EVALUATION OF IMMUNE RESPONSE AND PERFORMANCE IN STEERS OF

KNOWN GENETIC BACKGROUND VACCINATED AND CHALLENGED WITH

BOVINE VIRAL DIARRHEA VIRUS

A Thesis

by

CHASE ANTHONY RUNYAN

Submitted to the Office of Graduate Studies of
Texas A&M University
in partial fulfillment of the requirements for the degree of

MASTER OF SCIENCE

December 2010

Major Subject: Animal Science

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Approved by:

Chair of Committee,	Andy D. Herring
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ABSTRACT

Evaluation of Immune Response and Performance in Steers of Known Genetic

Background Vaccinated and Challenged with Bovine Viral Diarrhea Virus.

(December 2010)

Chase Anthony Runyan, B.S., Oklahoma State University

Chair of Advisory Committee: Dr. Andy D. Herring

This research was directed at investigating the variation in immune response of cattle when administered a known challenge from Bovine Viral Diarrhea Virus (BVDV) following different Bovine Respiratory Disease (BRD) vaccine treatments. Cattle were assigned vaccine treatments with sire and cow family was stratified across treatments to assess the role genetic differences may impact immune function. The same BVDV strain and challenge technique were used in two trials (2008 and 2009) in Angus-sired yearling steers. Data from these two years were analyzed separately because the cattle were managed and fed differently.

Blood antibody Immunoglobulin-G (IgG) titers for IBR, BVD Type 1 and BVD Type 2 were higher for cattle in the Killed vaccine group than the MLV or NON vaccinated groups ($P < 0.05$) in both years. In the 2008 study, average daily gain (ADG) was higher for cattle from the Killed vaccine group ($P < 0.05$) for the 28 d following BVDV challenge, but no cattle were classified as morbid based on rectal temperature. In the 2009 study, differences in rectal temperatures were observed, and a total of 35 of 93

having over 40.0^{o}C (28 in the first 14 d following challenge). Cattle in the MLV vaccine group had lower overall mean temperatures, with no animals having rectal temperatures over 40°C 14 d following viral challenges. Differences in rectal temperature were also observed due to sire. Differences in feed intake also occurred due to treatment, day, treatment × day interaction, and maternal-grandsire. The MLV vaccine group maintained more constant levels of intake as compared to Killed and NON vaccinated cattle at days 5 to 12. Although large differences in titers following BVDV challenge were observed, the relationships of this immune response with animal health and performance appears very complex.

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CHAPTER I

INTRODUCTION

Bovine Respiratory Disease (BRD) is costly to U.S. beef production due to effects on weight gain, production efficiency, antibiotic therapy, and in some cases animal death. In spite of improved efforts for prevention and treatment, there is also evidence that morbidity manifests as sub-clinical illness and can result in irritations of the lungs and, in severe cases, pulmonary tissue damage in animals never identified for illness. Breed differences pertaining to BRD occurrences have been reported, but the genetic influences of BRD still remain largely unknown.   There are also questions that remain about immune responses and protection offered by killed vs. modified-live BRD vaccines. Bovine Viral Diarrhea (BVD) is a disease that is a component of BRD and is a threat to cattle production systems not only from its threat of morbidity and mortality from transient infection, but also the incidence of cattle persistently infected with the virus. As a result, the objectives of this project were to: (1) evaluate genetic variation and vaccination effects on immune response and health measures in steers of known background that were challenged with Bovine Viral Diarrhea Virus (BVDV) following killed, modified-live or no BRD vaccination, (2) evaluate genetic and vaccination effects on weight gain and other traditional performance measures following vaccination and BVDV challenge, and (3) investigate potential genetic-vaccine treatment interactions for immune, health and performance measures.

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This thesis follows the style of the Journal of Animal Science.

CHAPTER II

LITERATURE REVIEW

Animal health can be summarized as the overall physical condition and well being of animals. In beef cattle, morbidity (sickness) accounts for reductions in performance and creates additional costs associated with treatment mortality (death loss), which can result in huge economic losses. Morbidity and mortality in growing cattle on pasture and in feedlots can be the result of numerous disorders, most commonly respiratory and digestive associated diseases (Smith, 1998).

It is well understood that Bovine Respiratory Disease (BRD) is an enormous burden in beef cattle operations. In terms of financial strain on the U.S. cattle industry, Griffin (1997) summarized an estimated annual loss of $750 million due to BRD incidence. Chirase and Greene (2001) reported a similar range of estimated loss of $800 to $900 million annually from death loss occurrence, reduced feed intake resulting in reduced gain performance, and treatment associated costs. McNeill et al. (1996) summarized Texas A&M Ranch to Rail data and concluded that a difference of $93 added expense per animal can be attributed to cattle treated for BRD versus cattle that have not been treated.

These financial strains reflect the concern over the incidence of morbidity due to BRD. Galyean et al. (1999) calculated that 52% of U.S. feedlot morbidity was a result of BRD. Snowder et al. (2006) reported incidence of BRD as high as 43.8% in 18,112 cattle over a 15-year period from herds at the U.S. Meat Animal Research Center at Clay Center, NE. Edwards (1996) reported morbidity of up to 82% as the result of BRD and

summarized that morbidity and mortality from BRD was most common in the first 45 days after arrival at the feedlot. Cattle are at a higher risk early in the feeding period when viral and bacterial exposure is compounded with high stress during arrival periods. Pre-exposed cattle from various geographic locations, breed types, immune status, and management systems are comingled in the feed yard shortly before arrival through regional auctions (Edwards, 2010). Efforts to reduce the amount of stress incurred during the receiving period is important especially for those cattle who are deemed as higher risk than others. Metaphylactic programs and prevention methods deserve attention when dealing with more susceptible, high risk cattle (Duff and Galyean, 2007).

Occurrences of BRD are the result of pathogenic bacteria and the presence of one or more viral infections (Daniels et al., 2000). The viral agents that contribute to BRD are *Bovine Herpes Virus-1* (BHV-1), *Parainfluenza-3 virus* (PI-3), *Bovine Respiratory Syncytial Virus* (BRSV) and *Bovine Viral Diarrhea Virus* (BVDV). These viruses create suppressed immune response activity which facilitates bacterial pneumonia caused by *Mannheimia haemolytica, Pasteurella multocida, Histophilus somni,* and *Mycoplasma spp.* (Fulton et al. 2000; Fulton et al. 2002; Fulton et al. 2006).

Incidence and morbidity of BRD has been recorded between breeds, although it has not been heavily researched. Muggli-Cockett et al. (1992) reported Pinzqauer offspring had a higher incidence frequency for BRD postweaning at 24.6% as compared to the lowest incidence frequency of 11.8% for the Angus in herds from the Germ Plasm Utilization project where 10,142 cattle were evaluated from 1983 to 1988. Snowder et al. (2006) also evaluated MARC herd BRD data from 1987 to 2001, and observed

incidence frequency for BRD was highest for Pinzqauer, Braunvieh, Simmental, and Limousin at 35%, 34%, 33%, and 32%, respectively. The Angus breed was the lowest at 10.2%, and the overall mean was 12.8% in the 12 breeds represented. Snowder et al. (2005) also reported MARC III composite (¼ Angus, ¼ Hereford, ¼ Pinzguaer, ¼ Red Poll) as not only having a lower incidence frequency of BRD at 9.7%, but also one of the highest mortality rates at 17.2%. They concluded that some breeds or breed combinations are inherently more sensitive to BRD than others.

Signs of clinical illness from BRD often include fever, depression and reduced appetite. Buhman et al. (2000) recorded lower frequency and duration of feed intake behavior in newly arrived feedlot cattle that were identified as sick vs. non-sick. Sick steers were identified using a scoring system that included a rectal temperature of greater the 40.0°C, visual signs of changes in attitude, cough, nasal secretion, ocular secretion, and hematologic examinations. A reduction in weight gain was reported by Gardner et al. (1999) as they observed 1.47 kg/d for treated cattle and 1.53kg/day for untreated cattle using visual symptoms and rectal temperature greater than 40°C. Roeber et al. (2001) also observed similar response in cattle treated more than once having .25 kg/d reduction in average daily gain (ADG) as compared to cattle not being treated based on visual diagnosis of morbidity. Schneider et al. (2009) reported cattle that were treated for BRD or had presence of lung lesions had a 0.07 kg/d reduction in ADG as compared to non treated cattle with no lung lesions.

There appears to be disconnecting evidence in the literature for ADG response and BRD incidence, however. Wittum et al. (1996) found no difference in ADG

between treated and untreated cattle, but did conclude that 68% of untreated steers displayed pulmonary lesions at slaughter. Further investigation by Wittum et al. (1996) in the same study concluded that steers exhibiting lung lesions, regardless of treatment status, did have a 0.076 kg/d reduction in ADG as compared to cattle exhibiting no signs of lung lesions upon harvest.

Traditionally, illness in cattle has been identified by changes in behavior that differ from healthy cattle. Trained animal handling personnel, experienced in pointing out these changes, use signs of depression, increased rectal temperature, lethargy, nasal and ocular secretion, gauntness, or coughing as signs of clinical illness; followed by therapy or treatments that are applied accordingly. Even still, a gap exists between observations and evidence of infection. Wittum et al. (1996) observed that 78% of cattle treated for BRD had pulmonary lesions evident at slaughter, whereas 68% of untreated steers also displayed pulmonary lesions at slaughter as compared to cattle with no pulmonary lesions. Gardner et al. (1999) also observed the presence of lung lesion in 37% of non-treated cattle. Similar observations were made by Schneider et al. (2009) where 60.6% of cattle never treated had lung abrasions present, and 74% of cattle that were treated also displayed lung tissue damage. These observations reflect the ability of infection to arise unnoticed by accepted clinical symptoms, however these subclinical infections are also harmful in terms of additional health stress which results in lost performance.

The focus of this thesis is the impact of Bovine Viral Diarrhea Virus (BVDV) on immune response and performance, so it is discussed in more detail than the other viral

components and pathogens of BRD. The greatest threat of BVDV comes primarily from persistently infected (PI) cattle. PI calves result from vertical transmission of BVDV from the infected dam's bloodstream to her fetus during pregnancy (Larson et al., 2002). BVDV hinders beef cattle production on a substantial economic scale as the estimated cost of a single occurrence of PI in a beef herd has been estimated to range from $14.85 to $24.84 per cow annually (Larson et al., 2002). It is important to note that the occurrence of PI calves in-utero happens between day 45 and day 175 of pregnancy (Grooms, 2004) during the time when the calf immune system is developing. Persistently infected calves fail to create an immune response to the BVD virus because they do not distinguish the virus from itself, and serve as reservoirs of infection for other cattle (Chase et al., 2008). Because a failure to respond to the infection has occurred, PI cattle continuously shed BVD virus through horizontal transmission from one animal to the other through mucosal secretions (Larson et al., 2002).

Bovine Viral Diarrhea virus can be classified into 2 genotypes (BVD type 1 and BVD type 2). Beyond these distinctions, each type has several subtypes such as BVD1a, BVD1b, BVD1c, etc; BVD1b is the most prevailing subtype in U.S. feedlots, whereas BVD1d is the most common in Australia (Fulton et al., 2003b; Fulton et al., 2009). Beyond these genotype separations, there is also a difference in biotypes for BVD. The biotype is defined as cytopathic or non-cytopathic (Ahn et al., 2005; Peterhans et al., 2003). These biotypes are classified based on the presence or absence of visible cytopathic, degenerative or degrading, effects in infected BVDV cell cultures. Ahn et al. (2005) recorded differences in occurrence of biotypes for BVDV as 30% of samples

were defined as cytopathic (CP) and 70% were non-cytopathic (NCP). Fulton et al. (2000) reported similar figures in vaccinated and non vaccinated steers with 20.8% of collected samples were CP, and 79.2% of samples were NCP.

Clinical symptoms of BVDV as reviewed by Peterhans et al. (2003) and Fulton et al. (2003a) include those similar to other respiratory infections. Further, the virus can affect the digestive tract, fetal development, and the immune system in addition to causing mucosal disease. BVDV has a dual nature for infection; BVDV itself serves as an infectious agent, but it also serves as an immunosuppressor of health defenses (Baker, 1995). With defense mechanisms suppressed, secondary bacterial colonization can occur which commonly leads to subsidiary infections (Edwards et al., 1986).

In terms of handling the occurrences of BVD, an important aspect is accurate screening and diagnosis of Persistently Infected (PI) calves. Sandvik (2005) described in detail numerous methods of laboratory testing for BVD virus itself through virus isolations, or immune function response to BVD, or via virus neutralizing antibodies. Cornish et al. (2005) summarized important characteristics of laboratory testing for detecting PI calves to have large capacity, be economical, accurate, and timely in order to be suitable for practical application.

Immunity is the ability to resist disease or infection from foreign substance that threatens homeostasis and wellbeing, and the complexity of the immune system is very extent. Two major components of immune function exist. The innate system, which is a non-specific response mechanism, and the adaptive immune system, which is antigen specific and leads to immunological memory (Abbas and Lichtman, 2007). The

principal components of innate immunity are (1) physical and chemical barriers, such as epithelia and antimicrobial substances produced at epithelial surfaces; (2) phagocytic cells (neutrophils, macrophages) and natural killer (NK) cells; (3) blood proteins, including members of the complement system and other mediators of inflammation; and (4) cytokine proteins that regulate and coordinate many of the activities of the cells of innate immunity.

The mechanisms of innate immunity are specific for structures that are common to groups of related microbes and may not distinguish fine differences between foreign substances. Innate immunity provides the early lines of defense against microbes (Abbas and Lichtman, 2007). Conversely, the adaptive immune system has an extraordinary capacity to distinguish among different, even closely related microbes and molecules, and for this reason is also called specific immunity (Abbas and Lichtman, 2007).

There are two types of adaptive immune responses; humoral immunity and cell-mediated immunity. Cell-mediated is derived by different components of the immune system and function to eliminate different types of microbes. Humoral immunity is mediated by antibodies in the blood and mucosal secretions and produced by B lymphocytes (also known as B cells). Cell-mediated immunity, also called cellular immunity, is mediated by T lymphocytes (also known as T cells). Intracellular microbes, such as viruses and some bacteria, survive and proliferate inside phagocytes and other host cells, where they are inaccessible to circulating antibodies. Defense against such infections is a function of cell-mediated immunity, which promotes the

destruction of microbes residing in phagocytes or the killing of infected cells to eliminate reservoirs of infection (Abbas and Lichtman, 2007).

Protective immunity against a microbe may be induced by the host's response to the microbe or by the transfer of antibodies or lymphocytes specific for the microbe. The form of immunity that is induced by exposure to a foreign antigen is called active immunity because the immunized individual plays an active role in responding to the antigen (Abbas and Lichtman, 2007).

Immunity can also be conferred on an individual by transferring serum or lymphocytes from a specifically immunized individual. The recipient of such a transfer becomes immune to the particular antigen without ever having been exposed to or having responded to that antigen. Therefore, this form of immunity is called passive immunity (Abbas and Lichtman, 2007), and is the primary mechanism by which cows provide immunity to their calves through colostrums.

Vaccine products generally contain modified-live (MLV) or killed viruses and bacteria toxins known to cause diseases. These products are administered to initiate the body's immune system into creating antibodies and develop immunological memory (Faries, 1999). Adjuvants are also included in available vaccines to slow the release of the antigen into the system to extend the immune response. Historically, most killed vaccine adjuvents are comprised of aluminum hydroxide or oil and water combinations, (Roth and Hednerson, 2001). Vaccines can contain an assortment biological agents, inactive toxins known as toxoids, killed bacteria known as bacterins and combinations of adjuvents which elevate the level of effectiveness of the antigens (Faries, 1999).

Faries (1999) also summarized that vaccines are identified as either infectious or noninfectious vaccines. Infectious vaccines contain an organism that is modified or altered to reduce its virulence so that it will not cause disease, but will still be infectious enough to provide immunity. MLV vaccines are infectious vaccines that achieve a desired level of infection. Immunity of the animal prevents the establishment of disease and provides immunity. Noninfectious vaccines are unable to infect or replicate infectious agents. Generally, noninfectious vaccines are weaker in their ability to illicit an immune response, so a second dose, or booster, is required 2-4 weeks later. The initial dose of vaccine is a priming, sensitizing dose that provides little to no protection and the booster provides protection for 6 to 12 months (Faries, 1999).

There appears to be some variance among reported studies pertaining to the duration of protection that vaccines provide. Cortese et al. (1998) concluded that vaccination with a MLV BVD Type 1 triggered antibodies to numerous strains of both, BVD type 1 and BVD type 2 that were detectable 18 months post vaccination. Reports from Fulton et al. (1995) indicate a decline in BVD antibody titers by day 140 following vaccination.

Some vaccines, which target specific antigens, such as toxoid vaccines, are meant to completely prevent disease from infection, whereas vaccines which are formulated for more complex agents of disease that tend to have more numerous antigenic strains are less likely to achieve a complete prevention level of protection (Faries, 1999). Callan and Garry (2002) summarized respiratory vaccines as disease modifiers rather than absolute preventative agents.

Additional benefits of vaccinatation exist which not only protects vaccinated individuals, but also reduces the ability to shed infections to pen mates or newly arrived comingled calves (Frank et al., 2003). Vaccinations are an important feature in reducing the spread of diseases, but practices that prevent pathogen introduction, limit exposure, and reduce transmission are all important steps upon entry into the feedlot (Snowder et al., 2006).

 Viruses use two different strategies to infect hosts. Peterhans (2003) reviewed that a virus may either cause a persistent infection in individual animals to infect new hosts, or, viruses can also use the 'hit-and-run" strategy which is a short duration followed by a rapid transfer to a different host and continuing.

Virulence of BVDV has been characterized as having a broad range of effects based upon the amount of infection or severity of infection induced (Ridpath and Neill, 2007). Liebler-Tenorio et al. (2003) observed differences in clinical observations and rectal temperature changes in colostrum deprived, non-vaccinated steers less than 4 months of age when challenged with 2 different BVDV type 2 strains. Observations were made following an intranasal challenge of a naturally occurring low virulent strain, or a highly virulent strain isolated from a severe field outbreak. Calves receiving the low virulent strain expressed a mildly elevated body temperature at day 7 following challenge with no changes in feed intake or observable behavior differences. Conversely, calves inoculated with the highly virulent strain exhibited elevated body temperature ($>40^{o}$C) shortly following challenge which persisted for several days. Calves in the high virulence group also became lethargic and apathetic in behavior.

Similar findings were made by Kelling et al. (2002) where 5 different isolates of BVDV type II (2 highly virulent strains and 2 lowly virulent strains) were experimentally induced into 6 to 9 month old crossbred calves. Kelling et al. (2002) observed that calves inoculated with high-virulence strains developed more signs of respiratory tract disease, displayed elevated rectal temperature 6 days following inoculation, and exhibited more lethargic demeanor as compared to calves challenged with low-virulent strains.

The variation in virulence exposes challenges from a research standpoint. Efficacy of vaccine and vaccine use protocol studies in the field have been limited by the lack of repeatability and under-developed accepted criteria for appropriate research models. Traditionally, results from studies using BVDV type 2 have been readily available; however results from BVDV type 1 challenges are less common. One such BVDV1 strain that has been used in vaccines as well as a popular challenge strain is BVDV NY-1. Isolated from a field case in New York, calves infected with BVDV NY-1 were reported to respond with symptoms of pyrexia, reduction in white blood cells, diarrhea, reduced appetite, depression, and reddening of the gums (Baker et al., 1954). Ridpath and Neill (2007) summarized two problems in using BVDV NY-1 when conducting viral challenges; one, was that clinical presentation of infection was reported as mild in comparison to BVDV strains R5013, R2360, and CA0401186A, and two, was the conflicting results in vaccine efficacy in studies using BVDV NY-1 in the treatment vaccine as well as the challenge virus simultaneously.

One of the aforementioned BVDV strains used by Ridpath and Neill (2007) that compared BVDV NY-1 to other field strains was used in this research as well. Bovine Viral Diarrhea Virus CA0401186A was discovered from a PI calf in California that was one of 24 calves with brain and skeletal deformities born to heifers in a single herd. Tissues containing CA0401186a were submitted to the USDA-ARS National Animal Disease Center in Ames, IA from the Tulare Laboratory of the California Animal Health and Food Safety Laboratory. It was characterized as a noncytopathic biotype and found to be of the BVDV1b subgenotype (Ridpath and Neill, 2007).

Wilson (1989) defined the efficacy of a vaccine as its ability to reduce the overall level of respiratory disease. With this broad definition, a limited amount of literature exists pertaining to positive results for the efficacy of vaccination strategies in field trials. Reasons for this limitation were summarized by Wilson (1989) in that the methods of identifying respiratory disease are often based on clinical observation only. Variables including severity of illness and length of illness are un-detected. Finally, differences of repeatability issues which are unavailable in field studies as compared to laboratory evaluations (Wilson, 1989).

Considering the complex characteristics of BVDV, there is also complexity in BVDV vaccination strategies. Fulton et al. (2002) observed that BVDV was an important contributing factor to BRD in 2 studies, with BVDV subtype 1b being the most predominant. Fulton et al. (2002) and Fulton et al. (2005) also briefly reviewed popular BRD vaccines containing BVDV, and summarized that only a small number of commercially available vaccines actually contained BVDV 1b antigenic material. The

problem here is that even though cattle are vaccinated with BVDV 1a or 2, they may still be susceptible to the most common BVDV subtype, 1b. Fulton et al. (2005) concluded that vaccination with a MLV vaccine containing BVDV subtypes 1a and 2 did not prevent infection in vaccinated calves when exposed to PI calves with BVDV1b. Fulton et al. (2003a) also reported that vaccines containing BVDV1a induce lower antibodies to BVDV1b than to BVDV1a in 3 separate USDA licensed vaccines.

Summary

Bovine Respiratory Disease Complex is costly and remains widespread in beef cattle production systems. Morbidity and mortality result from exposure to viral and bacterial infections that work in concert to activate immune responses. Breed types and combinations of breeds have also been linked to a lower incidence of BRD. One major component of BRD is Bovine Viral Diarrhea Virus. One of the reasons BVDV draws much attention is due to Persistently Infected (PI) calves that go unidentified as a major source of spreading infection. Even in events of immunization, exposure to varying subtypes to BVDV can still result in infection leaving questionable results of vaccine efficacy. Thus, research is needed that addresses the genetic influences related to health, immune responses and animal performance in cattle exposed to BVDV.

CHAPTER III

EVALUATION OF WEIGHT GAIN, RECTAL TEMPERATURE AND IMMUNE

RESPONSE OF ANGUS-SIRED STEERS CHALLENGED WITH BOVINE VIRAL

DIARRHEA VIRUS

Introduction

Growing cattle on pasture and in feedlots can experience limited productivity

from the presence of viral and bacterial agents that make-up a complicated disease

system known as Bovine Respiratory Disease (BRD) complex (Gardner et al., 1999;

Roeber et al., 2001). During the growing and feeding phases in the beef cattle

production, a variety of stress elements work in concert with BRD pathogens to reduce

immune function and ultimately allow for infection and morbidity (Fulton et al., 2000).

Genetic variation in BRD occurrence is another subject with very little reported findings.

In those studies however, differences between breed types in susceptibility have been

identified (Muggli-Cockett et al., 1992; Snowder et al., 2006).

One disease that is an important component in the BRD complex is Bovine Viral

Diarrhea (BVD). As a serious threat to worldwide production (Peterhans et al., 2003),

BVD infections suppress immune function and initiate morbidity. Furthermore, BVD is

important from a cow-calf production standpoint because, BVD can cause abortions,

and, cow herds are the point of origin for persistently infected (PI) cattle. Calves born PI

are the result of their dams becoming infected with BVD in mid gestation before the

calf's immune system has fully developed. As a result, PI calves are born

immunotolerant to BVD infection and serve as harbors of the virus and facilitate constant exposure to non-PI cattle (Grooms, 2004).

Vaccines for BVD are commercially available as a means to manage and hopefully minimize occurrences of BVD infection or offset effects of viral infection. Efficacy of BVD vaccines have been questioned based on the genotypes and biotypes of virus included (Fulton et al., 2003a); for instance, BDV type 1a is typically in vaccines, but BDV type 1b has shown to be the most prevalent strain in the USA (Fulton et al., 2003a; Fulton et al., 2009). Additionally, virulence of BVDV seems to be subject of much variation as well in terms of symptoms associated with infection (Ridpath and Neill, 2007).

With such complicated factors that go into BRD complex from BVDV infection, the objective of this research was to investigate the health measures and performance traits of growing cattle of known genetic background that were vaccinated for BRD and then challenged with BVDV.

Materials and Methods

Animal methods were approved by the Texas A&M University Institutional Animal Care and Use Committee (AUP 2007-130).

Animals. Seventy-three Angus-sired steers of known genetic background were used in this trial. Steers were born in spring of 2007 and out of half *Bos indicus* cows from the Texas A&M University McGregor Genomics project. No prior exposure to viral vaccinations occurred before the trial began, and all steers were confirmed to be seronegative to BVD and IBR prior to trial initiation. Cattle were confirmed to be PI-

free through evaluation of ear notch sample by antigen capture ELISA at the Texas Veterinary medical Diagnostic Laboratory (TVMDL, Amarillo, TX).

Vaccine treatments. Steers were stratified by sire and genomics cow family across the vaccine treatments of Killed group (Killed, n = 33), modified-live group (MLV, n = 32), and non-vaccinated group (NON, n = 8).

Steers in the MLV treatment group were administered Arsenal 4.1® (Novartis Animal Health US, Inc.) on day -21 as per label instructions. Steers in the Killed treatment group were administered Vira-shield® (Novartis Animal Health US, Inc.) on day -42 and a booster at day -21 as per label instructions. Eight steers (4 from killed group and 4 from MLV group) were not vaccinated. Steers in the MLV group were kept separate from killed and NON steers for 5 days following vaccination.

Viral challenge. At day 0, steers were challenged with a type 1b non-cytopathic BVDV strain (CA0401186a) obtained from the USDA-ARS National Animal Disease Center (NADC) (Ridpath and Neill, 2007). This strain was isolated from a persistently infected calf and submitted to the NADC from Tulare laboratory of the California Animal Health and Food Safety Laboratory. Each steer was administered 5 mL of inoculum (1×10^5 TCID/mL) by placing 2.5 mL in each nasal passage; the animal's nose was elevated until it was visually observed that the steer had swallowed to confirm challenge virus was ingested. This particular strain of BVDV was chosen for this study because it had previously been reported to cause recognized immunological and clinical signs of morbidity, but without risk of extreme illness or death (Ridpath and Neill, 2007).

Sample collection. Rectal temperatures were measured via digital rectal thermometer on days 0, 1, 3, 9, 14, 28, and 42 following viral challenge. Body weights were also recorded, with those observations made at days -42, -21, 0, 14, 28, and 42. Average daily gain (ADG) was calculated for three phase increments of increment 1 (days 0 to 14), increment 2 (14 to 28), and increment 3 (28 to 42).

Serology. Collection of blood was conducted via jugular venipuncture into evacuated tubes on days -42 and -21 before the viral challenge, day 0, and days 14, 28, and 42 following viral challenge. Following collection, blood serum was harvested via centrifuge 2 to 4 hours following collection; centrifuge settings were to spin samples for 20 minutes at 4°C at 4000 RPM. Serum samples were then stored at -20°C until they were sent to Texas Veterinary Medical Diagnostic Laboratory (TVMDL) for quantification of serum neutraliazing (SN) antibody immunoglobulin G (IgG) titers for BVD Type 1, BVD Type 2, and IBR.

Clinical evaluation. Clinical observations were conducted twice daily for 14 days following challenge to assess apparent health with a score of 0 (no symptoms), or 1 to 5 (least severe to most severe) for commonly associated symptoms of BVD/BRD (cough, ocular secretion, nasal secretion, depression, diarrhea, and gauntness/shrink). From days 15 to 42 observations were only conducted once daily.

Diet. Steers were on wheat pasture from days -42 to day 0. From days 0 to 14, steers were fed in a set of holding pens. From days 14 to 42 steers were held in a small (1 hectare) pasture. During the period following BVDV challenge, the diet consisted of

average quality coastal bermudagrass hay fed, *ad libitum*, with no additional supplement following challenge.

Statistical analysis. Mixed model procedures of SAS (SAS Inst. Inc., Cary, NC) were used so that rectal temperature and antibody titers (transformed to a log base of 2) were analyzed as repeated measures with a model that contains vaccine treatment, day, sire, vaccine treatment × day, and vaccine treatment × sire, as fixed effects. An autoregressive covariance structure was utilized. Least squares means were separated with two-tailed t-tests following significant F-tests ($P < 0.05$) from the mixed model analyses. Average daily gain was analyzed with a similar model that included vaccine treatment, day, vaccine treatment × day, pen, maternal grandsire, and vaccine treatment × MGS interaction and day 0 weight as a covariate. Eight steers (n = 4 from killed vaccine group, n = 4 from MLV vaccine group) were removed from this analysis due to them not receiving viral challenge, and are not included in these results.

Results and Discussion

A general summary of the traits evaluated is presented in Table 3.1.

Table 3.1 Summary statistics for traits measured in 2008 trial

Variable	Mean	SD	Minimum	Maximum
IBR[1]	2.19	2.3	0	9.0
BVD Type 1[1]	3.65	3.8	0	13.0
BVD Type 2[1]	3.44	3.7	0	13.0
Rectal Temperature, °C	38.8	1.58	37.44	40
ADG, kg/day	.38	.26	-.87	.89

[1] Titer transformed to log base of 2

Results of the statistical analyses are presented in Table 3.2.

Table 3.2 Significance levels from mixed model analysis for health measures

Effect	IBR	BVD1	BVD2	Rectal temperature
Vaccine treatment	< 0.001	< 0.001	<0 .001	0.66
Day	<0 .001	< 0.001	< 0.001	0.12
Vaccine treatment × Day	<0 .001	< 0.001	< 0.001	0.94
Sire	0.33	0.17	0.12	0.80
Vaccine treatment × Sire	0.54	0.06	0.09	0.98

IBR titers. Titers for IBR were evaluated in an attempt to evaluate vaccine effectiveness and pathogen exposure. Steers in both vaccinated groups should have developed titers to IBR, whereas those not vaccinated should not have developed titers, and, this was observed. Differences in IgG titer levels for IBR were observed due to vaccine treatment, day, and vaccine treatment × day interaction ($P < 0.01$). Least squares means for the interaction are presented in Figure 3.1.
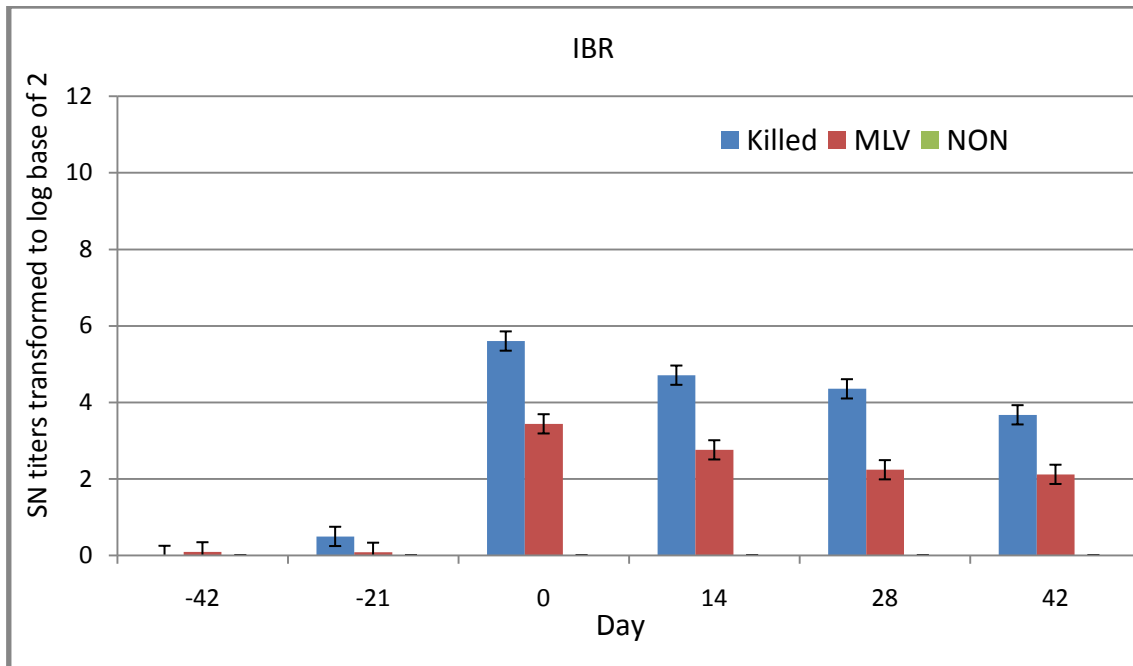
Figure 3.1. Least squares means for serum neutralizing antibody titer (log$_2$) for IBR in steers of killed vaccine (Killed), modified-live vaccine (MLV), or non-vaccinated control (NON) groups across time in 2008

Large differences between treatment groups were observed with steers from the Killed group being the highest at days 0, 14, 28, and 42 as compared to MLV and NON vaccinated cattle. Titer levels within the Killed group were highest at day 0 ($P < 0.01$), followed by day 14, then 28, and, these were higher than days -42, -21, and 42 for cattle within the Killed group. Titers for IBR within the Killed group at day 42 were higher than days -42 and -21.

Steers in the MLV group had higher IBR titers than steers from the NON group at all dates measured after the viral challenge including day 0. Differences within the MLV group across collection dates did vary however, with IBR titers for the MLV group being highest at day 0 and decreasing steadily through day 42 ($P < 0.01$). After the

challenge on day 0, serum neutralizing IBR titers for MLV steers were lowest at days 28, and 42 ($P < 0.01$).

BVD Type 1 titers. Differences in BVD Type 1 titers were observed due to vaccine treatment, day, and vaccine treatment × day interaction ($P < 0.01$). Figure 3.2 shows the least squares means from the interaction.

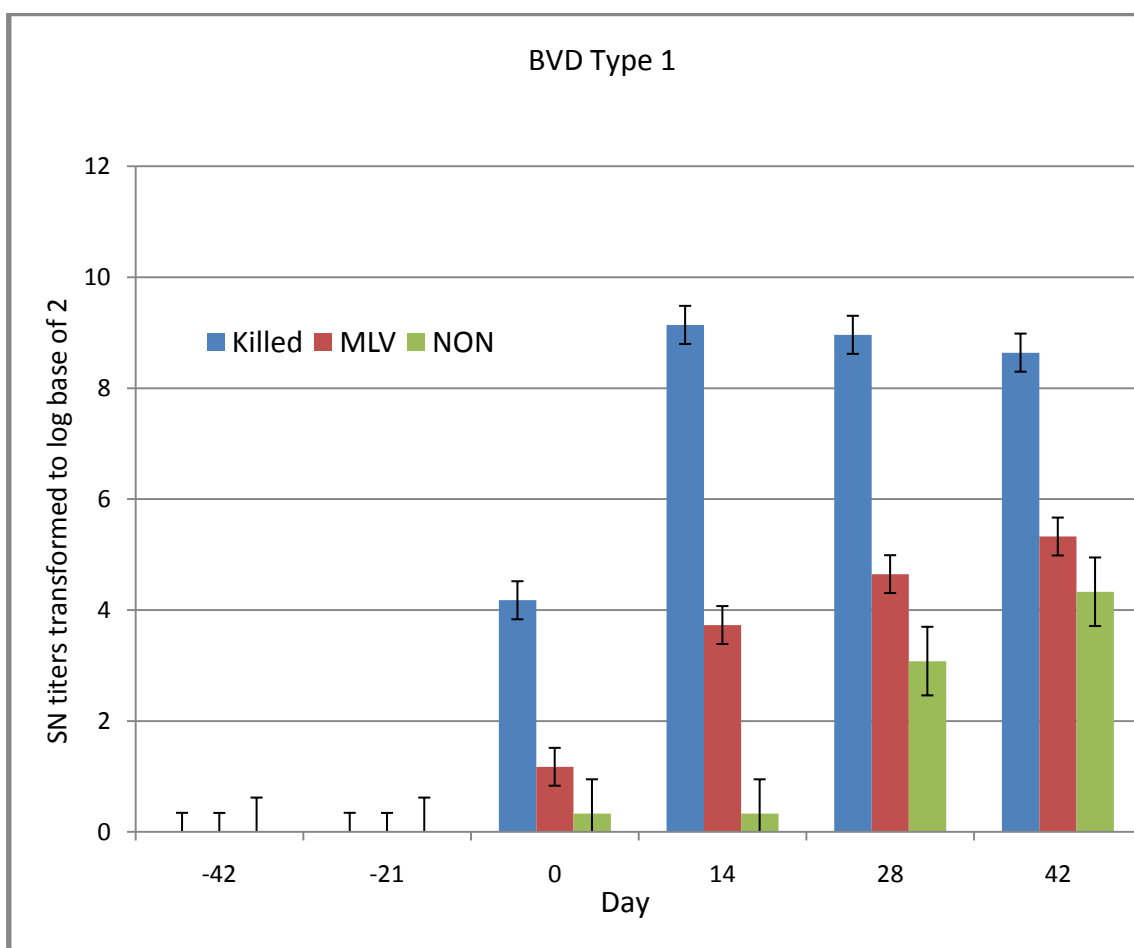Figure 3.2 Least squares means for serum neutralizing antibody titer (log$_2$) for BVD Type 1 in steers of killed vaccine (Killed), modified-live vaccine (MLV), or non-vaccinated control (NON) groups across time in 2008

On days 0 through 42 it was observed that BVD Type 2 titers from the Killed group were higher than the MLV or NON vaccinated group at each day. A large difference in titers from day 0 to day 14 was observed ($P < 0.01$). Following this peak BVD Type 1 titers from the Killed group remained similar through day 42.

Calves in the MLV group displayed higher IgG titers for BVD type 1 than the NON group at day 14, 28, and 42. Titers from the MLV group steadily increased from day 0 through 42, which was the day with the highest level observed for cattle in this group ($P < 0.01$).

Titers for BVD Type 1 from calves within the NON group were observed to resemble titers from the MLV group. Titers from the NON group increased gradually until their highest point was observed at day 42.

BVD Type 2 titers. Differences in BVD Type 2 titers were observed due to vaccine treatment, day, and vaccine treatment × day ($P < 0.01$). Least squares means for the vaccine × day interaction are presented in Figure 3.3.
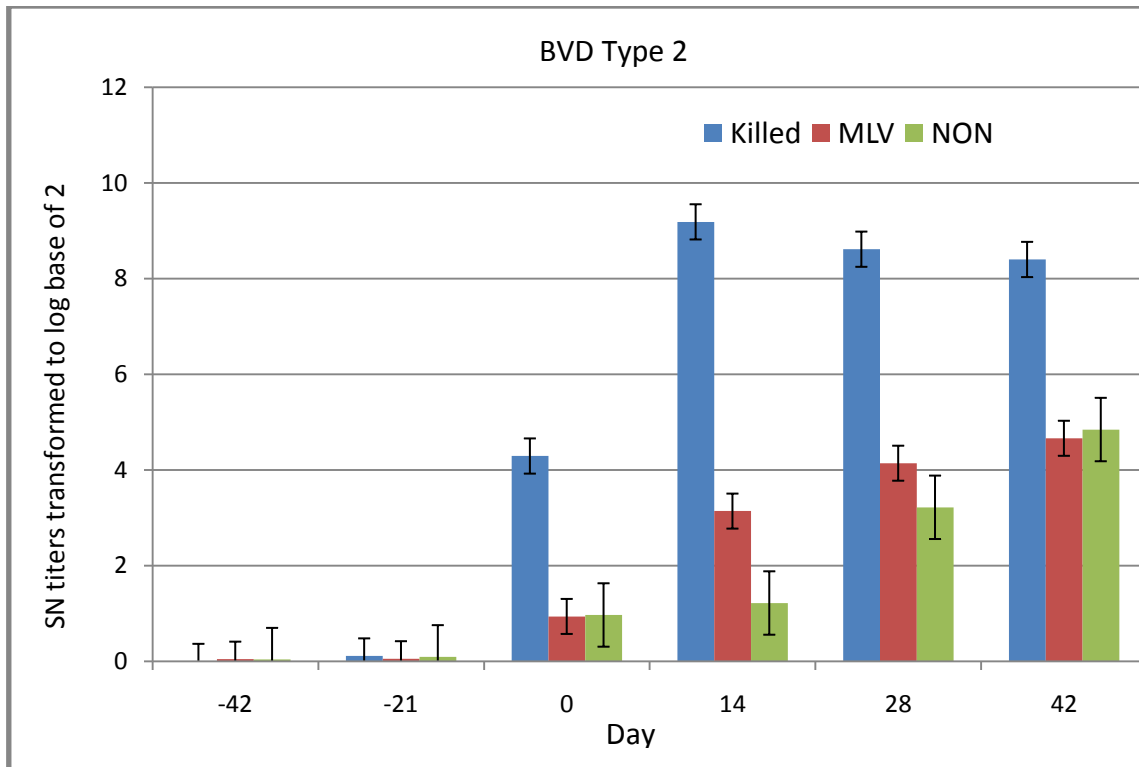
Figure 3.3. Least squares means for serum neutralizing antibody titer (log$_2$) for BVD Type 2 in steers of killed vaccine (Killed), modified-live vaccine (MLV), or non-vaccinated control (NON) groups across time in 2008

Serum neutralizing titers for BVD Type 2 from cattle in the Killed group were the highest at all days measured following the viral challenge including day 0 as compared to cattle in the MLV and NON groups ($P < 0.01$). A large increase in blood titers for BVD Type 2 from the Killed group was observed from day 0 to day 14. Titer levels for BVD Type 2 within the Killed group peaked at day 14 ($P < 0.05$). Following this peak a slight decrease in titers were observed and titer held at this level with no significant changes from day 28 through 42.

Blood titer levels for BVD Type 2 from the MLV group steadily increased from day 0 through 42. Unlike the pattern of Killed group BVD Type 2, titers in the MLV group peaked at days 28 and 42 ($P < 0.01$). Serum titers for BVD Type 2 from cattle in the NON group steadily increase from day 0 to peak at day 42 ($P < 0.01$).

Average daily gain. Differences in average daily gain were also observed due to vaccination treatment groups. Least squares means for ADG are shown in Table 3.3.

Table 3.3. Least squares means and SE for average daily gain for the three, 2-week periods following BVDV challenge

	Vaccine Treatment		
	Killed	MLV	NON
ADG d 0-14 (kg)	$0.45^a \pm .15$	$0.00^b \pm .15$	$-0.50^b \pm .29$
ADG d 14-28 (kg)	$1.79^a \pm .08$	$1.27^b \pm .08$	$1.36^b \pm .15$
ADG d 28-42 (kg)	$0.27^a \pm .10$	$0.14^a \pm .11$	$0.14^a \pm .20$

[ab]Least squares means with different superscripts within a row differ ($P < 0.05$)

In 2 out of 3 two-week observation periods, the ADG for the Killed group was higher than MLV and NON treatment groups ($P < 0.05$). Additionally, it was observed that cattle from the NON group lost 0.5 kg/d, which was 0.95kg/d lower than the mean of the Killed group ($P < 0.05$).

Summary. Vaccine strategies are beneficial when incorporated into growing cattle systems. Across the entire trial, cattle vaccinated with either an MLV or Killed vaccine had higher IgG titer for IBR, BVD Type 1, and BVD Type 2 than non-vaccinated cattle. For the first 14 d following challenge the Killed group gained 0.45

kg/d, the MLV group gained no weight, and the non-vaccinated steers lost 0.50 kg/d. All cattle gained weight in the second 14-d period, but the killed group still had an advantage over the MLV and NON groups. Although steers in the killed vaccine group had a higher ADG than the MLV and non-vaccinated steers in the first 28 d, there was no obvious reason for this separation in ADG between the killed and MLV vaccinated groups, particularly since there were no differences in rectal temperatures. In this trial, the use of vaccines sufficiently stimulated the immune system before the viral challenge which aided in maintaining higher ADG in vaccinated steers as compared to non-vaccinated animals.

No clear differences due to sire and sire × treatment were observed in any measurement taken. It was unexpected that no animals had high enough rectal temperature (> 40.0 C) to be treated with an antimicrobial, and it appears that the relationship between these IgG titers and rectal temperature is not clear. Very large differences in titers were observed among individuals.

CHAPTER IV

EVALUATION OF FEED INTAKE, HEALTH AND IMMUNE RESPONSE IN

ANGUS-SIRED STEERS CHALLENGED WITH

BOVINE VIRAL DIARRHEA VIRUS

Introduction

Bovine Respiratory Disease (BRD) can be an enormous burden in beef cattle operations. In terms of financial strain on the U.S. cattle industry, Griffin (1997) summarized an estimated annual loss of $750 million due to BRD incidence. Galyean et al. (1999) calculated that 52% of U.S. feedlot morbidity was a result of BRD. Morbidity from BRD is most common in the first 45 days after arrival, as during this time cattle are subjected to and recovering from added levels of stress which requires additional management (Edwards, 2010). Illness from BRD often results in reduced appetite along with other signs of disease. Buhman et al. (2000) observed reduced intake behavior and lower frequency of intake in newly arrived feedlot cattle identified as sick as compared to non-sick contemporaries.

Morbidity due to BRD comes from pathogenic bacteria, typically following the presence of one of more viral infections (Daniels et al., 2000). The viral components of BRD create suppressed immune function and leads to infectious bacterial pneumonia, most commonly caused my *Mannheimia haemolytica* (Fulton et al., 2000; Fulton et al., 2002; Fulton et al., 2006). Differences among breed types have also accounted for BRD occurrences. Muggli-Cockett et al. (1992) reported Angus cattle had the lowest percentage of occurrence compared to several other breeds.

Bovine Viral Diarrhea Virus is a component of BRD which is a subject of great interest due to the production nuisances BVD creates in cow herds which carries over into the growing and finishing phases of beef production (Larson et al., 2002). Vaccines containing BVDV antigenic material are available and contain modified-live or killed forms of viral agents which initiate cattle immune systems (Faries, 1999). The efficacy of vaccines tends to be difficult to measure in field studies, mainly from the lack of standardized methods for detecting clinical illness and illness severity.

Due to the amount of variation in results that involve vaccine efficacy combined with the lack of reported information pertaining to the genetic influence in respiratory illness, the objective of this research was to investigate the immune response, feed intake, growth performance, and genetic variability of steers with known genetic background when challenge with BVDV following different vaccination strategies.

Materials and Methods

Animal methods were approved by the Texas A&M University Institutional Animal Care and Use Committee (AUP 2007-130).

Animals. Ninety-three Angus-sired, yearlings steers produced from the Texas A&M University McGregor Genomics Project cows, born in the spring of 2008 were used to conduct this trial. No prior exposure to viral vaccinations occurred before the trial began and all steers were confirmed to be seronegative to BVD and IBR prior to trial initiation. Cattle were confirmed to be PI free through evaluation of ear notch

sample by antigen capture ELISA at the Texas Veterinary medical Diagnostic

Laboratory (TVMDL, Amarillo, TX).

Vaccine treatments. Steers were stratified by sire and genomics cow family

across 3 vaccine treatment groups; killed vaccine (Killed) group (n = 29), modified-live

(MLV) group (n = 34), and non vaccinated (NON) group (n = 31).

Steers in the Killed group received initial Vira-shield® (Novartis Animal Health

US, Inc.) vaccine on day -56 and a booster dose on day -35 as per label instruction.

Steers in the MLV group were vaccinated with Arsenal 4.1® (Novartis Animal Health

US, Inc.) on day -35 prior to the challenge and remaining 31 steers of the NON group

remained as non-vaccinated. Steers receiving the MLV were kept separate from Killed

and NON steers for 7 days following vaccination. These were the same vaccines used in

the 2008 trial.

Viral challenge. At day 0, steers were challenged with a type 1b non-cytopathic

BVDV strain (CA0401186a) obtained from the USDA-ARS National Animal Disease

Center (Ridpath and Neill, 2007). This strain was isolated from a persistently infected calf

and submitted to the NADC from Tulare laboratory of the California Animal Health and

Food Safety Laboratory. Each steer was administered 5 mL of inoculum (1×10^5

TCID/mL) by placing 2.5 mL in each nasal passage; the animal's nose was elevated until

it was visually observed that the steer had swallowed to confirm challenge virus was

ingested. This particular strain of BVDV was chosen for this study because it had

previously been reported to cause recognized immunological and clinical signs of

morbidity, but without risk of extreme illness or death (Ridpath and Neill, 2007). This was

the same BVDV strain and challenge technique used as in the previous year's (2008) trial.

Sample collection. Body weights were measured and rectal temperatures were evaluated via digital rectal thermometer on days 0, 1, 3, 7, 10, 14, 28, and 42 following viral challenge. Average daily gain (ADG) was calculated for three periods: period 1 (days 0 to 14), period 2 (days 14 to 28), and period 3 (days 28 to 42).

Serology. Collection of blood was conducted via jugular venipuncture into evacuated tubes on days -56 and -35 before the viral challenge, days 0, 14, 28, and 42 following viral challenge. Following collection, blood serum was harvested via centrifuge 2 to 4 hours following collection, centrifuge settings were to spin samples for 20 minutes at 4^oC at 4000 RPM. Serum samples were then stored at -20^oC until they were sent to Texas Veterinary Medical Diagnostic Laboratory (TVMDL) for quantification of serum neutraliazing (SN) antibody immunoglobulin G (IgG) titers for BVD Type 1, BVD Type 2, and IBR.

Clinical evaluation. Clinical observations were conducted twice daily for 14 days following challenge to assess apparent health with a score of 0 (no symptoms), or 1 to 5 (least severe to most severe) for commonly associated symptoms of BVD/BRD (cough, ocular secretion, nasal secretion, depression, diarrhea, and gauntness/shrink). From days 15 to 42 observations were only conducted once daily. Animals exhibiting rectal temperatures over 40^oC were administered a commercially available antimicrobial approved for cattle and dosed per label directions.

Diet and feed intake monitoring. Cattle were fed a high-forage growing diet that consisted of 35.5% corn, 32.5% chopped alfalfa hay, 24.5% dry distillers grains, 2.5% commercial premix, and 5% molasses. Cattle were housed and fed at the Texas A&M University Beef Systems Research Unit on FM 50 where 4 pens are equipped with GrowSafe® feed intake observation units. Cattle were housed so that the treatment groups were stratified across the 4 pens with approximately 23 steers per pen.

Statistical analysis. Mixed model procedures of SAS (SAS Inst. Inc., Cary, NC) were used so that rectal temperature and titer level (transformed to a log-base of 2) were analyzed as repeated measures with vaccine treatment, day, vaccine treatment × day, sire or family, and vaccine treatment × sire evaluated as fixed effects. An autoregressive covariance structure was utilized. Average daily gain and feed intake were analyzed with similar model that included vaccine treatment, day, vaccine treatment × day interaction, pen, maternal grandsire and vaccine treatment × maternal grandsire interaction. Least squares means were compared for effects that produced a significant F-test ($P < 0.05$).

Results and Discussion

General summary statistics for traits evaluated are provided in Table 4.1.

Table 4.1 Summary statistics for traits studied in 2009 trial

Variable	Mean	SD	Minimum	Maximum
IBR[1]	1.61	2.13	0	7.0
BVD Type 1[1]	3.51	3.69	0	13.0
BVD Type 2[1]	3.08	3.55	0	14.04
Rectal temperature, ^{o}C	39.29	.54	37.2	41.30
ADG, kg	1.7	.38	.7	2.8
Daily intake, kg	12.18	3.58	0	24.6

[1]Titer transformed to log base of 2

Results from statistical analyses are provided in Table 4.2.

Table 4.2 Significance levels from mixed model analysis for IBR, BVD Type 1, BVD Type 2, and rectal temperature

Effect	IBR	BVD Type 1	BVD Type 2	Rectal Temperature
Vaccine treatment	< 0.001	< 0.001	< 0.001	< 0.05
Day	< 0.001	< 0.001	< 0.001	< 0.001
Vaccine ×day	< 0.001	< 0.001	< 0.001	< 0.05
Sire	0.71	0.06	0.20	< 0.05
Vaccine × sire	0.81	0.14	0.60	0.64

IBR titers. Titers for IBR were evaluated as a control variable as cattle that were vaccinated for BRD should be the only animals with IBR titers. Vaccine treatment, day, and vaccine treatment × day interaction affected IBR titers ($P < 0.01$). Figure 4.1 shows titers for IBR across vaccine groups and sample dates.
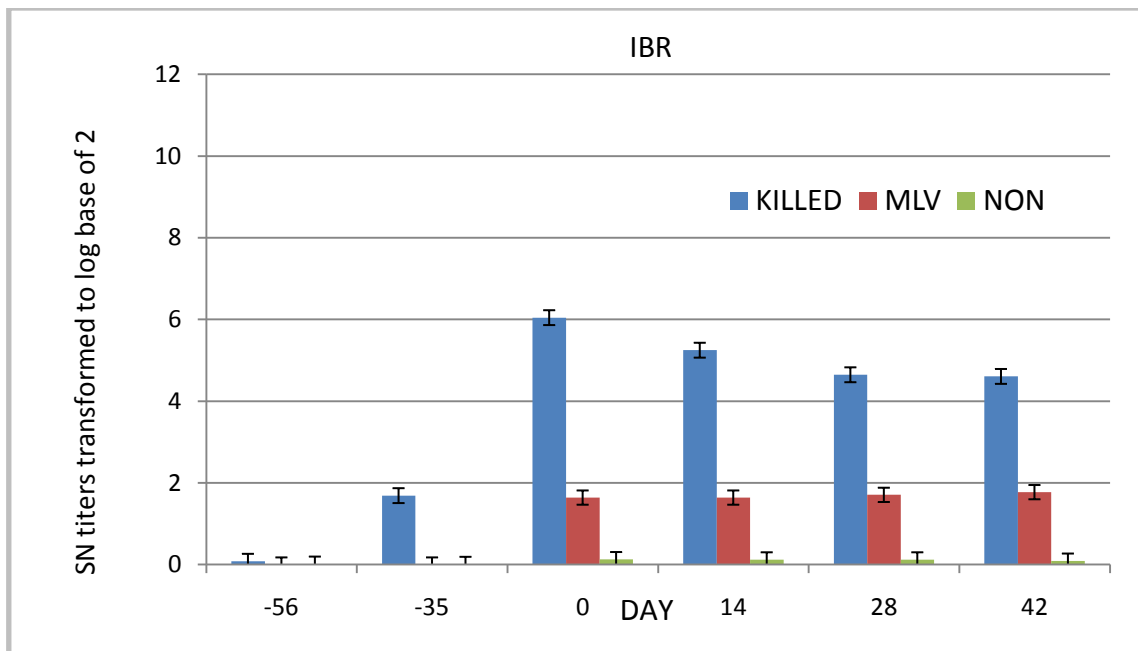
Figure 4.1. Least squares means for serum neutralizing antibody titer response ($\log_2$) of IBR across days in steers of killed vaccine (Killed), modified-live vaccine (MLV), or non-vaccinated control (NON) groups in 2009

Large titer differences for cattle in the Killed group were observed at days -35, 0, 14, 28, and 42 as compared to MLV and non-vaccinated (NON) ($P < 0.01$). The highest IBR titers for the Killed group were observed at day 0 compared to days -56, -35, 14, 28, 42 ($P < 0.01$). IBR titers in the Killed group at day 14 were also higher than days -56, -35, 28, and 42 ($P < 0.01$). IBR titers for the Killed group at days 28 and 42 were not different from each other ($P = 0.86$), but were higher than days -56 and -35 ($P < 0.01$).

Titer differences for IBR in the MLV group were higher than the NON group at days 0, 14, 28, 42 ($P < 0.01$) but differences in IBR titers for the MLV group for days 0, 14, 28, 42 were not different from each other ($P > 0.10$).

BVD Type 1 titers. Vaccine treatment, day, and vaccine treatment by day interaction affected BVD Type 1 titers ($P < 0.01$). Figure 4.2 shows least squares means for BVD Type 1 titers across vaccine group and sample date combinations.

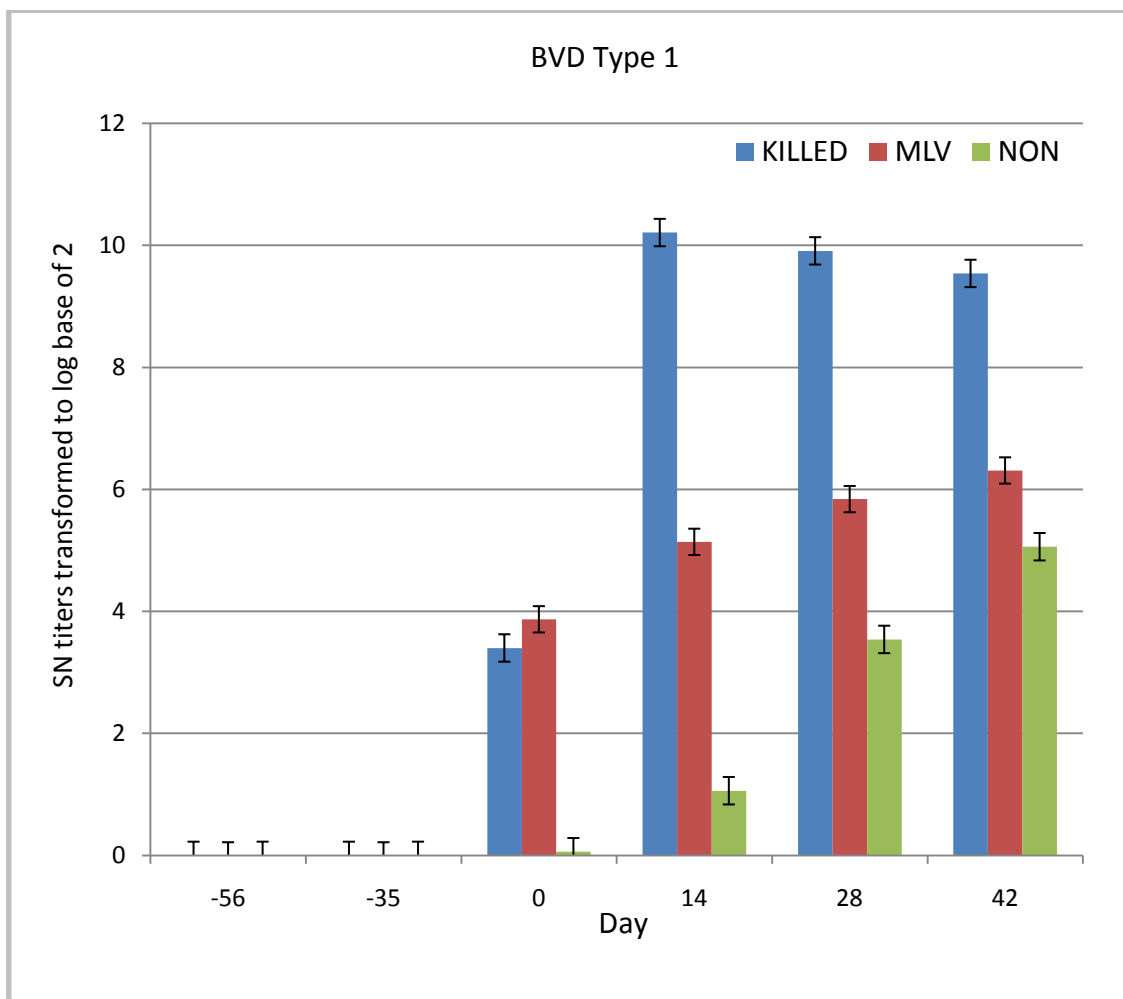

Figure 4.2. Least squares means for serum neutralizing antibody titer response (log_2) of BVD Type 1 across days in steers of killed vaccine (Killed), modified-live vaccine (MLV), or non-vaccinated control (NON) groups in 2009

Titers for BVD Type 1 in the Killed group were higher than MLV and NON groups at days 14, 28, and 42 ($P < 0.01$). Differences between days within the same vaccine group were also observed. Titers in the Killed group were highest at days 14 and 28. Titers for BVD Type 1 in the Killed group at day 14 were higher than days -56, -35, 0, and 42 ($P < 0.01$) and titers at day 28 were higher than -56, -35, and 0, but were not significantly different from day 14 or day 28 ($P = 0.16$, $P = 0.08$ respectively). Titers in the Killed group at day 0 were higher than -56 and -35 ($P < 0.01$), but not days 14, 28, and 42.

Titers for BVD Type 1 in the MLV group were higher than titers from the NON vaccine group at days 0, 14, 28, 42 ($P < 0.01$). Titers from the MLV group were observed to be similar to Titers from the Killed group at day 0 but were not significantly different ($P = 0.13$). Titers for BVD Type 1 in the MLV group were highest at day 42 as compared to any other day ($P < 0.05$).

Titers for BVD Type 1 in the NON group progressively increased over time with the highest recorded titers being observed at day 42 ($P < 0.01$).

BVD Type 2 titers. Figure 4.3 shows least squares means for BVD Type 2 titers across vaccine group and sample dates observed.
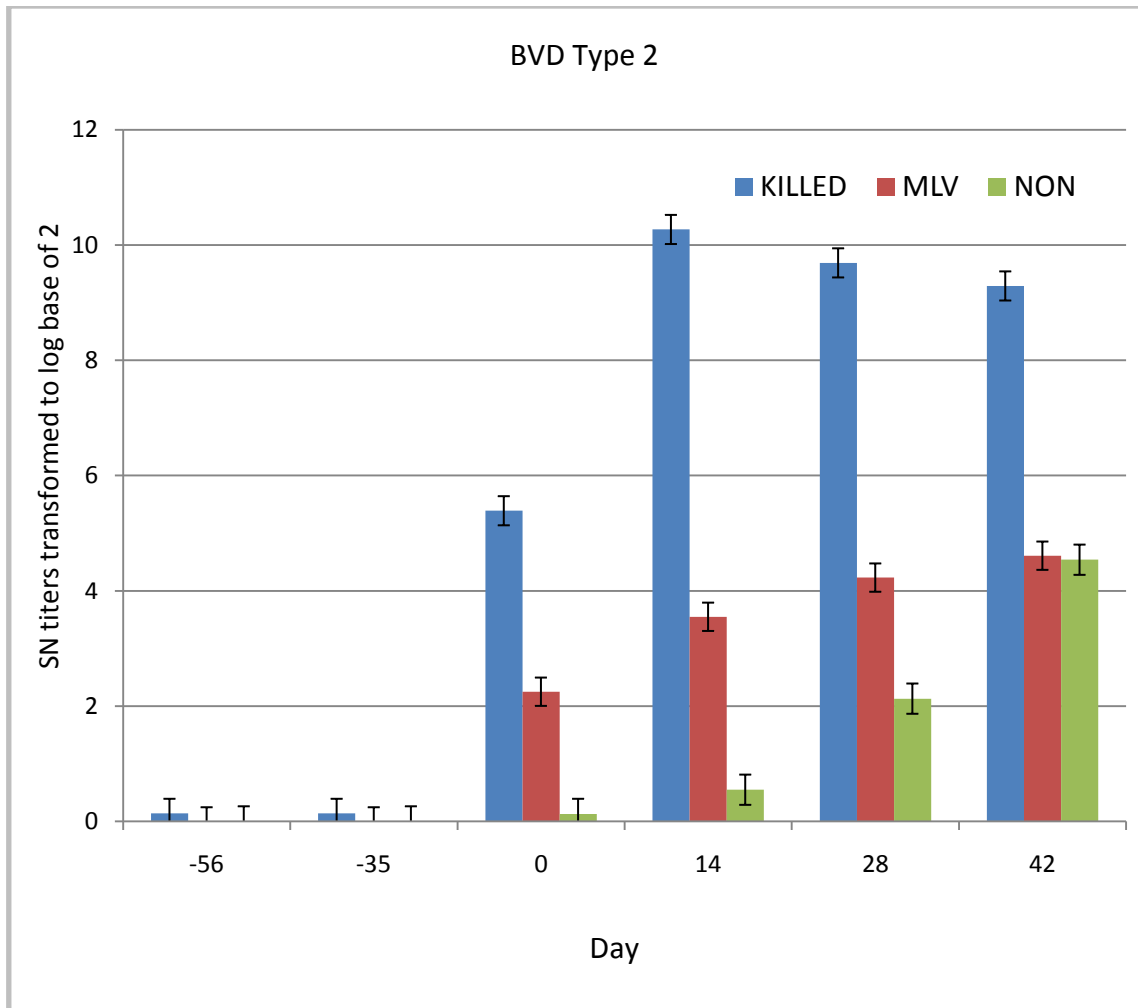
Figure 4.3. Least squares means for serum neutralizing antibody titer response ($\log_2$) of BVD Type 2 across days in steers of killed vaccine (Killed), modified-live vaccine (MLV), or non-vaccinated control (NON) groups in 2009

Titers for BVD Type 2 in the Killed group were higher than titers from the MLV and NON treatment groups at all dates from day 0 through day 42 ($P < 0.01$). Killed group titers were highest at day 14, which was significant as compared to other dates within that treatment group ($P < 0.01$). Titers in the Killed group for BVD Type 2 on

days 28 and 42 were not significantly different ($P = 0.06$), but were still substantially

higher than days 0, -56, -35 in the Killed group ($P < 0.01$).

Titers for BVD Type 2 in the MLV group were higher than the NON group at

days 0, 14, 28 ($P < 0.01$), but were not significantly different the NON group at day 42

($P = 0.87$). Titers for BVD Type 2 in the MLV group were highest at days 28 and 42.

Similar to BVD Type 1, BVD Type 2 titers from cattle in the NON group progressively

increased over time and were highest at day 42.

Pen assignment was not a significant source of variation for titer response for

IBR, BVD Type 1, or BVD Type 2 ($P = 0.34$, 0.63, and 0.56, respectively).

Rectal temperature. Differences in rectal temperature were observed due to

vaccine treatment, day, vaccine treatment by × day interaction, and sire. Figure 4.4

shows differences in rectal temperatures. Peak least squares means for rectal

temperature response occurred at day 7. Steers from the MLV group remained lower

(39.2°C) for rectal temperatures than steers in the Killed group and NON group ($P <$

0.05), which were highest at 39.8°C and 39.8°C, respectively on day 7.
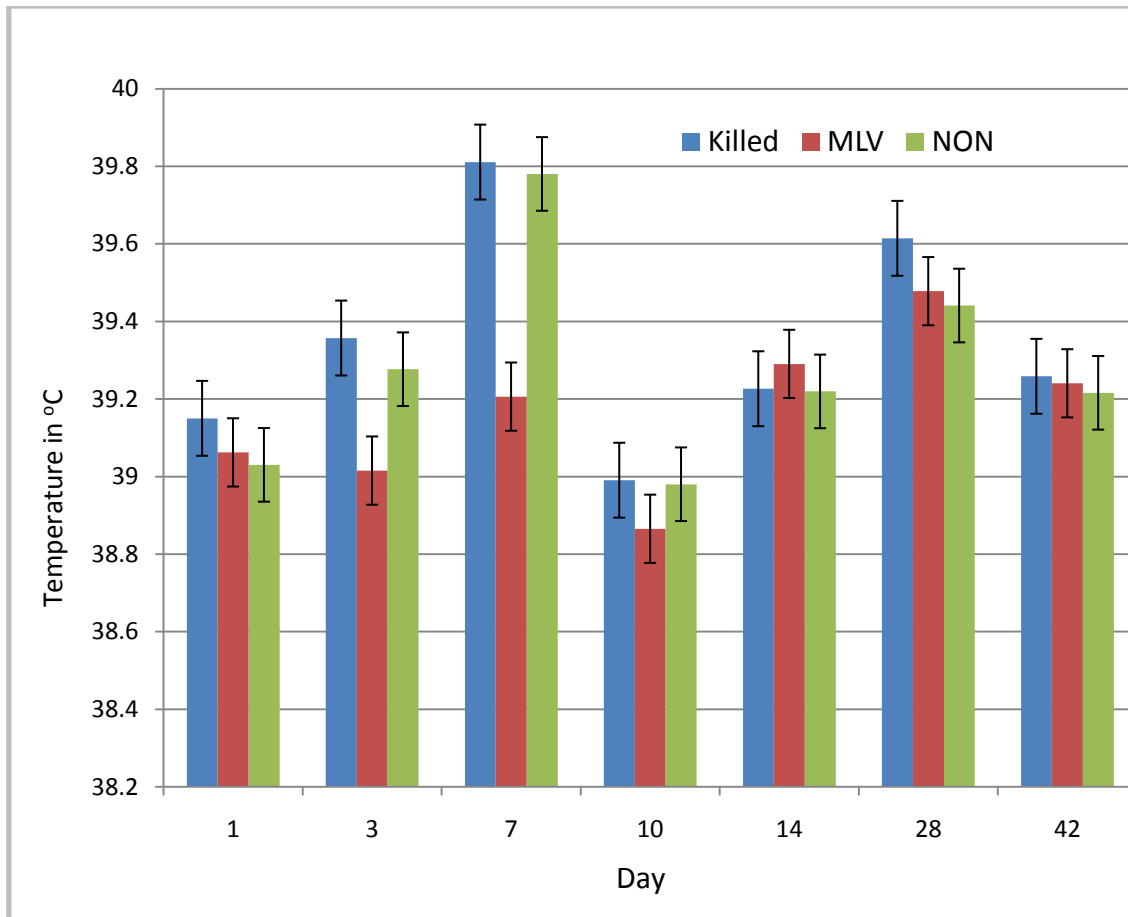
Figure 4.4. Least squares means for rectal temperature (^{o}C) over time of steers in killed vaccine (Killed), modified-live vaccine (MLV), or non-vaccinated (NON) groups following BVDV challenge on day 0

Following day 7, rectal temperatures dropped below 39.0^{o}C for all vaccine groups then steadily rose through day 28 when it was observed that temperatures for cattle in the Killed group reached a secondary peak which was significantly higher than all measured rectal temperatures within the killed group days except day7 ($P < 0.05$). A similar pattern was observed for the NON cattle as their temperatures appeared to peak again at day 28 as compared to other measured dates, excluding days 3 and 7 ($P \leq 0.05$).

Antimicrobial treatment was assessed when observed rectal temperatures were greater than 40°C. A summary of the number of cattle treated across vaccine treatment group are presented in Table 4.3.

Table 4.3. Number of cattle treated (rectal temperature > 40°C) days 0 to 14 across vaccine groups

	Killed	MLV	NON	TOTAL
NO	14	34	17	65
YES	14	0	14	28

Yes – rectal temperature > 40°C
No – rectal temperature ≤ 40°C

Following viral challenge and ensuing effects of inflammation, the observed rectal temperatures at day 42 across all three returned to within 0.1°C of each other at 39.3°C, 39.2°C, and 39.2°C for Killed, MLV, and NON groups, respectively.

Differences in rectal temperature due to sire were also observed, and means are presented in Table 4.4.

Table 4.4. Least squares means and standard errors for rectal temperature for sire groups across all days

Sire	Rectal Temperature SE
006N[a]	39.28°C ± 0.05
012R[ab]	39.34°C ± 0.07
023N[bc]	39.45°C ± 0.06
032N[abc]	39.33°C ± 0.12
178N[ad]	39.15°C ± 0.09
215R[de]	39.07°C ± 0.11
252P[de]	39.12°C ± 0.07
548P[abc]	39.36°C ± 0.04

[abcde]Least squares means with different subscripts are different ($P < 0.05$).

Calves sired by 252P and 215R displayed the lowest overall least squares mean temperature (39.12°C and 39.12°C, respectively) for all days collected as compared to calves sired by 548P, 032N, 023N, 012R, and 006N regardless of vaccine treatment group ($P < 0.05$).

Feed intake. Differences in daily feed intake were observed due to vaccine treatment, day, and vaccine treatment × day. Figure 4.5 shows least squares means for daily intake across vaccine groups. Steers from the MLV group consumed more than steers in the Killed group and the NON group. The largest variation in differences between the vaccine groups appeared to occur from days 7 to 10 following the BDVD challenge where NON steers consumed 0.59 to 4.92 kg/day less that MLV and Killed groups. Figure 4.6 shows least squares means for daily intake across vaccine groups for the first 14 days following viral challenge, including the separation of cattle that were treated and not treated within those vaccine groups based on rectal temperature (>40.0°C). These results are similar to (Buhman et al., 2000) in that cattle that are treated displayed a reduction in appetite.
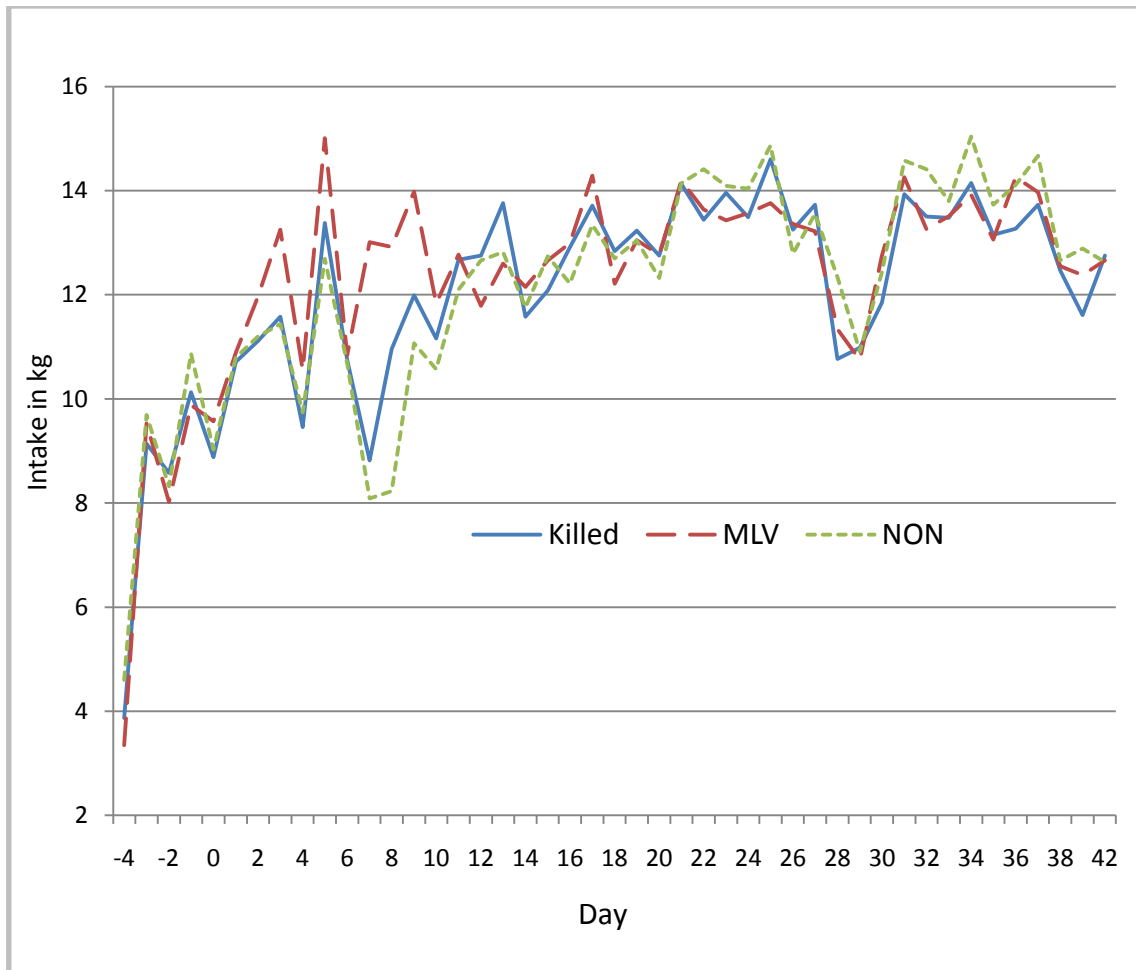
Figure 4.5. Least squares means for daily feed intake (kg) for the vaccine treatment by day interaction ($P < 0.05$)
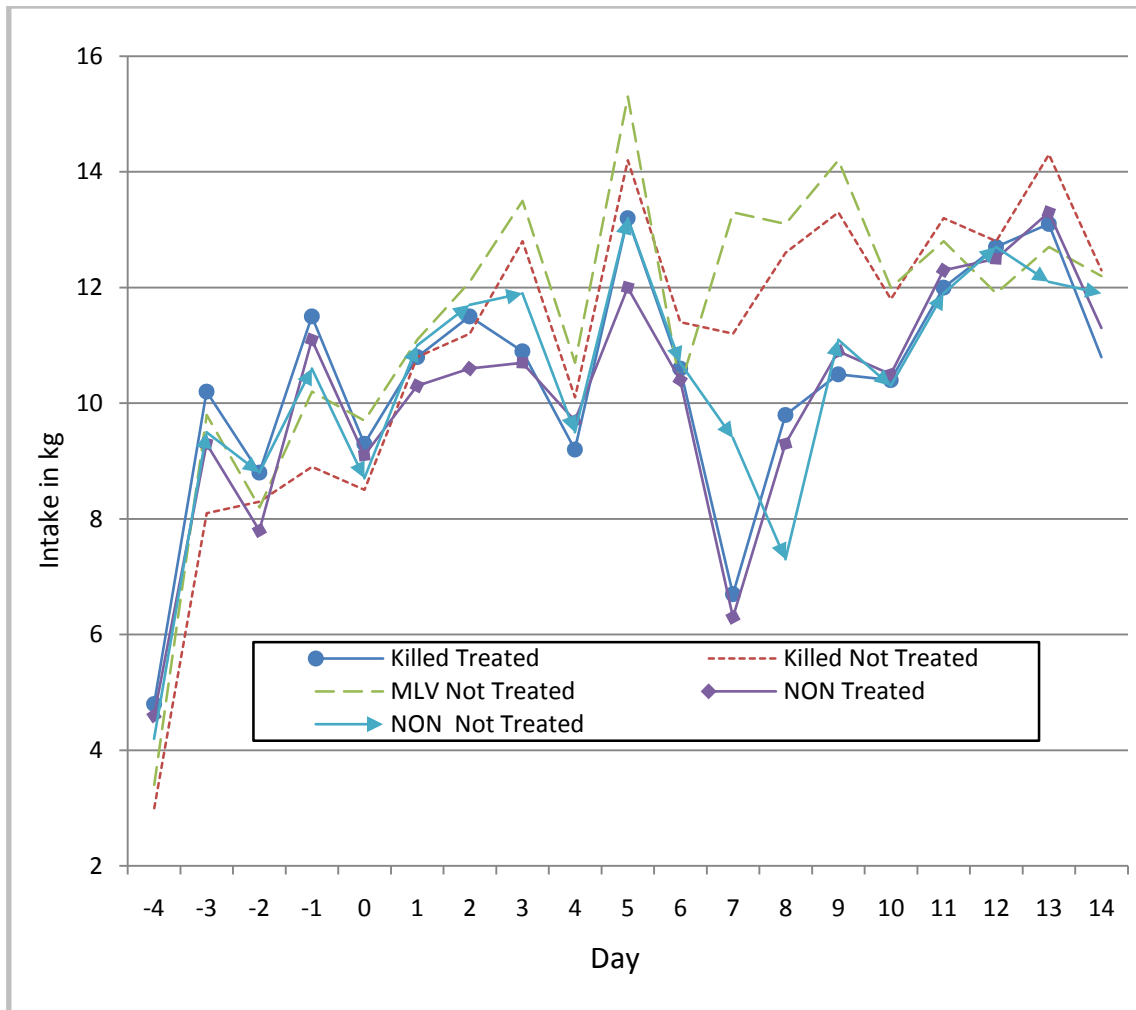
Figure 4.6. Least squares means for daily feed intake of steers in the 14 days following BVDV challenge due to combinations of vaccine treatment group and whether or not they were treated (based on rectal temperature > 40.0 C)

Differences in daily feed intake were observed due to maternal grand sire (MGS) and MGS × vaccine treatment interaction (least squares means are presented in Table 4.5)

Table 4.5. Least squares means for overall daily feed intake for vaccine treatment by maternal-grandsire (MGS) interaction

	Vaccine treatment		
MGS	Killed ± SE	MLV ± SE	NON ± SE
297J	$13.2^{ax} \pm 0.23$	$12.9^{ax} \pm 0.23$	$12.3^{ay} \pm 0.25$
432H	$11.9^{bx} \pm 0.19$	$12.4^{ay} \pm 0.18$	$11.8^{abx} \pm 0.19$
437J	$11.9^{bcx} \pm 0.25$	$12.9^{ay} \pm 0.21$	$12.5^{acy} \pm 0.21$
551G	$11.2^{cdx} \pm 0.25$	$11.6^{bx} \pm 0.23$	$11.4^{bx} \pm 0.25$

[abc]LS means with different superscripts within maternal grandsire column are different ($P < 0.05$)
[xy]LS means with different superscripts within vaccine treatment row are different ($P < 0.05$)

Steers in the Killed group from MGS 297J consumed more than steers from MGS 432H, 437J, and 551G ($P < 0.05$). When comparing grand-progeny of 297J across vaccine treatments, cattle in the NON group consumed 0.9 kg/day less than those in the Killed group and 0.6 kg/day less than those in the MLV group ($P < 0.05$). Similar patterns of differences across vaccine treatments within a MGS group were observed for steers with 432H in their pedigree. Steers from 432H in the MLV group consumed 0.5 kg/day and 0.6 kg/day more than steers from the Killed group or the NON group, respectively ($P < 0.05$). Steers from MGS 437J consumed 1 kg/day and .6 kg/day less as part of Killed group as compared to steers from 437J in the MLV and NON groups,

respectively. Calves from MGS 551G, regardless of vaccine group, consumed the least in all cases ($P < 0.05$).

Average daily gain. Calculations of ADG were conducted but no differences existed in any 14-day period, which is similar to observations made by Wittum et al, (1996) in that no differences in ADG were observed due to treatment or vaccine status.

Summary. Similar to results in the 2008 trial, vaccinated cattle responded with higher titers for IBR, BVD Type 1 and BVD Type 2 as compared to non-vaccinated cattle following the viral challenge. Differences in rectal temperature were also observed due to sire. Cattle in the MLV treatment group remained lower following viral challenge, and as a result no cattle in the MLV group were treated in the first 14 days post challenge. Additionally, all steers in the MLV group and fourteen steers in the Killed vaccine group that were not treated maintained an increasing daily intake days 6 through 9 post challenge when all cattle in the NON vaccine group and fourteen steers in the Killed vaccine group that were treated exhibited a sharp decrease in daily feed consumption. Differences in daily feed intake were observed due to MGS and MGS × treatment interaction. Preliminary analysis for ADG and daily feed intake included sire, but it was not a significant source of variation. MGS was then investigated and accounted for differences and was therefore left in the analysis. These interactions that exist between sires and treatments as well as MGS and treatments provide evidence that genetics along with vaccine strategy management plays a significant role in the differences of animal health response to BVDV infection. The role of genetic background on vaccination response to BVD has not been reported in the literature.

Genetic background has been shown to influence BRD incidence (Muggli-Cockett et al., 1992; Snowder et al., 2006). If future studies find genetic background influences BVDV immune response then this information would need to be documented for improved BVD control strategies and could impact producers' animal selection and vaccine choices, and even veterinarian recommendations.

CHAPTER V

SUMMARY

This chapter is included to compare and contrast the results of the 2008 and 2009 trials. While these trials were very similar in the objectives and sources of animals and methodology, substantial differences between the trials were evident.

The summary statistic Tables 3.1 and 4.1 present some differences; the most obvious are the results for ADG between the 2 years. The mean for the 2008 trial was .38 kg/day as compared to the 2009 trial where the mean was 1.7 kg/day. This difference can be accounted for by the types of diets each set of steers was fed during the observation period; steers in 2008 were fed average quality bermudagrass hay whereas steers in 2009 were fed a mixed growing ration. Rectal temperature differences were also evident in terms of the maximum temperatures recorded. In the 2008 study a maximum value of $40.0^{\circ}C$ was observed and no animals were treated with an antimicrobial, whereas in 2009 the maximum temperature achieved was $41.3^{\circ}C$, and there were 35 of 93 (37%) treated with an antimicrobial. In both trials the source of cattle, vaccines administered, and virus exposure were similar, so no clear explanation exists for this difference in rectal temperature response between years. It is important to note that the diets, timing between vaccination and viral challenge, along with weather conditions between each trial were different, thus further investigation is needed to determine the effects these differences have on vaccine response in growing steers.

Serum IgG titer for IBR for cattle in the 2008 trial and 2009 trial were very similar for the killed vaccine group being the highest at day 0 and descending through

day 42. Cattle in the MLV vaccine group between the 2 years was less similar however, as the IBR titer from the 2009 trial seemed to remain at a constant level unlike the descending pattern observed in the 2008 trial for IBR titer.

Serum IgG titer for BVD Type 1 was very similar between the 2 years after day 0. Titer levels for BVD Type 1 from the Killed group was higher than the MLV and non-vaccinated groups except at day 0 of the 2009 trial where no differences existed between the MLV and Killed groups. In each trial, the non-vaccinated group displayed an ascending pattern with most of the increase occurring from day 14 to day 28. Less of an increase occurred from day 28 to 42 in both years. In both trials the only exposure to BVDV occurred at d 0 and since the vaccinated cattle were immunized, their titer response was higher and more rapid as compared to the non vaccinated steers.

Serum IgG titer for BVD type 2 in the Killed vaccine group was similar between the 2 years. In both years a large increase from day 0 to day 14 occurred and titer levels for the Killed vaccine group descended from day 14 to day 42. The MLV vaccine group between the 2 years displayed less similar patterns at day 0 where MLV group titer level for BVD type 2 was not different than the NON vaccine group in 2008 but was significantly different in 2009. Titer levels for the MLV group between the 2 trials were reflective in the ascending pattern through day 42. This similar ascending pattern was also observed in non-vaccinated steers.

Differences in the source of variation can also be observed in measurements that were taken in both trials. Sire accounted for more of the difference in rectal temperature in the 2009 trial as compared to the 2008 trial, and some of this difference may have

been due to the improved balance of sires across treatments in 2009. Vaccine treatment was a large source of variation for ADG in the 2008 trial but not in the 2009 trial, this may be due to an improved plane of nutrition for the 2009 trial since a complete mixed ration was fed.

Overall, this study shows that the relationships among health and performance traits are complex when cattle are exposed to BVD virus. The reduction of feed intake among the non-vaccinated cattle that showed no apparent increase in rectal temperature needs more research. There appears to be substantial genetic variation to select for IgG titers in response to BVDV, but whether or not this is the best genetic strategy to follow needs further investigation.

LITERATURE CITED

Abbas, A. K. and A. H. Lichtman. 2007. Cellular and Molecular Immunology. Elsevier Saunders Philadelphia, PA. 4-6.

Ahn, B.C., P.H. Walz, G.A. Kennedy, S. Kapil. 2005. Biotype, genotype, and clinical presentation associated with bovine viral diarrhea virus (BVDV) isolates from cattle. Intern. J. Appl. Res. Vet. Med. 3:319-325

Baker, J., C. York., J. Gillepsie, G. Mitchell. 1954. Virus diarrhea in cattle. Am. J. Vet. Res. 15:525-531.

Baker, J.C. 1995. The clinical manifestations of bovine viral diarrhea infection. Vet Clin North Am Food Anim Pract. 11:425-445.

Buhman, M.J., L.J. Perino, M.L. Gaylean, T.E. Wittum, T.H. Montgomery, and R.S. Swingle. 2000. Association between changes in eating and drinking behaviors and respiratory tract disease in newly arrived calves at a feedlot. Am. J. Vet. Res. 61:1163-1168.

Callan, R. J., and F. B. Garry. 2002. Biosecurity and bovine respiratory disease. Vet. Clin. North. Am. Food. Anim. Pract. 18:57-77.

Chase, C.L., D.J. Hurley, and A.J. Reber. 2008. Neonatal immune development in the calf and its impact on vaccine response. Vet. Clin. North. Am. Food. Anim. Pract. 24:87-104.

Chirase, N.K., and L.W. Greene. 2001. Dietary zinc and manganese sources administered from the fetal stage onwards affect immune response of transit stressed and virus infected offspring steer calves. Anim. Feed Sci. and Tech. 93:217-228.

Cornish, T.E., A.L. van Olphen, J.L. Cavendar, J.M. Edwards, P.T. Jaeger, L.L. Vieyra, L.F. Woodward, D.R. Miller, D. O'Toole. 2005. Comparison of ear notch immunohistochemistry, ear notch antigen-capture ELISA, and buffy coat virus isolation for detection of calves persistently infected with bovine viral diarrhea virus. J. Vet. Diagn. Invest. 17:110-117.

Cortese, V. S., R. Whittaker, and J. Ellis. 1998. Specificity and duration of neutralizing antibodies induced in healthy cattle after administration of a modified-live virus vaccine against bovine viral diarrhea. Am. J. Vet. Res. 59:848-850.

Daniels, T.K., J.G.P. Bowman, B.F. Sowell, M.E. Branine, and M.E. Hubbert. 2000. Effects of metaphylactic antibiotics on behavior of feedlot calves. Professional Animal Scientist. 16:247-253.

Duff, G.C. and M.L. Galyean. 2007. Recent advances in management of highly stressed, newly received feedlot cattle. J. Anim. Sci. 85:823-840.

Edwards, A.J. 1996. Respiratory diseases of feedlot cattle in the central USA. Bovine Pract. 30:5-7.

Edwards, S., L. Wood, C. Hewitt-Taylor, and T.W. Drew. 1986. Evidence for an immunocompromising effect of bovine pestivirus on bovid herpesvirus-1 vaccination. Vet. Res. Commun. 10:297-302.

Edwards, T.A. 2010. Control methods for bovine respiratory disease for feedlot cattle. Vet. Clin. North. Am. Food. Anim. Pract. 26:273-284.

Faries, F. C. Jr. 1999. Cattle Vaccines. bull. pub. No. L-5289. Texas Ag. Ext. Service. Texas A&M University, College Station, TX.

Frank, G. H., R. E. Briggs, G. C. Duff, and H. S. Hurd. 2003. Effect of intranasal exposure to eukotoxin-deficient *Mannheimia haemolytica* at the time of arrival at the feedyard on subsequent isolation of *M. haemolytica* from nasal secretions of calves. Am. J. Vet. Res. 64:580-585.

Fulton, R. W. 2007. Vaccines. Pages 209-222 in Bovine Viral Diarrhea Virus: Diagnosis, Management and Control. S. M. Goyal and J. F. Ridpath eds. Blackwell Publishing, Ames, IA.

Fulton, R. W., and L. J. Burge. 2000. Bovine viral diarrhea virus types 1 and 2 antibody response in calves receiving modified live virus or inactivated vaccines. Vaccine. 19:267-274.

Fulton, R. W., A. W. W. Confer, and L. J. Burge. 1995. Antibody responses by cattle after vaccination with commercial viral vaccines containing bovine herpesvirus-1, bovine viral diarrhea virus, parainfluenza-3 virus, and bovine respiratory syncytial virus immunogens and subsequent revaccination at day 140. Vaccine. 13:725-733.

Fulton, R.W., C.W. Purdy, A.W. Confer, J.T. Saliki, R.W. Loan, R.E. Briggs, L.J. Burge. 2000. Bovine viral diarrhea viral infections in feeder calves with respiratory disease: Interactions with *Pasteurella* spp*., parainfluenza-3 virus,* and bovine respiratory syncytial virus. Can. J. Vet. Res. 64:151-159.

Fulton, R.W., J.F. Ridpath, J.T. Saliki, R.E. Briggs, A.W. Confer, L.J. Burge, C.W. Purdy, R.W. Loan, F.C. Duff, and M.E. Payton. 2002. Bovine viral diarrhea virus (BVDV) 1b: predominant BVDV subtype in calves with respiratory disease. Can. J. Vet. Res. 66:181-190.

Fulton R. W., J. T. Saliki, L. J. Burge, and M. E. Payton. 2003a. Humoral immune response and assessment of vaccine virus shedding in calves receiving modified live virus vaccines containing bovine herpesvirus-1 and bovine viral diarrhea virus 1a. J. Vet. Med B 50:31.

Fulton, R.W., J.F. Ridpath, A.W. Confer, J.T. Saliki, L.J. Burge, and M.E. Payton. 2003b. Bovine viral diarrhea virus antigenic diversity: impact on disease and vaccination programmes. Biologicals. 31:89-95.

Fulton, F.W., R.E. Briggs, J.F. Ridpath, J.T. Saliki, A.W. Confer, M.E. Payton, G.D. Duff, D.L. Step, and D.A. Walker. 2005. Transmission of bovine viral diarrhea virus 1b to susceptible and vaccinated calves by exposure to persisitently infectd calves. Can. J. Vet. Res. 69:161-169.

Fulton, R.W., B. Hessman, B.J. Johnson, J.F. Ridpath, J.T.Saliki, L.J. Burge, D. Sjeklocha, A.W. Confer, R.A. Funk, and M.E. Payton. 2006. Evaluation of diagnostic tests used for detection of bovine viral diarrhea virus and prevalence of subtypes 1a, 1b, and 2a in persistently infected cattle entering a feedlot. J. Am. Vet. Med. Assoc. 228:578-584.

Fulton, R.W., J.F. Ridpath, B. Hessman, and K.S. Blood. 2009. Bovine viral diarrhea virus variability and prevalence of BVDV subtypes in persistently infected cattle entering feedlots: BVDV1b as predominant subtype. Proc. 4th U.S. BVDV Symposium, Jan. 25-27, Phoenix, AZ .

Galyean, ML., L. J. Perino, and G. C. Duff. 1999. Interaction of cattle health / immunity and nutrition. J. Anim. Sci. 77:1120-1134.

Gardner, B. A., H. G. Dolezal, L. K. Bryant, F. N. Owens, and R. A. Smith. 1999. Health of finishing steers: Effects on performance, carcass traits, and meat tenderness. J. Anim. Sci. 77:3168-3175.

Griffin, D. 1997. Economic impact associated with respiratory disease in beef cattle. Vet. Clin. North. Am. Food Anim. Pract. 13:367-377.

Grooms, D.L. 2004. Reproductive consequences of infection with bovine viral diarrhea virus. Vet. Clin. North. Am. Food. Anim. Pract. 20:5-19.

Grooms, D. L., K. V. Brock, and L. A. Ward. 1998. Detection of cytopathic bovine viral diarrhea virus in the ovaries of cattle following immunization with a modified live bovine viral diarrhea virus vaccine. J. Vet. Diagn. Invest. 10:130-140.

Kelling, C.L., D.J. Steffen, C.L. Topliff, K.M. Eskridge, R.O. Donis, D.S. Higuchi. 2002. Comparitive virulence of isolates of bovine viral diarrhea virus type II in

experimentally inoculated six to nine month old calves. Am. J. Vet. Res. 63(10):1379-1384.

Larson, R.L., V.L. Pierce, and D.M. Grotelueshen, and T.E. Wittum. 2002. Why control BVD: Economic and production costs, tools to accomplish. Available: http://www.ars.usda.gov/SP2UserFiles/Place/36253000/BVD2005/Produce1_Larson_Hout.pdf. Accessed June 10, 2008.

Liebler-Tenorio, E.M. J.F. Ridpath, J.D. Neill. 2003. Lesions and tissue distribution of viral antigen in severe acute versus subclinical acute infection with BVDV2. Biologicals. 31:119-122.

Loneragan, G.H., D.A. Dargats, P.S. Morley, and M.A. Smith. 2001. Trends in mortality ratios among cattle in US feed-lots. J. Am. Vet. Med. Assoc. 219:1122-1127.

Martin, S.W., A.H. Meek, D.G. Davis, J.A. Johnson, and R.A. Curtis. 1982. Factors associated with mortality and treatment costs in feedlot calves: The Bruce County beef project, years 1978, 1979, 1980. Can. J. Comp. Med. 46:341-349.

McNeill, J.W., J.C. Paschal, M.S. McNeill and W.W. Morgan. 1996. Effect of morbidity on performance and profitability of feedlot steers. J. Anim. Sci. 74(Suppl.1):135.

Meyling, A., H. House, and A.M. Jensen. 1990. Epidemiology of bovine virus diarrhea virus. Rev. Sci. Tech. Off. Int. Epiz. 9:75-93.

Muggli-Cockett, N.E., L.V. Cundiff, and K.E. Gregory. 1992. Genetic analysis of bovine respiratory disease in beef calves during the first year of life. J. Anim. Sci. 70:2013-2019.

Peterhans, E., T.W., Jungi, and M. Schweizer, 2003. BVD and innate immunity. Biologicals. 31:107-111.

Ridpath, J.F., J.D. Neill. 2007. Impact of variation in acute virulence of BVD1 strains on design of better vaccine efficacy challenge models. J. Vaccine. 47:8058-8066.

Roeber, D.L., N.C. Speer, J.G. Gentry, J.D. Tatum, C.D. Smith, J.C. Whittier, G.F. Jones, K.E. Belk, and G.C. Smith. 2001. Feeder cattle health management: Effects on morbidity rates, feedlot performance, carcass characteristics, and beef palatability. Prof. Anim. Sci. 17:39-44.

Roth, J. A., and L. M. Henderson. 2001. New technology for improved vaccine safety and efficacy. Vet. Clin. North. Am. Food Anim. Pract. 17(3)585-597.

Saliki, J.T., and Dubovi, E.J. 2004. Laboratory diagnosis of bovine viral diarrhead virus infections. Vet. Clin. North. Am. Food Anim. Pract. 20:69-83

Sandvik, T. 2005. Selection and use of laboratory diagnostic assays in BVD control programmes. Preventitive Veterinary Medicine 72:3-16.

Schneider, M.J., R.G. Tait Jr., W.D. Busby, and J.M, Reecy. 2009. An evaluation of bovine respiratory disease complex in feedlot cattle: impact on performance and carcass traits using treatment records and lung lesion scores. J. Anim. Sci. 87:1821-1827.

Smith, R.A. 1998. Impact of disease on feedlot performance: A review. J. Anim. Sci. 76:272-274.

Snowder G.D., L.D. Van Vleck, L.V. Cundiff and G.L. Bennett. 2005. Influence of breed, heterozygosity, and disease incidence on estimates of variance components of respiratory disease in preweaned beef calves. J. Anim. Sci. 83:1247–1261.

Snowder G.D., L.D. Van Vleck, L.V. Cundiff and G.L. Bennett. 2006. Bovine respiratory disease in feedlot cattle: Environmental, genetic, and economic factors. J. Anim. Sci. 84:1999-2008.

Warner, C.M., D.L. Meeker, and M.F. Rothschild. 1987. Genetic control of immune responsiveness: A review of its use as a tool for selection for disease resistance. J. Anim. Sci. 64:394-406.

Wilson, S.H. 1989. Why are meaningful field trials difficult to achieve for bovine respiratory disease vaccines? Can. Vet. J. 30:299-302.

Wittum, T.E., N.E. Woollen, L.J. Perino, and E.T. Littledike. 1996. Relationships among treatment for respiratory tract disease, pulmonary lesions evident at slaughter, and rate of weight gain in feedlot cattle. J. Am. Vet. Med. Assoc. 209:814-818.

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