PEER REVIEWED

Effects of delayed respiratory viral vaccine and/or inclusion of an immunostimulant on feedlot health, performance, and carcass merits of auction-market derived feeder heifers

K.C. Rogers,[1] DVM, MS; **D.G. Miles,**[1] DVM, MS; **D.G. Renter,**[2] DVM, PhD; **J.E. Sears,**[3] DVM; **J.L. Woodruff,**[4] DVM
[1]Veterinary Research and Consulting Services, LLC, Greeley, CO 80634
[2]Center for Outcomes Research and Education, Kansas State University, Manhattan, KS 66506
[3]Bayer Animal Health, Shawnee Mission, KS 66216
[4]Boehringer Ingelheim Vetmedica, Inc., St. Joseph, MO 64506
Corresponding author: Dr. K.C. Rogers, vrcskcr@aol.com

Abstract

A total of 5,179 high-risk heifer calves were used to define the potential benefits of delaying the on-arrival respiratory viral vaccine (Pyramid® 5, Boehringer Ingelheim Vetmedica, Inc., St. Joseph, MO) for 30 d with and without the addition of a DNA immunostimulant (Zelnate® DNA Immunostimulant, Bayer Animal Health, Shawnee Mission, KS) at feedlot entry on health, performance, and carcass characteristics. The products were evaluated in a 2 x 2 factorial, randomized complete-block design comparing the following: delayed vaccine (DP), on-arrival vaccine (AP), delayed vaccine plus immunostimulant (DPZ), and on-arrival vaccine plus immunostimulant (APZ) amongst 60 total pens of heifers (15 pens/treatment). Pen-level linear mixed models, including a random effect for allocation block (source), were used for all statistical analyses. There was no P x Z interaction ($P \geq 0.35$) for any outcome. At 60 DOF, reimplant (116 d), and close-out, delaying the viral vaccine decreased ($P \leq 0.05$) the percent of calves treated twice for bovine respiratory disease (BRD). The inclusion of immunostimulant reduced ($P \leq 0.05$) BRD mortality and overall mortality at both 60 and 116 DOF. Additionally, the reduction ($P = 0.04$) in overall mortality and the tendency to lessen BRD mortality ($P = 0.06$) was maintained through close-out for cattle administered the immunostimulant. No differences in final gain performance, dry matter intake or feed conversion were observed among treatments. There was a tendency for heifers to have heavier finished body weights ($P = 0.08$) and HCW ($P = 0.07$) when vaccinated on arrival. No differences in carcass quality or yield grade categories were evident. In conclusion, delaying the viral vaccine and including the immunostimulant both appeared to improve cattle health by significantly reducing BRD retreatment risk and overall mortality, respectively.

Key words: bovine, respiratory, BRD morbidity, DNA immunostimulant, delayed viral vaccination

Résumé

On a utilisé un total de 5 179 génisses à haut risque pour déterminer les bénéfices potentiels de reporter de 30 jours le vaccin contre les infections virales respiratoires administré à l'arrivée (Pyramid® 5, Boehringer Ingelheim Vetmedica, Inc., St. Joseph, MO) avec ou sans l'ajout d'un immunostimulant à base d'ADN (Zelnate® DNA Immunostimulant, Bayer Animal Health, Shawnee Mission, KS) lors de l'entré au parc d'engraissement sur la santé, la performance et les caractéristiques de la carcasse. Ces produits ont été utilisés dans un plan d'expérience factoriel 2 sur 2 avec blocs aléatoires complets afin de comparer les groupes suivants : vaccin reporté, vaccin à l'arrivée, vaccin reporté avec immunostimulant et vaccin à l'arrivée avec immunostimulant. Il y avait un total de 60 enclos de génisses et 15 enclos par traitement. On a utilisé des modèles linéaires mixtes au niveau de l'enclos incluant le bloc d'allocation (source) comme effet aléatoire pour toutes les analyses statistiques. Il n'y avait pas d'interaction entre le vaccin et l'immunostimulant ($P \geq 0.35$) pour toutes les mesures. Après 60 d'engraissement, à la réimplantation (116 jours) et à la fin de l'engraissement, le pourcentage de veaux traités à deux reprises pour le complexe respiratoire bovin (CRB) était moindre avec le vaccin reporté ($P \leq 0.05$). L'ajout de l'immunostimulant a réduit la mortalité associée au CRB ($P \leq 0.05$) et la mortalité globale après 60 et 116 jours d'engraissement. De plus, la réduction ($P = 0.04$) de la mortalité globale et la tendance à la réduction de la mortalité associée au CRB ($P = 0.06$) se sont maintenues jusqu'à la fin de la période d'engraissement chez les veaux qui avaient reçu l'immunostimulant. Il n'y a pas eu de différence entre

les traitements pour le gain final de performance, la prise de matière sèche ou la conversion alimentaire. Le poids à la finition ($P = 0.08$) et le poids de la carcasse chaude ($P = 0.07$) étaient marginalement plus élevés chez les génisses vaccinées à l'arrivée. Il n'y avait pas de différence évidente entre les traitements en ce qui concerne la qualité de la carcasse et la catégorie de rendement. En conclusion, reporter le vaccin viral et l'inclusion d'un immunostimulant ont tous les deux un impact positif sur la santé des bovins en réduisant le risque de retraitement pour le CRB et en diminuant la mortalité globale, respectivement.

Introduction

The economic impact of bovine respiratory disease (BRD) is substantial and spans markedly beyond observable death loss. Indirect costs of treatment and prevention, loss of production, and reduced carcass value must also be taken into consideration to fully realize the expanse of this multifactorial complex. Rarely does BRD stem from a single pathogen, but rather it has a diverse origin that relies on the interaction between host susceptibility, pathogens, and the environment. *Mannheimia haemolytica* serotype A1 has been identified as the most common bacterial pathogen in BRD during the receiving period,[1,5,8,9,13,26] with prevalence ranging from 65 to 75%.[5] Moreover, fibrinous pneumonia ascribed to *M. haemolytica* is the most important cause of BRD mortality.[4] A commensal organism of the nasopharynx and tonsillar crypts, *M. haemolytica* is an opportunist, gaining access to the lungs when host defenses are compromised by stress and/or viral-induced immune dysfunction.[19] Bovine respiratory disease is definitively one of the best known examples of stress-associated infectious disease.[12]

Many of the physiologic stress effects incurred by feedlot placements (weaning,[6,10] handling,[6] commingling,[22] acute pain,[28] castration/dehorning,[15] and transportation[6,10,15,27]) are known to cause increased plasma cortisol concentrations, which in turn can compromise the immune system. The transportation stress period alone has been demonstrated to endure for as long as 15 d post-arrival based on serum hemolytic complement concentration.[18] Prevention of disease by vaccination is the foundation of animal health management; however, stress and previous exposure to BRD pathogens may decrease vaccine efficacy. All manufacturers of modified-live virus (MLV) vaccines recommend that the vaccine be given prior to risk of disease and only to healthy cattle, as would align with many preconditioning programs.[1,11] However, present day marketing channels and cattle incubating disease prior to sale may preclude this from being achievable. Nearly all cattle are vaccinated with bovine viral diarrhea virus (BVDV; 95.1%) and infectious bovine rhinotracheitis virus (IBRV; 93.2%) antigens upon arrival in US feedlots.[30] Whether vaccination on arrival is due to tradition, convenience or merely uncertainty in prior history, it is contrary to label recommendations and attests our timing as an industry may be inappropriate. Thus, delaying respiratory vaccination may reduce taxation of the immune system and improve vaccine efficacy because high-risk cattle are allowed to overcome stress-induced immune dysfunction and prevent a blunted vaccine response.[22]

Strategic measures to prevent or mitigate BRD over the last several decades have been met with disappointing results as it continues to be the most common cause of feedlot death loss.[14] As an industry, we will be implored by consumers to reduce antimicrobial use in the production of wholesome beef. Other means of intervention to positively impact health will need to be devised.

Brand selection of the MLV vaccine for this study was based on previous work documenting beneficial health and economic outcomes when using this product.[3] Confidence in the vaccine enabled the focus of the study to center on the effects of timing rather than efficacy. However, it would be worthwhile to replicate this study using other MLV vaccine brands to examine if the effects of delaying vaccination are repeatable. Thus, the objectives of this study were to evaluate the potential benefits of delaying the MLV vaccine until stress and *M. haemolytica* challenges had declined, and also examine if the addition of a DNA immunostimulant would improve health and performance outcomes in a large commercial feedlot setting.

Materials and Methods

Cattle

A total of 5,179 heifer calves were allocated to 15 blocks of 4 treatments within each block (60 pens total), to compare the effects of delayed MLV pentavalent viral administration, with and without the inclusion of a DNA immunostimulant, on health, performance, and carcass merits of lightweight, auction-derived feeder calves. Crossbred heifers of English, Brahman, and exotic origin were procured from livestock markets in Oklahoma and Texas and delivered to a commercial feedlot from August 24, 2015 to October 16, 2015. Calves were unloaded and penned by source within a truckload upon arrival, and those that were sick, injured, or males were placed in separate pens and excluded from study. Prior to weighing cattle at initiation of the study, reimplant, and shipping, the load scale was certified, as is standard for scales utilized for commerce. Cattle remained separated by source and were randomized as they entered the chute. Treatment group sequence was established by drawing numbers (1, 2, 3, and 4) from a hat to determine order for test article administration. The first number drawn was the treatment group assigned for the first animal in the chute, the second number was the treatment group assigned for the second animal in the chute, and so on. Calves of common origin/purchase followed the same randomization scheme within a block until pens were filled to capacity to ensure that cattle within arrival block were equally represented in each treatment group. There was a new drawing for each block to decide processing order. As

a result, calves were nearly equally represented in all groups dependent on procured headcount and number of long-bred pregnant heifers removed from study consideration. Cattle in different treatment groups were fed in different but adjacent, study pens for each respective block. Pen square footage and bunk space allowance were equivalent within a given study block. Pens housed approximately 86 calves/pen, and average processing pen weight was 624 lb (284 kg) (range 602 to 650 lb; 274 to 295 kg).

Processing

Upon arrival, cattle were placed in pens according to origin/truckload and provided *ad libitum* access to prairie grass hay and water, prior to processing within 72 hours of arrival. Routine processing procedures for all heifers included the following:

- Serially-numbered lot ear tag
- *Mannheimia haemolytica* toxoid[a] (2 mL) administered SC in left neck
- Tilmicosin[b] (2 mL/100 lb (45.5 kg) of body weight) administered SC in the right neck to all replicates due to high-risk status of calves
- Moxidectin[c] (1 mL/110 lb (50 kg) of body weight) administered SC in the right neck
- Oxfendazole[d] (0.9 mL/100 lb (45.5 kg) of body weight) administered orally
- Determination of pregnancy status .

Trial vaccine and/or immunostimulant were administered to calves randomized to 1 of 4 experimental groups: DP heifers received vaccine at 30 d; AP received vaccine on-arrival and 30 d; DPZ received immunostimulant on-arrival and vaccine at 30 d; and APZ received vaccine and immunostimulant on-arrival and vaccine at 30 d. Test articles evaluated and location administered are as follows:

- Modified-live IBRV, parainfluenza-3 virus (PI_3V), BVDV types 1 and 2, and bovine respiratory syncytial virus (BRSV) vaccine[e] (2 mL) administered SC in the right neck
- DNA immunostimulant[f] (2 mL) administered IM in the left neck.

All heifers, with the exception of Replicate 6 (manual palpation), were examined in the hydraulic chute by ultrasound to determine pregnancy status. A total of 429 short-bred heifers (less than 90 days in gestation) were confirmed pregnant and administered a prostaglandin[g] (5 mL, right neck) at the time of processing. Heifers bred longer than 90 days in gestation were not considered eligible for the trial. Cattle were weighed in drafts on a livestock ground scale by treatment after processing to establish a starting pen weight. At 30 ± 5 days on feed (DOF), all calves received their initial growth promoting implant[h], were administered viral vaccine[e], and pen weights were collected. A terminal implant[i] was administered at approximately 116 DOF (range 104 to 127 DOF). No subsequent revaccination with viral respiratory products was performed either in the hospital as adjunct therapy or at time of reimplant.

Treatment assignment

Vaccine treatment and immunostimulant administration were determined as cattle entered the chute, with each animal receiving the appropriate test article based upon the number sequence established by blindly drawing from a hat prior to processing. The same treatment sequence was used for all sources within a replicate. Truckloads were kept together to ensure that cattle in each replicate were of similar background, age, and weight. A total of 15 pen replicates were included in the study, averaging 86 calves/pen (range 79 to 98). Study pens were 38,500 sq ft (3575 m^2) consisting of dirt floors with 140 feet (43 m) of linear concrete bunk space and fence-line overflow water tanks.

BRD case definition

Pen riders and treatment personnel were blinded to experimental treatment, and cattle exhibiting clinical signs of disease were removed from their home pen and evaluated. Standard feedlot protocol specified that cattle must have a rectal temperature of ≥ 104 °F (40 °C), and any 1 of the following clinical signs of BRD, including depression, lowered head carriage, nasal and/or ocular discharge, coughing, stiff gait, or depressed ruminal fossa, to qualify for treatment for respiratory disease. In rare instances, at the discretion of treatment personnel, cattle deemed to be in advanced stages of BRD (late pull, as defined by extreme depression, sunken eyes, crusted nose, and/or yellow diarrhea) were treated with an antimicrobial regardless of rectal temperature for humane reasons. The same treatment regimen was utilized for all cattle taken to the hospital that qualified for therapy. Treated cattle were generally returned to their home pen; however, those that were treated 3 or more times were allowed to recover in hospital pens or railed (culled), if deemed necessary. Feed consumed for animals retained in the hospital was prorated back to the appropriate home pen prior to data analysis. Health records for all treated cattle were maintained throughout the trial. Cattle not considered capable of reaching market weight in the same amount of time as their pen mates due to illness (i.e., chronic respiratory disease, lameness, or failure to thrive due to an undiagnosed condition) were removed from the pen and marketed via alternate channels. These animals were not included in the final growth performance analysis for deads-out gain determination. Dead cattle were necropsied by either a veterinarian or trained feedlot personnel.

Feed

Cattle were fed 3 times daily. Diets were consistent across treatments and replicates and consisted of steam-flaked corn, high moisture corn, dry distillers grain, corn silage, alfalfa hay, corn stalks, and liquid supplement. Monen-

sin[j] and tylosin[k] were fed for the entire feeding period. At approximately 35 DOF, melengestrol acetate[l] was incorporated into the third step-up ration. Ractopamine[m] was included in the ration when heifers were within 30 days of harvest.

Marketing

Heifers were harvested when they were visually estimated to have adequate finish for market; DOF ranged from 196 to 221 (average 209). A total of 338 heifers either died or were marketed through alternate channels prior to the end of the study. All heifers from a given replicate were harvested on the same day, with 6 harvest dates. Carcass data were provided by lot rather than individual ID, as cattle were sold on the grid. Heifers were shipped to a packing plant in Liberal, Kansas, from March 11 to April 29, 2016, and carcass data were collected for all cattle.

Statistical analyses

The analyses were dictated by the study design: a 2 x 2 factorial treatment structure (with viral vaccine on-arrival or delayed, and with and without immunostimulant upon feedlot entry) in a randomized complete block design with 15 blocks (replicates) and pen (within block) as the experimental unit. General and generalized linear mixed models, for continuous and categorical outcomes respectively, were used for all analyses.[n] In all models, fixed effects included the vaccine x immunostimulant (P x Z) interaction and both main effects of vaccine timing (P) and immunostimulant (Z), and a random effect (intercept) for block was included to account for the design structure (lack of independence among pens within blocks). A P value of ≤ 0.05 was considered statistically significant. When the vaccine x immunostimulant (P x Z) interaction was not significant, model-adjusted means for main effects were compared. Model-adjusted means for main effects or for individual treatment groups, and standard errors of those means, are reported (back transformed to the original scale for generalized models).

Results and Discussion

There were a total of 429 short-bred (< 90 days gestation) heifers allocated to 60 pens in 15 replicates with no difference in the number of pregnant heifers among the four treatment groups ($P = 0.72$). There was also no difference ($P = 0.17$) in the mean allocation body weight among treatments: 622, 625, 625, and 625 lb, (283, 284, 284, and 284 kg) for DP, AP, DPZ, APZ, respectively, indicating consistent dispersion of initial body weight across treatment groups. With no evidence for a significant interaction (P x Z) between treatments for any of the health, performance or carcass variables analyzed (i.e. the effects of vaccine and immunostimulant at arrival were not affected by each other), only the main effects of viral vaccine and immunostimulant are discussed.

Health outcomes at 60 DOF by treatment group are presented in Table 1. Typically most cases of respiratory disease and fatality are experienced in calves within the first

Table 1. Health performance of feedlot heifers (60 d) for the effects of vaccination timing and immunostimulant inclusion (model-adjusted means, (SEM)).

	Experimental group*						
Item	**DP†**	**AP**	**DPZ‡**	**APZ**	***P*-value P§**	***P*-value Z‖**	***P*-value P x Z¶**
No. calves (pens)	1,296 (15)	1,290 (15)	1,293 (15)	1,300 (15)	-	-	-
BRD 1 treatment, %	22.92 (1.82)	23.17 (1.83)	22.92 (1.82)	21.78 (1.76)	0.70	0.55	0.55
BRD 2 treatments, %	8.36 (1.05)	9.39 (1.14)	7.49 (0.98)	9.67 (1.16)	0.04	0.66	0.44
BRD 3 treatments, %	4.34 (0.68)	4.98 (0.74)	3.53 (0.60)	3.95 (0.64)	0.35	0.10	0.92
BRD case fatality, %	11.88 (1.93)	14.03 (2.07)	8.61 (1.66)	11.63 (1.95)	0.16	0.12	0.70
BRD mortality, %	2.85 (0.56)	3.45 (0.64)	1.98 (0.45)	2.54 (0.52)	0.19	0.05	0.86
Overall mortality, %	3.26 (0.60)	3.79 (0.66)	2.15 (0.46)	2.79 (0.54)	0.20	0.03	0.74

*DP, AP, DPZ, APZ = delayed vaccine, arrival vaccine, delayed vaccine plus DNA immunostimulant, and arrival vaccine plus DNA immunostimulant, respectively

†P = Pyramid® 5, Boehringer Ingelheim Vetmedica, Inc., St. Joseph, MO

‡Z = Zelnate® DNA Immunostimulant, Bayer Animal Health, Shawnee Mission, KS

§*P*-value for the main effect of vaccine timing

‖*P*-value for the main effect of arrival inclusion of DNA immunostimulant

¶*P*-value for the interaction of vaccine and DNA immunostimulant

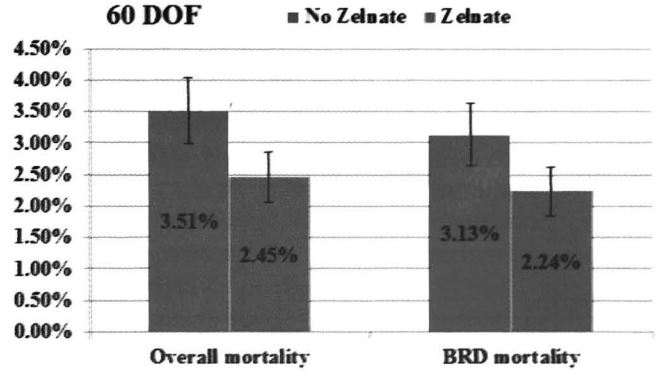
2 months of feedlot arrival; hence the selected interim data time point. There was no effect of vaccination ($P = 0.70$) or immunostimulant ($P = 0.55$) for percentage of calves treated once for respiratory disease; however, delaying MLV vaccination resulted in a decrease ($P = 0.04$) in the percent of calves treated twice for BRD. Richeson et al[23] compared the effects of vaccine administration (unvaccinated, arrival, and delayed 14 d) on high-risk stocker calves and did not observe any differences in health parameters due to timing; however, there was a tendency for more of unvaccinated calves to require 2 antimicrobial treatments. Also, the relapse rate was numerically increased for the on-arrival versus delayed treatment in this study (30.1 vs 23.3%; $P = 0.51$). Other studies[20,21] reported similar morbidity when calves were administered the initial MLV respiratory vaccine either on day 0 (arrival) or 14. Likewise, Duff et al[7] compared the effects of vaccine administration (intranasal vs intramuscular vs unvaccinated control) in newly received beef calves and observed no differences in morbidity. It is widely accepted that stress reduces the immune response to vaccination in cattle[26] yet distinct immunological consequences are attributed to acute vs chronic stress.[22] It is thought acute stress typically occurs for well-handled calves on a ranch during routine vaccination procedures, whereas chronic stress is encountered more in high-risk calves during initial processing at stocker or feedlot facilities.[22] It is plausible that vaccine response is enhanced for cattle encountering acute stress due to immunopriming effects of short-term stress, while chronically stressed cattle will exhibit a blunted vaccine response due to the immunosuppressive effects of chronic stress.[22] The nature of the vaccine antigen (replicating MLV) also deserves special consideration in determining vaccine efficacy/response on the ultimate antibody production in chronically stressed cattle. Richeson et al[24] demonstrated increased replication of MLV agents, antigenicity, and nasal shedding in beef steers challenged with immunosuppressive levels of dexamethasone that were also administered a pentavalent MLV vaccine. Thus, excessive replication of MLV agents in naturally stressed calves administered MLV vaccine on-arrival might translate to a greater display of clinical signs of BRD and subsequent pulls/treatments.

The indication for administering the DNA immunostimulant on arrival was to activate the innate immune system to fight BRD pathogens, especially *M. haemolytica*, at the time of stress and disease challenge. There was no statistical difference among treatments in the percentage of calves treated once for BRD that received immunostimulant vs those that did not, but there tended ($P = 0.10$) to be a reduction in third treatments (Table 1). The inclusion of immunostimulant reduced percentage BRD mortality ($P = 0.05$) and overall mortality ($P = 0.03$) at 60 d (Figure 1). Thus, the reduction in mortality and tendency for percent case fatality risk (Table 2; $P = 0.07$) to be lower in heifers receiving the immunostimulant suggests the product positively influenced the outcome (survivability) of those calves that were treated for respiratory disease.

Table 2 demonstrates health outcomes by treatment group at 116 d (the mean reimplant DOF). The main effect of delaying vaccination was associated with a reduction in both the percent of calves treated twice (data not shown) for BRD ($P = 0.04$) and the overall retreatment risk ($P = 0.01$). Inclusion of immunostimulant was again associated with a decrease in BRD ($P = 0.04$) and overall mortality ($P < 0.01$).

Health outcomes at close-out are presented in Table 3, in addition to Figures 2 and 3. In total 1,335 (25.8 %) of the heifers were treated for BRD with 547 (10.6%) being treated more than once. Additionally, 270 heifers died (5.2%) prior to study completion with 190 (3.7%) attributed to respiratory disease. Deaths due to acute interstitial pneumonia (n = 18) were minor, distributed nearly evenly, and tabulated as non-BRD mortalities. Heifers receiving the delayed MLV vaccine required less therapy as evidenced by significant differences in the number being treated twice for BRD and the mean retreatment risk. Across all pens, mean retreatment risk was 37.05% for calves receiving viral vaccine at 30 DOF (delayed administration) vs 43.97% for calves administered vaccine on arrival (Figure 2). Martin et al[16] reported an increased incidence of mortality when a respiratory vaccine was administered within 14 d of arrival in calves fed a silage-based diet, but no difference in mortality due to timing of vaccine when calves were fed a hay-based diet. The incidence of chronics in the present study did not differ based on either effects of vaccination ($P = 0.78$) or immunostimulant ($P = 0.39$). Overall mortality was less for cattle receiving the immunostimulant ($P = 0.04$), and there was a tendency for BRD mortality ($P = 0.06$) and case fatality risks ($P = 0.10$) to follow the same

Figure 1. Model-adjusted means (and standard errors of the means) for overall mortality and BRD mortality at 60 days on feed, demonstrating the significant reductions due to inclusion of the DNA immunostimulant.*



*Zelnate® DNA Immunostimulant, Bayer Animal Health, Shawnee Mission, KS
Overall mortality, $P = 0.03$
BRD mortality, $P = 0.05$

Table 2. Health performance of feedlot heifers (116 d) for the effects of vaccination timing and immunostimulant inclusion (model-adjusted means, (SEM)).

	Experimental group*						
Item	**DP†**	**AP**	**DPZ‡**	**APZ**	***P*-value P§**	***P*-value Z‖**	***P*-value P x Z¶**
BRD 1 treatment, %	24.83 (1.95)	24.76 (1.95)	24.43 (1.94)	23.82 (1.91)	0.78	0.58	0.82
BRD retreatment,# %	39.48 (3.17)	43.95 (3.24)	37.06 (3.15)	46.78 (3.31)	0.01	0.96	0.35
BRD case fatality, %	13.78 (1.97)	16.24 (2.13)	9.97 (1.72)	13.19 (1.97)	0.13	0.07	0.71
BRD mortality, %	3.54 (0.63)	4.22 (0.71)	2.44 (0.50)	3.22 (0.59)	0.13	0.04	0.74
Overall mortality, %	4.30 (0.71)	4.99 (0.78)	2.67 (0.52)	3.75 (0.64)	0.08	< 0.01	0.50

*DP, AP, DPZ, APZ = delayed vaccine, arrival vaccine, delayed vaccine plus DNA immunostimulant, and arrival vaccine plus DNA immunostimulant, respectively

†P = Pyramid® 5, Boehringer Ingelheim Vetmedica, Inc., St. Joseph, MO

‡Z = Zelnate® DNA Immunostimulant, Bayer Animal Health, Shawnee Mission, KS

§*P*-value for the main effect of vaccine timing

‖*P*-value for the main effect of arrival inclusion of DNA immunostimulant

¶*P*-value for the interaction of vaccine and DNA immunostimulant

#Retreatment risk for each pen was calculated as the percentage of calves first treated for BRD that were subsequently treated again for BRD.

Table 3. Health performance of feedlot heifers at close-out for the effects of vaccination timing and immunostimulant inclusion (model-adjusted means, (SEM)).

	Experimental group*						
Item	**DP†**	**AP**	**DPZ‡**	**APZ**	***P*-value P§**	***P*-value Z‖**	***P*-value P x Z¶**
BRD 1 treatment, %	26.03 (2.09)	25.50 (2.07)	25.18 (2.05)	25.16 (2.05)	0.82	0.63	0.83
BRD 2 treatments, %	9.64 (1.21)	11.05 (1.33)	8.93 (1.15)	11.01 (1.32)	0.04	0.63	0.66
BRD 3 treatments, %	5.53 (0.79)	5.79 (0.82)	4.35 (0.68)	5.50 (0.79)	0.24	0.22	0.42
BRD re-treatment risk,# %	37.95 (3.09)	43.59 (3.20)	36.17 (3.09)	44.35 (3.22)	0.01	0.84	0.64
BRD case fatality, %	13.44 (1.90)	16.08 (2.08)	10.28 (1.71)	13.11 (1.90)	0.14	0.10	0.85
BRD mortality, %	3.61 (0.64)	4.36 (0.73)	2.65 (0.53)	3.36 (0.61)	0.15	0.06	0.88
Overall mortality, %	5.35 (0.80)	5.88 (0.86)	3.79 (0.64)	5.02 (0.77)	0.13	0.04	0.45
BRD outs (deads + removals), %	4.17 (0.78)	4.98 (0.89)	3.47 (0.68)	4.21 (0.79)	0.16	0.18	0.95
Total outs (deads + removals), %	6.34 (0.96)	6.88 (0.10)	5.26 (0.84)	6.16 (0.94)	0.27	0.17	0.73

*DP, AP, DPZ, APZ = delayed vaccine, arrival vaccine, delayed vaccine plus DNA immunostimulant, and arrival vaccine plus DNA immunostimulant, respectively

†P=Pyramid® 5, Boehringer Ingelheim Vetmedica, Inc., St. Joseph, MO

‡Z=Zelnate® DNA Immunostimulant, Bayer Animal Health, Shawnee Mission, KS

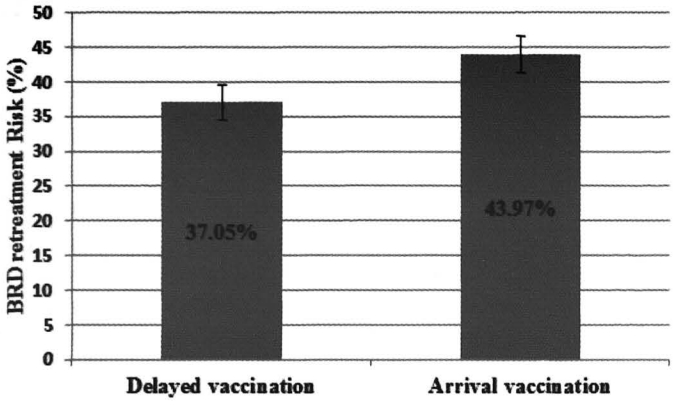
§P-value for the main effect of vaccine timing

‖P-value for the main effect of arrival inclusion of DNA immunostimulant

¶P-value for the interaction of vaccine and DNA immunostimulant

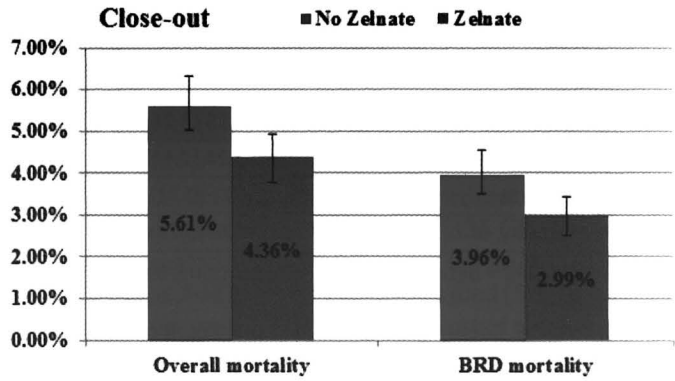
#Retreatment risk for each pen was calculated as the percentage of calves first treated for BRD that were subsequently treated again for BRD.

Figure 2. Model-adjusted means (and standard errors of the means) for BRD retreatment risk at close-out, demonstrating the statistically significant reductions due to delaying MLV vaccine* administration (P=0.01).



*Pyramid® 5, Boehringer Ingelheim Vetmedica, Inc., St. Joseph, MO

Figure 3. Model-adjusted means (and standard errors of the means) for overall mortality and BRD mortality at close-out demonstrating the significant reductions due to inclusion of the DNA immunostimulant.*



*Zelnate® DNA Immunostimulant, Bayer Animal Health, Shawnee Mission, KS

Overall mortality, P = 0.04

BRD mortality, P = 0.06

Table 4. Live performance of feedlot heifers at close-out for the effects of vaccination timing and immunostimulant inclusion (model-adjusted means, (SEM)).

	Experimental group*						
Item	**DP†**	**AP**	**DPZ‡**	**APZ**	***P*-value P§**	***P*-value Z‖**	***P*-value P x Z¶**
Final body weight, lb	1234 (7.94)	1242 (7.94)	1235 (7.94)	1243 (7.94)	0.08	0.85	0.95
HCW, lb	794 (6.04)	799 (6.04)	794 (6.04)	802 (6.04)	0.07	0.62	0.74
ADG, deads in#	2.67 (0.06)	2.68 (0.06)	2.72 (0.06)	2.72 (0.06)	0.96	0.30	0.82
ADG, deads out**	2.92 (0.03)	2.95 (0.03)	2.92 (0.03)	2.95 (0.03)	0.16	0.90	0.83
DMI, lb	17.63 (0.19)	17.84 (0.19)	17.58 (0.19)	17.87 (0.19)	0.12	0.96	0.82
F:G‡‡ (deads in)	6.64 (0.11)	6.69 (0.11)	6.48 (0.11)	6.60 (0.11)	0.33	0.12	0.64

*DP, AP, DPZ, APZ = delayed vaccine, arrival vaccine, delayed vaccine plus DNA immunostimulant, and arrival vaccine plus DNA immunostimulant, respectively

†P = Pyramid®5, Boehringer Ingelheim Vetmedica, Inc., St. Joseph, MO

‡Z = Zelnate® DNA Immunostimulant, Bayer Animal Health, Shawnee Mission, KS

§*P*-value for the main effect of vaccine timing

‖*P*-value for the main effect of arrival inclusion of DNA immunostimulant

¶*P*-value for the interaction of vaccine and DNA immunostimulant

#Gain is total net slaughter weight plus total weight of animals shipped as railers for salvage minus total initial weight (processing pen weight) divided by the total number of animal days (slaughter, railer and dead cattle DOF).

**Gain is average slaughter weight minus average initial weight divided by average days on feed.

‡‡Based on unshrunk initial weights and 4% shrunk final weights

trend. Across all pens, means for overall mortality and BRD mortality are 4.36% vs 5.61% and 2.99% vs 3.96% for calves receiving the immunostimulant compared to those that did not, respectively (Figure 3).

Live performance at close-out is depicted in Table 4. Cattle (pens) were fed for an average of 209 days in the study, having an overall mean out-weight of 1238 lb (563 kg). No differences in final gain performance, dry matter intake or feed conversion were observed among treatments. However, there was a tendency (P = 0.08) for heifers that finished the study to have heavier finished body weights in the arrival vaccination groups compared to those that were delayed.

Immunological challenges from antigens contained in a vaccine could divert an animal's resources towards mounting a protective response to the vaccine at the expense of growth, but this was not validated in the present study. Likewise, Stokka and Edwards[29] reported no detrimental effects on gain of stressed calves receiving a polyvalent MLV vaccine on arrival. Other vaccination timing studies differ in performance results, possibly due to disease stage at receiving, degree of challenge during the marketing process or perhaps even pen dynamics. Richeson et al[23] reported greater ADG for calves administered delayed (14 d) respiratory vaccination; however, Richeson et al[21] and Poe et al[17] reported no difference in vaccination timing (d 0 or 14) on gain performance during a 42-d receiving period.

Data were collected on 4,834 total carcasses, with an overall mean hot weight of 797 lb (362 kg) and a mean dressing percentage of 64.32%. There was no evidence of treatment effects on dressing percentage; however, HCW tended ($P = 0.07$) to be heavier for cattle receiving vaccine at arrival (Table 4). There was no evidence that the distribution of carcasses in the quality and yield categories were different (data not shown; all P values ≥ 0.15).

Conclusion

In this study, delaying the administration of MLV vaccine for 30 d resulted in a significant decrease in the number of calves requiring additional treatment for BRD, suggesting that the delay may have provided the opportunity for treated cattle to respond more favorably because the interaction between stress and MLV antigens was not a factor in convalescence or clinical display for the delayed vaccinate cattle. Additionally, the inclusion of a DNA immunostimulant consistently improved survivability as evidenced by a significant reduction in total mortality at 60 d, 116 d, and close out, resulting in a 22% reduction in overall death loss.

Endnotes

[a]Presponse® SQ, Boehringer Ingelheim Vetmedica, Inc., St. Joseph, MO
[b]Micotil®, Elanco Animal Health, Greenfield, IN
[c]Cydectin®, Boehringer Ingelheim Vetmedica, Inc., St. Joseph MO
[d]Synanthic®, Boehringer Ingelheim Vetmedica, Inc., St. Joseph, MO
[e]Pyramid® 5, Boehringer Ingelheim Vetmedica, Inc., St. Joseph, MO
[f]Zelnate® DNA Immunostimulant, Bayer Animal Health, Shawnee Mission, KS
[g]Lutalyse®, Zoetis Animal Health, Florham Park, NJ
[h]Component® with Tylan® TE-IH, Elanco Animal Health, Greenfield, IN
[i]Component® with Tylan® TE 200, Elanco Animal Health, Greenfield, IN
[j]Rumensin®, Elanco Animal Health, Greenfield, IN
[k]Tylan®, Elanco Animal Health, Greenfield, IN
[l]HeifermaX, Elanco Animal Health, Greenfield, IN
[m]Optaflexx®, Elanco Animal Health, Greenfield, IN
[n]SAS Glimmix, SAS Institute Inc., Software Version 9.3, Cary, NC

Acknowledgement

We especially wish to thank Shelly Kirby, Mike Veltri, Michael Persinger, and staff of Supreme Cattle Feeders, Kismet, KS for their assistance and dedication in conducting this trial. Funding for this study was provided by Bayer Animal Health and Boehringer Ingelheim Vetmedica, Inc.; however, no employees of the sponsors were directly involved in the conduct of the study.

References

1. Bayley A. Canadian Compendium of Veterinary Pharmaceuticals, Biologicals and Specialists. 1st ed. Hensall, Ontario: CCVPBS 774569 Ontario Ltd., 1989.
2. Booker CW, Abutarbush SM, Morley PS, Jim GK, Pittman TJ, Schunicht OC, Perrett T, Wildman BK, Fenton RK, Guichon PT, Janzen ED. Microbiological and histopathological findings in cases of fatal bovine respiratory disease of feedlot cattle in Western Canada. *Can Vet J* 2008; 49:473-481.
3. Bryant TC, Rogers KC, Stone ND, Miles DG. Effect of viral respiratory vaccine treatment on performance, health, and carcass traits of auction-origin feeder steers. *Bov Pract* 2008; 42:98-103.
4. Cavirani S, Taddei S, Cabassi CS, Ghidini F, Piancastelli C, Flammini CF. Antibody response to *Mannheimia haemolytica* leukotoxin in cattle with respiratory tract disease. *Open Vet Sci J* 2007; 1:7-10.
5. Confer AW, Clarke CR. Pasteurellosis in cattle. In: Howard JL, Smith RA, eds. *Current Veterinary Therapy 4. Food Animal Practice.* Philadelphia: WB Saunders, 1999; 369-372.
6. Crookshank HR, Elissalde MH, White RG, Clanton DC, Smalley HE. Effect of transportation and handling of calves upon blood serum composition. *J Anim Sci 1979;* 48:430-435.
7. Duff GC, Malcolm-Callis DA, Walker MW, Wiseman MW, Galyean ML, Perino LJ. Effects of intranasal versus intramuscular modified-live vaccines and vaccine timing on health and performance by newly received beef cattle. *Bov Pract* 2000; 34:66-71.
8. Frank GH. The role of *Pasteurella haemolytica* in the bovine respiratory disease complex. *Vet Med* 1986; 81:838-846.
9. Griffin D. Etiology, pathogenesis and clinical signs of bovine respiratory disease. In: Smith RA, ed. *Bovine respiratory disease. Sourcebook for the veterinary professional.* Trenton, NJ: Veterinary Learning Systems, 1996; 6-11.
10. Gwazdauskas FC, Gross WB, Bibb TL, McGilliard ML. Antibody titers and plasma glucocorticoid concentrations near weaning in steer and heifer calves. *Can Vet J* 1978; 19: 150-154.
11. Harland RJ, Potter AA, van Drunen-Little-Van den Hurk S, Van Donkersgoed J, Parker MD, Zamb TJ, Janzen ED, The effect of subunit or modified-live bovine herpesvirus-1 vaccines on the efficacy of a recombinant *Pasteurella haemolytica* vaccine for the prevention of respiratory disease in feedlot calves. *Can Vet J* 1992; 33:734-741.
12. Hoerlein AB. Shipping fever. In: Amstutz HE, ed. *Bovine medicine and surgery.* Santa Barbara, CA: Am Vet Publ, 1980; 99-106.
13. Ives S, Drouillard J, Anderson D, Stokka G, Kuhl G. Comparison of morbidity and performance among stressed feeder calves following vaccination with PYRAMID MLV 4 or PYRAMID 4 + PRESPONSE SQ. *Kansas State University, Cattlemen's Day, Report of Progress* 1999; 831:126-129.

14. Jensen R, Pierson RE, Braddy PM, Saari DA, Lauerman LH, England JJ, Keyvanfar H, Collier JR, Horton DP, McChesney AE, Benitez A, Christie RM. Shipping fever pneumonia in yearling feedlot cattle. *J Am Vet Med Assoc* 1976; 169:500-506.
15. Johnston JD, Buckland RB. Response of male Holstein calves from seven sires to four management stresses as measured by plasma corticoid levels. *Can J Anim Sci* 1976; 56:727-732.
16. Martin SW, Meek AH, Davis DG, Johnson JA, Curtis RA. Factors associated with mortality and treatment costs in feedlot calves: the Bruce County beef project. *Can J Comp Med* 1982; 46:341-349.
17. Poe KD, Beck PA, Richeson JT, Gadberry MS, Kegley EB, Hess TW, Hubbell DS. Effects of respiratory vaccination timing and growth-promoting implant on health, performance, and immunity of high-risk, newly received stocker cattle. *Prof Anim Sci* 2013; 29:413-419.
18. Purdy CW, Loan RW, Straus DC, Briggs RE, Franks GH. Conglutinin and immunoconglutinin titers in stressed calves in a feedlot. *Am J Vet Res* 2000; 61:1403-1409.
19. Rice JA, Carrasco-Medina L, Hodgins DC, Shewen PE. *Mannheimia haemolytica* and bovine respiratory disease. *Anim Health Res Rev* 2007; 8:17-28.
20. Richeson JT, Beck PA, Gadberry MS, Gunter SA, Hess TW, Hubbell III DS, Jones C. Effects of on- arrival versus delayed modified-live virus vaccination on health, performance, and serum infectious bovine rhinotracheitis titers of newly received beef calves. *J Anim Sci* 2008; 86:999-1005.
21. Richeson JT, Kegley EB, Gadberry MS, Beck PA, Powell JG, Jones CA. Effects of on-arrival versus delayed clostridial or modified-live respiratory vaccinations on health, performance, bovine viral diarrhea virus I titers and stress and immune measures of newly received beef calves. *J Anim Sci* 2009; 87:2409-2418.
22. Richeson JT. Vaccinating high-risk calves against BRD. *Bov Pract* 2015; 48:1-4.
23. Richeson JT, Beck PA, Poe KD, Gadberry MS, Hess TW, Hubbell III DS. Effects of administration of a modified-live virus respiratory vaccine and timing of vaccination on health and performance of high-risk beef stocker calves. *Bov Pract* 2015; 49:37-42.
24. Richeson JT, Carroll JA, Burdick Sanchez NC, May ND, Hughes HD, Roberts SL, Broadway PR, Sharon KP, Ballou MA. Dexamethasone treatment differentially alters viral shedding and the antibody and acute phase protein response after multivalent respiratory vaccination in beef steers. *J Anim Sci* 2016-0572; doi: 10.2527.
25. Roth JA. Cortisol as mediator of stress-associated immunosuppression in cattle. In: Moberg GP, ed. *Animal stress.* New York: Springer, 1985; 225-243.
26. Roth JA. How cattle defend themselves against *P. haemolytica* pneumonia. *Vet Med* 1988; 83:1067-1072.
27. Shaw KE, Nichols RE. Plasma 17-hydroxycorticosteroids in calves: the effects of shipping. *Am J Vet Res* 1964; 25:252-253.
28. Stephens DB. Stress and its measurement in domestic animals. A review of behavioral and physiological studies under field and laboratory situations. *Adv Vet Sci Comp Med* 1980; 24:179-210.
29. Stokka GL, Edwards AJ. Revaccination of stressed calves with a multiple polyvalent MLV vaccine. *Agri Pract* 1990; 11:18-20.
30. USDA. Vaccine usage in U.S. feedlots. USDA:APHIS:VS:CEAH. Fort Collins, CO. 2013; #672.0513.

Draxxin®
(tulathromycin)
Injectable Solution

OTHER BRD TREATMENTS WERE 50% AS EFFECTIVE AS DRAXXIN® IN SEVERAL STUDIES.

Treat bovine respiratory disease (BRD) the right way with DRAXXIN® (*tulathromycin*) Injectable Solution. DRAXXIN demonstrated 50% fewer re-treats and 50% fewer dead or chronic animals[1] versus competitive products in several large pen studies.[2] Which means your cattle stay healthier, and that helps keep your bottom line healthier, too.

BRD SOLUTIONS
FROM ZOETIS

Get the numbers on DRAXXIN at draxxin.com.

IMPORTANT SAFETY INFORMATION: DRAXXIN has a pre-slaughter withdrawal time of 18 days in cattle. Do not use in female dairy cattle 20 months of age or older. Do not use in animals known to be hypersensitive to the product. See Brief Summary of Prescribing Information on adjacent page and full Prescribing Information at draxxin.com/pi.

[1] Data on file, Study Report Nos. 1133R-60-05-491, A131R-US-12-028, 2132T-60-01-050, 1133R-60-02-376, 2132T-60-01-063, 1133R-60-03-388 and 11RGDRA01, Zoetis Services LLC.
[2] Data on file, Study Report Nos. 1133R-60-05-491, 1133R-60-05-492, 1133R-60-05-493, A131R-US-12-028, 2132T-60-01-050, 1133R-60-02-376, 2132T-60-01-063 and 1133R-60-03-388, Zoetis Services LLC.

zoetis

Brief Summary for use in Cattle:
See Package Insert for full Prescribing Information

Draxxin®
(tulathromycin)
Injectable Solution

Antibiotic
100 mg of tulathromycin/mL

For use in beef cattle (including suckling calves), non-lactating dairy cattle (including dairy calves), veal calves, and swine. Not for use in female dairy cattle 20 months of age or older.
CAUTION: Federal (USA) law restricts this drug to use by or on the order of a licensed veterinarian.

DESCRIPTION
DRAXXIN Injectable Solution is a ready-to-use sterile parenteral preparation containing tulathromycin, a semi-synthetic macrolide antibiotic of the subclass triamilide. Each mL of DRAXXIN contains 100 mg of tulathromycin as the free base in a 50% propylene glycol vehicle, monothioglycerol (5 mg/mL), with citric and hydrochloric acids added to adjust pH.

DRAXXIN consists of an equilibrated mixture of two isomeric forms of tulathromycin in a 9:1 ratio.

The chemical names of the isomers are (2R,3S,4R,5R,8R,10R, 11R,12S,13S,14R)-13-[[2,6-dideoxy-3-C-methyl-3-O-methyl-4-C-[(propylamino) methyl]-α-L-ribo-hexopyrano-syl] oxy]-2-ethyl-3,4,10-trihydroxy-3,5,8,10,12,14-hexamethyl-11-[[3,4,6-trideoxy-3-(dimethylamino)-β-D-xylo-hexopyrano-syl]-oxy]-1-oxa-6-azacyclopentadecan-15-one and (2R,3R,6R, 8R,9R,10S,11S,12R)-11-[[2,6-dideoxy-3-C-methyl-3-O-methyl-4-C-[(propylamino)methyl]-α-L-ribo-hexopyrano-syl]oxy]-2-[(1R,2R)-1,2-dihydroxy-1-methylbutyl]-8-hydroxy- 3,6,8,10,12-pentamethyl-9-[[3,4,6-trideoxy-3-(dimethylamino)- β-D-xylo-hexopyranosyl] oxy]-1-oxa-4-azacyclotridecan-13-one, respectively.

INDICATIONS
Beef and Non-Lactating Dairy Cattle
BRD – DRAXXIN Injectable Solution is indicated for the treatment of bovine respiratory disease (BRD) associated with *Mannheimia haemolytica, Pasteurella multocida, Histophilus somni,* and *Mycoplasma bovis*; and for the control of respiratory disease in cattle at high risk of developing BRD associated with *Mannheimia haemolytica, Pasteurella multocida, Histophilus somni,* and *Mycoplasma bovis.*
IBK – DRAXXIN Injectable Solution is indicated for the treatment of infectious bovine keratoconjunctivitis (IBK) associated with *Moraxella bovis.*
Foot Rot – DRAXXIN Injectable Solution is indicated for the treatment of bovine foot rot (interdigital necrobacillosis) associated with *Fusobacterium necrophorum* and *Porphyromonas levii.*
Suckling Calves, Dairy Calves, and Veal Calves
BRD - DRAXXIN Injectable Solution is indicated for the treatment of BRD associated with *M. haemolytica, P. multocida, H. somni,* and *M. bovis.*

DOSAGE AND ADMINISTRATION
Cattle
Inject subcutaneously as a single dose in the neck at a dosage of 2.5 mg/kg (1.1 mL/100 lb) body weight (BW). Do not inject more than 10 mL per injection site.

Table 1. DRAXXIN Cattle Dosing Guide

Animal Weight (Pounds)	Dose Volume (mL)
100	1.1
200	2.3
300	3.4
400	4.5
500	5.7
600	6.8
700	8.0
800	9.1
900	10.2
1000	11.4

CONTRAINDICATIONS
The use of DRAXXIN Injectable Solution is contraindicated in animals previously found to be hypersensitive to the drug.

WARNINGS
FOR USE IN ANIMALS ONLY.
NOT FOR HUMAN USE.
KEEP OUT OF REACH OF CHILDREN.
NOT FOR USE IN CHICKENS OR TURKEYS.

RESIDUE WARNINGS
Cattle
Cattle intended for human consumption must not be slaughtered within 18 days from the last treatment. Do not use in female dairy cattle 20 months of age or older.

PRECAUTIONS
Cattle
The effects of DRAXXIN on bovine reproductive performance, pregnancy, and lactation have not been determined. Subcutaneous injection can cause a transient local tissue reaction that may result in trim loss of edible tissue at slaughter.

ADVERSE REACTIONS
Cattle
In one BRD field study, two calves treated with DRAXXIN at 2.5 mg/kg BW exhibited transient hypersalivation. One of these calves also exhibited transient dyspnea, which may have been related to pneumonia.

NADA 141-244, Approved by FDA

To report a suspected adverse reaction or to request a safety data sheet call **1-888-963-8471**. For additional information about adverse drug experience reporting for animal drugs, contact FDA at 1-888-FDA-VETS or online at http://www.fda.gov/AnimalVeterinary/SafetyHealth.

zoetis

Distributed by:
Zoetis Inc. Kalamazoo, MI 49007

For additional DRAXXIN product information call: **1-888-DRAXXIN** or go to **www.DRAXXIN.com**

TAKE TIME  OBSERVE LABEL DIRECTIONS

032908ZO
Made in Brazil
Revised: February 2014