDS

Feeds Selection and Customization

- Go to Feeds Library - click on Feeds tab
- Enter Native Grass Hay (2023) manually
- Select several potential feeds
  - 1 – Line 53 (manually added hay '23)
  - 2 – Line 94 (Soy hulls - $105/ton)
  - 3 – Line 95 (Wheat mids - $85/ton)
  - 4 – Line 100 (Corn Gluten Feed - $110/ton)
  - 5 – Line 104 (Dried Distillers Grains - $165/ton)
  - 6 – Line 138 (Corn - $3.63/bu…*$130/ton*)

Where To Find Feed Prices?

By-product feeds
http://agebb.missouri.edu/dairy/byprod/bplist.asp

Corn
https://www.ams.usda.gov/market-news/state-grain-reports

Hay
https://www.ams.usda.gov/market-news/hay-reports

Using BRANDS

Nutrient Requirements

- Go to Cow Module – click on Cow tab
- Enter Herd Parameters
  - Feeding Period – 1
  - 1/19/23 thru 3/25/23
  - Cow Size – Medium
  - Breed type - British
  - High - Milk
  - BCS - 5
  - BCS Desired - Maintenance

Using BRANDS

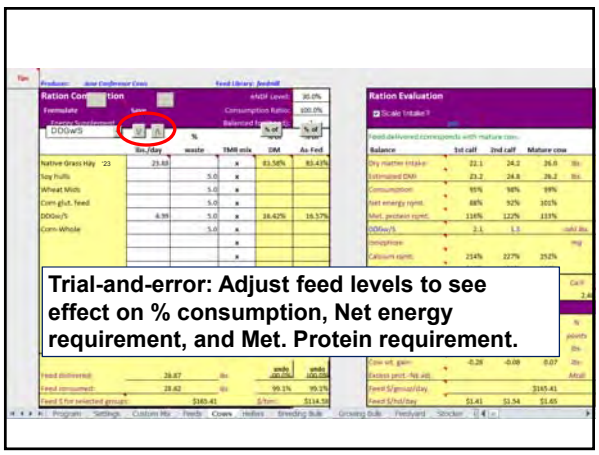
Nutrient Requirements

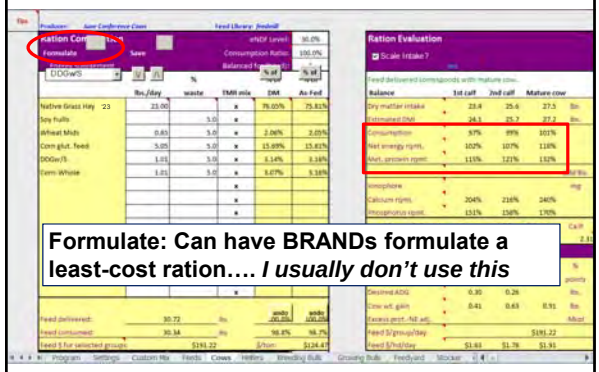
- Go to Cow Module – click on Cow tab
- Enter Herd Parameters
  - Prod. Stage - 3rd trimester
  - Calf Birth Weight - Moderate
  - Wind - Normal
  - Hair Condition – Clean & Dry
  - Hair coat - Winter
  - Temperature - Normal
  - Enter Data, Name File, AND PRESS SAVE

Using BRANDS

Ration Balancing Screen

- Determine amount of base forage to be consumed (usually 1.5 to 2.5% of BW)
- Determine amount of DDG needed to meet requirements
- Trial-and-Error method of ration balancing
- Use of ↓ ↑ buttons (next to Energy Supplement window)





Using BRANDS

Nutrient Requirements

- See what happens if you change BCS to "4" and BCS Desired to "+1/2 CS/mo" …

Ration Evaluation

☑ Scale Intake?

yes

Feed delivered corresponds with mature cow.

Balance	1st calf	2nd calf	Mature cow	
Dry matter intake	22.1	24.2	26.0	*lbs.*
Estimated DMI	23.2	24.8	26.2	*lbs.*
Consumption	95%	98%	99%	
Net energy rqmt.	69%	73%	80%	
Met. protein rqmt.	82%	87%	95%	
DDGw/S	6.2	5.6	4.1	*add lbs.*

Using BRANDS

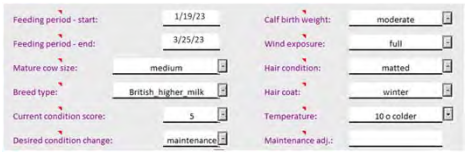
Nutrient Requirements

- Changes needed – increase DDG…

Using BRANDS

Nutrient Requirements

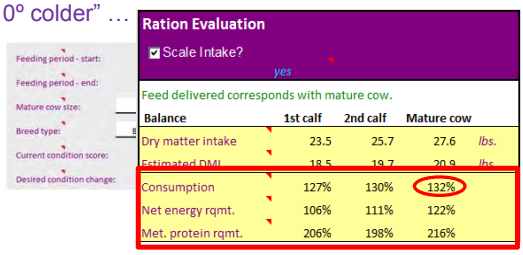
- See what happens if you change Wind exposure to "full", Hair condition to "Matted", and Temperature to "10º colder" …



Using BRANDS

Nutrient Requirements

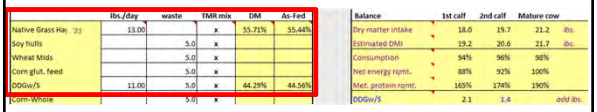
- See what happens if you change Wind exposure to "full", Hair condition to "Matted", and Temperature to "10º colder" …



Using BRANDS

Nutrient Requirements

- Changes needed – increase DDG…



Using BRANDS

Nutrient Requirements

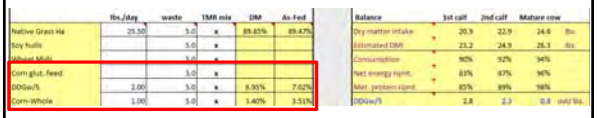
- See what happens if you started with a higher quality forage (good brome hay) …

Forage intake increases and need for supplement greatly decreases !

Using BRANDS

Nutrient Requirements

- Start by using corn and DDG

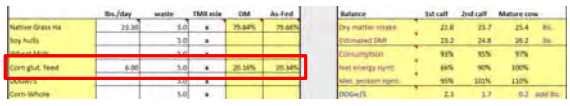


$0.93 / cow / day

Using BRANDS

Nutrient Requirements

- Try using Corn Gluten Feed

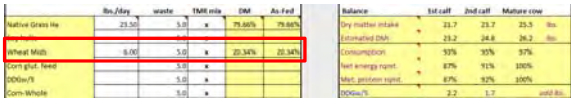


$0.98 / cow / day

Using BRANDS

Nutrient Requirements

- Try using Wheat Mids



$0.90/ cow / day

Using BRANDS

Nutrient Requirements

- The point is not that currently wheat mids are less expensive than distillers grains
- The point is that BRANDS can be used to compare options

Summary:

- Mature cows in moderate BCS (i.e. 5) that are not presented with weather stress and have average or better quality forage available will maintain or gain body weight on forage alone.
- All other situations (i.e. thin cows, cows consuming less than average quality forage, cows facing weather stress, heifers) require 3 to 15 lbs. of an energy-dense supplement (e.g. distillers grain, soy hulls, corn gluten feed, corn, etc.) to meet their caloric needs.

Heifers are not cows!!

Heifers' nutritional requirements from weaning to breeding are very different from mature cows' primarily because heifers are still growing (require NE partitioned toward growth in addition to maintenance)

Interpreting Target Weight for Puberty

What is the appropriate target weight?

- 50% - 55% - 60% - 65% of mature weight?
- Real question is…

  "What ration should I feed cohort of replacement heifers to result in the desired number reaching puberty and becoming pregnant at the desired date?"

  Need to know target weight in order to determine desired average daily gain from weaning to breeding

  ADG = (Target weight – Starting weight) / Number of days

  I would rather know yearling wt. (not % of mature wt) that meets the herd's goals

Interpreting Target Weight for Puberty

What is the appropriate target weight?

- How is target weight calculated?
- What is your goal?
  - Nearly all heifers in replacement pool reach puberty?
    - Set a high target weight (actual lbs. or 65% of mature wt.)
  - Only small-framed heifers (low mature wt.) or early maturing heifers reach puberty?
    - Use herd average mature weight – and set a low target weight (actual lbs. or 55% of mature wt.)

Interpreting Target Weight for Puberty

What is the appropriate target weight?

- How is target weight calculated?
- What is your goal?
- Answer: Monitor herd – what weight is needed to reach targeted number of pubertal heifers?
  - If I know – that is the target weight (assuming constant genetic potential for mature wt. and age-at-puberty)
  - If I don't know – base target weight on producer's goal

Summary (moderate wt. gain):

Situation (animal requirement / forage quality)	Outcome (Supplement Required)
BCS 5 cows with average quality forage	Maintain (or even gain) weight on forage alone
BCS 5 cows with poor quality forage	Lose weight on forage alone – need 3 lbs (as fed) DDG to maintain body wt.
BCS 3-4 cows (need to gain 1.7 lbs. daily) with average quality forage	Need ≈7 lbs (as fed) DDG to obtain BCS 5 w/n 90 days
BCS 3-4 cows (need to gain 1.7 lbs. daily) with poor quality forage	Need ≈11 lbs (as fed) DDG to obtain BCS 5 w/n 90 days
Heifers that need to gain 1.5 lbs. daily with good quality forage	Need ≈4.5 lbs (as fed) DDG to meet targeted gain
Heifers that need to gain 1.5 lbs. daily with average quality forage	Need ≈5.5-6.0 lbs (as fed) DDG to meet targeted gain
Heifers that need to gain 1.5 lbs. daily with poor quality forage	Need ≈7.5-8.0 lbs (as fed) DDG to meet targeted gain

Summary (rapid wt. gain):

Situation (animal requirement / forage quality)	Outcome (Supplement Required)
BCS 3-4 cows (need to gain 2.5 lbs. daily) with average quality forage	Need ≈12-12.5 lbs (as fed) DDG to obtain BCS 5 w/n 60 days
BCS 3-4 cows (need to gain 2.5 lbs. daily) with poor quality forage	Need ≈15-15.5 lbs (as fed) DDG to obtain BCS 5 w/n 60 days
Heifers that need to gain 2.5 lbs. daily with good quality forage	Need ≈11.0-11.5lbs (as fed) DDG to meet targeted gain (not much forage)
Heifers that need to gain 2.5 lbs. daily with average quality forage	Need ≈11.5-12.0 lbs (as fed) DDG to meet targeted gain (not much forage)
Heifers that need to gain 2.5 lbs. daily with poor quality forage	Need ≈12.5-13.0 lbs (as fed) DDG to meet targeted gain (not much forage)