

# Effect of ambient temperature on viral replication and serum antibody titers following administration of a commercial intranasal modified-live infectious bovine rhinotracheitis-parainfluenza-3 virus vaccine to beef cattle housed in high- and moderate-ambient temperature environments

Gretchen P. Grissett, DVM; Brad J. White, DVM, MS; David E. Anderson, DVM, MS; Robert E. Larson, DVM, PhD; Matt D. Miesner, DVM, MS

**Objective**—To evaluate the effect of ambient temperature on viral replication and serum antibody titers following administration of an intranasal modified-live infectious bovine rhinotracheitis (IBR)-parainfluenza-3 (PI3) virus vaccine to beef calves housed in high- (> 32°C) and moderate- (21°C) ambient temperature environments.

**Animals**—28 calves (mean weight, 206.8 kg).

**Procedures**—Calves were randomly allocated to 4 treatment groups (housed outdoors during high ambient temperature with [HAT; n = 10] or without [HAC; 4] vaccination or housed indoors in a moderate ambient temperature with [MAT; 10] or without [MAC; 4] vaccination). Rectal and nasal mucosal temperatures were recorded every 2 hours from 8 AM to 8 PM on days 0 (vaccination) and 1. Nasal swab specimens were obtained on days 0 through 7 for virus isolation. Serum samples were collected on days 0, 7, 14, and 28 for determination of antibody titers.

**Results**—Mean rectal temperature did not differ among the treatment groups. Mean nasal temperature for the HAT group was significantly higher than that for the MAT group at 6, 24, 30, 32, and 38 hours after vaccination. Viable IBR virus was isolated from all vaccinated calves on days 1 through 6. Two weeks after vaccination, vaccinated calves had anti-IBR antibody titers that were significantly greater than those for unvaccinated calves. Mean anti-IBR antibody titers did not differ significantly between the HAT and MAT groups.

**Conclusions and Clinical Relevance**—Results indicated that, following vaccination with an intranasal modified-live IBR-PI3 virus vaccine, IBR viral replication and serum antibody titers did not differ significantly between calves housed in high- and moderate-ambient temperature environments. (*Am J Vet Res* 2014;75:1076–1082)

**B**ovine respiratory disease is a multifactorial disease in which infectious agents, compromised host immune systems, and environmental factors interact to cause bronchopneumonia in affected cattle. It is a prom-

Received June 26, 2014.

Accepted August 4, 2014.

From the Department of Clinical Sciences, College of Veterinary Medicine, Kansas State University, Manhattan, KS 66506. Dr. Anderson's present address is Department of Large Animal Clinical Sciences, College of Veterinary Medicine, University of Tennessee, Knoxville, TN 37996.

This manuscript represents a portion of a thesis submitted by Dr. Grissett to the Kansas State University Department of Clinical Sciences as partial fulfillment of the requirements for a Master of Science degree.

Supported by Merck Animal Health.

Presented as a poster presentation at the American Association of Bovine Practitioners Meeting, Milwaukee, September 2013.

The authors thank David Amrine, Miles Theurer, and Carrie Wheeler for technical assistance.

Address correspondence to Dr. White (bwhite@vet.k-state.edu).

## ABBREVIATIONS

BRD	Bovine respiratory disease
CIS	Clinical illness score
IBR	Infectious bovine rhinotracheitis virus
IN	Intranasal
MLV	Modified-live virus
PI3	Parainfluenza-3 virus

inent disease of the feedlot industry. Economic losses caused by death, reduced feed efficiency, and treatment costs are estimated at \$23.60/treated calf, and 96.9% of US feedlots have cattle affected with BRD.<sup>1</sup> Infectious bovine rhinotracheitis virus and PI3 are pathogens associated with the BRD complex. A variety of clinical manifestations are associated with IBR, including rhinotracheitis, vulvovaginitis, balanoposthitis, abortion, and encephalitis.<sup>2,3</sup> Additionally, IBR plays an important role in the BRD complex through immune suppression,

which increases susceptibility to secondary bacterial infections.<sup>2</sup> The primary role of PI3 in the BRD complex is to predispose the bovine respiratory tract to subsequent infections caused by other viruses and bacteria.<sup>2</sup> Results of 1 study<sup>4</sup> indicate that 12% (68/569) and 23% (131/569) of cattle entering a feedlot had antibody titers against PI3 and IBR, respectively. Because of the prominent roles that IBR and PI3 play in the BRD complex, substantial focus is placed on their prevention.

A mainstay of BRD prevention is vaccination. In 2011, the National Animal Health Monitoring System survey<sup>1</sup> estimated that 93.7% of US feedlots had used IBR vaccines, with 53.7% of those administering the IBR vaccine by the IN route, and 85.1% had used PI3 vaccines.<sup>1</sup> Currently available vaccines for BRD contain either inactivated viral antigens or MLV, and MLV vaccines are considered to have superior efficacy because of their ability to induce both humoral and cell-mediated immunity and interferon.<sup>5</sup> Results of 1 study<sup>6</sup> indicate that calves that were vaccinated against IBR with an MLV vaccine had significantly higher serum anti-IBR antibody titers than did calves that were vaccinated against IBR with an inactivated vaccine 2 weeks after administration of a second (booster) dose of the assigned vaccine. The goal of IN administration of a modified-live IBR-PI3 vaccine to cattle is to provide immunity against viral colonization of the nasal passages and subsequent disease development.<sup>7</sup> Similar to parenteral administration, IN administration of a modified-live IBR-PI3 vaccine induces systemic immunity; however, IN administration results in a quicker onset of local immunity than does parenteral administration.<sup>7</sup> Temperature-sensitive vaccines are MLV vaccines in which the viral components have been modified so that viral replication decreases as temperature increases; therefore, at high temperatures, these vaccines have only limited viral replication to stimulate the immune response. In *in vivo* models,<sup>8</sup> temperature-sensitive vaccines have diminished viral replication at temperatures > 39°C; thus, IN is the preferred route of administration for these vaccines because nasopharyngeal temperatures generally only reach 32° to 34°C, which allows the vaccine virus to replicate unhindered and produce an immune response. Extreme environmental temperatures may affect the efficacy of temperature-sensitive vaccines or the ability of vaccinated animals to respond to the immunization.

Cattle are often administered IN vaccines when environmental conditions of ambient temperature and humidity are adversely high. Results of 1 study<sup>9</sup> indicate that nasal mucosal temperature exceeded 39°C in 2 of 8 healthy calves and 9 of 10 calves with experimentally induced BRD during a period of high ambient temperatures (mean daily temperature,  $\geq$  32°C; humidity,  $\geq$  40%). To our knowledge, the effect of ambient temperature on viral replication and the immunologic response of beef calves following IN administration of a modified-live IBR-PI3 vaccine is unknown. The purpose of the study reported here was to compare the serologic response (anti-IBR and anti-PI3 antibody titers) and extent of viral replication in beef calves following administration of an IN modified-live IBR-PI3 vaccine during summer conditions (high ambient temperatures) with those of calves administered the same vac-

cine during moderate conditions. The hypothesis was that calves housed in a high-ambient temperature environment would have a decreased serologic response and decreased virus replication, compared with the serologic response and virus replication in calves housed in a climate-controlled environment with a moderate ambient temperature.

## Materials and Methods

**Animals**—All study procedures were conducted in accordance with a protocol approved by the Kansas State University Institutional Animal Care and Use Committee. Thirty weaned black heifer calves were obtained for the study and housed at the Kansas State University Large Animal Research Center in Manhattan, Kan, during July and August when the maximum daily temperature was forecasted to be > 32.2°C. On arrival at the research center, all calves were individually weighed, identified by the application of individual ear tags, and metaphylactically treated with ceftiofur crystalline-free acid<sup>a</sup> (6.6 mg/kg, SC in the base of an ear). A random number generator<sup>b</sup> was used to allocate 28 calves to 1 of 4 treatment groups (housed outdoors during a period of high ambient temperatures with [HAT; n = 10] or without [control; HAC; 4] vaccination or housed indoors in a moderate ambient temperature with [MAT; 10] or without [control; MAC; 4] vaccination). The 2 remaining calves served as substitutes in case any of the other calves developed clinical disease during the 21-day observation period prior to vaccine administration; one was randomly allocated to be housed with the HAC group and the other to the MAC group so that they would be acclimatized if needed.

After initial processing, calves were placed in their appropriate group and housing environment. The HAT and HAC groups were housed in separate but identical outdoor pens (12.2 × 24.4 m) on opposite sides of a 3.7-m-wide alley, which ensured separation and prevented direct contact between the 2 groups. Both groups had access to an open-faced shed for shelter. Those 2 groups were handled and processed separately through the same outdoor chute to prevent nose-to-nose contact between groups, and the HAC group was processed first except on the day of vaccination. The MAT group was housed in 1 pen (6.1 × 9.1 m) in an indoor climate-controlled facility (temperature, approx 21°C). The MAC group was housed in 2 identical pens (3.0 × 3.0 m; 2 calves in 1 pen and 3 calves in the other pen during the 21-day observation period prior to vaccination [day 0] and then 2 calves/pen after day 0) in the same indoor climate-controlled facility as the MAT group, but was kept separate from the MAT group so that there was no possibility of direct contact between the 2 groups. The calves in the MAT and MAC groups were processed separately through an indoor chute and alley to maintain the proper temperature acclimation at all times, and the MAC group was processed first except on the day of vaccination.

Throughout the study, calves were fed a complete starter grain ration<sup>c</sup> and had *ad libitum* access to grass hay, supplemental salt blocks, and water. The starter ration contained 13.5% crude protein and 2.79% crude fat with a total digestible nutrient proportion of 78.18%

and was fed twice a day at approximately 8 AM and 4 PM. For the first week after arrival, calves were fed 1.4 kg of starter ration/calf/d. The amount of starter ration fed was gradually increased to 1.6 kg/calf/d for the last 2 weeks of the study.

Eight days after the calves arrived at the research facility, biothermal radiofrequency chips<sup>d</sup> were implanted in all calves in the HAT group and 7 randomly selected (by means of a random number generator<sup>a</sup>) calves in the MAT group. The chips were implanted in the submucosa of the right nares approximately 100 mm caudal to the alar cartilage and 2 mm deep to the nasal mucosa as described.<sup>9,10</sup> Once implanted, the nasal mucosal temperature as measured by the biothermal chip was monitored by use of a digital electronic microchip scanner.<sup>d</sup>

**Study design**—The study had a 2 × 2 factorial design. Because of the differences in housing environment and the number of calves assigned to the vaccinated and control groups, investigators were aware of the treatment group assignment for all calves throughout the study (ie, the investigators were not blinded).

**Vaccine administration**—Study initiation was determined on the basis of predicted outdoor temperatures consistently exceeding 32.2°C for the first week after vaccination. Twenty-one days after arrival at the research facility, a commercial IN modified-live IBR-PI3 vaccine<sup>e</sup> or sterile saline (0.9% NaCl) solution was administered to all calves (day 0). Calves were restrained in a head gate and chute. Calves in the HAT and MAT groups were administered 1 mL of the vaccine in each nostril in accordance with the label instructions. Following vaccine administration to the HAT and MAT groups, the head gates and chutes were disinfected and personnel changed attire before administering the saline solution to the control groups to prevent exposure of the control calves to the vaccine. Calves in the HAC and MAC groups were administered 1 mL of sterile saline solution in each nostril. One veterinarian (GPG) administered all treatments.

**Clinical observation of calves**—One veterinarian (GPG) trained in the detection of clinical illness and BRD observed all calves twice daily for the duration of the study. During each observation, each calf was assigned a CIS ranging from 1 to 4 (1 = normal behavior; 2 = slight illness, mild lethargy, or cough; 3 = moderate illness, severe lethargy, labored breathing, or cough; and 4 = severe illness, moribund, or little response to human approach) on the basis of health characteristics. During the 21-day observation period prior to vaccine administration, calves that were assigned a CIS ≥ 2 had a physical examination performed, which included measurement of rectal temperature. Calves with a CIS ≥ 2 and rectal temperature ≥ 40°C at any time during that observation period were administered appropriate treatments and removed from the study. Beginning on day 0 and for next 28 days, calves with a CIS ≥ 2 and rectal temperature ≥ 40°C had physical examinations performed, were administered appropriate treatments, and remained in their assigned treatment groups for data collection.

**Temperature monitoring**—Nasal mucosal temperatures for the calves in the HAT and MAT groups

and rectal temperatures for all calves were measured immediately prior to and every 2 hours between 8 AM and 8 PM for the first 48 hours after administration of the vaccine or saline solution. Nasal mucosal and rectal temperatures were measured twice daily on days 2 through 7 and were obtained once weekly during processing thereafter for the remainder of the study.

**Sample collection**—During sample collection, control calves (HAC and MAC groups) were processed first to minimize their risk of exposure to the vaccine strains of IBR and PI3. For each calf, body weight was measured on days -21 (arrival at research facility), 7, 14, 21, and 28. A nasal swab specimen was obtained from each nostril immediately prior to administration of the vaccine or saline solution (baseline) and on days 1 through 7 for virus isolation. Each nostril was swabbed approximately 10 cm caudal to the alar cartilage with a sterile cotton-tipped swab. The 2 nasal swab specimens obtained from each calf during each collection time were placed in the same viral culture media and immediately frozen and stored at -80°C until analysis.

A blood sample was obtained from each calf at baseline and on days 7, 14, 21, and 28. Following collection, each sample was allowed to clot and then centrifuged so that the serum could be harvested. Serum samples were stored at -80°C until analysis.

**Sample processing**—Once collection of each respective sample type was completed, all nasal swab specimens and serum samples were submitted to the Kansas State Veterinary Diagnostic Laboratory for analysis. Laboratory personnel that performed the analyses were blinded to the treatment group assignment for all calves. Nasal swab specimens obtained at baseline were evaluated for the presence of IBR DNA and PI3 RNA by means of PCR assays to determine whether any of the calves were infected with either virus prior to vaccination. Virus isolation for IBR and PI3 was performed on all nasal swab specimens. The TCID<sub>50</sub> (ie, the endpoint dilution required to produce a cytopathic effect in 50% of the inoculated tissue culture cells<sup>11</sup>) was determined for each sample from which IBR or PI3 was isolated. Serum neutralization for determination of antibodies against IBR was performed on all serum samples. Samples with an anti-IBR antibody titer ≥ 1:4 were considered positive.<sup>12</sup> Serum neutralization for determination of antibodies against PI3 was not performed because it was discovered that the HAT and MAC groups contained PI3-infected calves, and it would have been impossible to distinguish between antibodies induced by infection and those induced by the vaccine.

**Environmental monitoring**—Throughout the study, outdoor temperature and humidity were measured hourly with wireless weather monitoring technology.<sup>f</sup> For the first 48 hours after vaccine administration, indoor temperature and humidity were measured hourly with an environmental data monitor.<sup>g</sup> Beginning on day 2 and for the remaining duration of the study, the maximum indoor temperature and humidity were measured daily with a commercial monitor.<sup>h</sup>

**Statistical analysis**—Data were imported into a commercial statistical software package<sup>i</sup> for descriptive

and statistical analyses. Outcome variables of interest included serum anti-IBR antibody titer, IBR TCID<sub>50</sub>, nasal mucosal temperature, and rectal temperature, and the distributions of those variables were assessed for normality by means of visual assessment. Data for variables that were not normally distributed (anti-IBR antibody titer) were logarithmically transformed for analysis and then back-transformed for reporting purposes. For each outcome of interest, a mixed generalized linear model was used to assess its association with treatment group (HAT, HAC, MAT, and MAC), time of sample collection relative to vaccine administration (time), and the interaction between treatment group and time. All models included a random effect to account for repeated measures on individual calves. For all analyses, values of  $P \leq 0.05$  were considered significant.

## Results

**Calves**—On arrival to the research facility, the mean  $\pm$  SD weight of the calves was  $206.8 \pm 25.4$  kg. During the 21-day observation period prior to vacci-

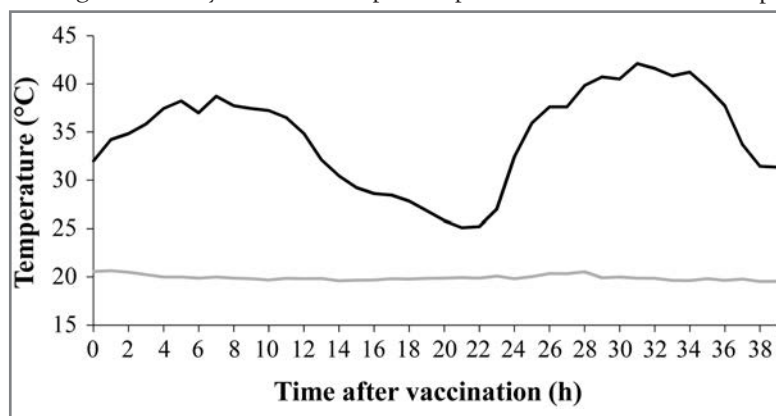


Figure 1—Indoor (solid gray line) and outdoor (solid black line) environmental temperatures for the 38 hours immediately after IN administration (8 AM; hour 0) of a modified-live IBR-PI3 vaccine or sterile saline (0.9% NaCl) solution to 28 weaned black beef calves.

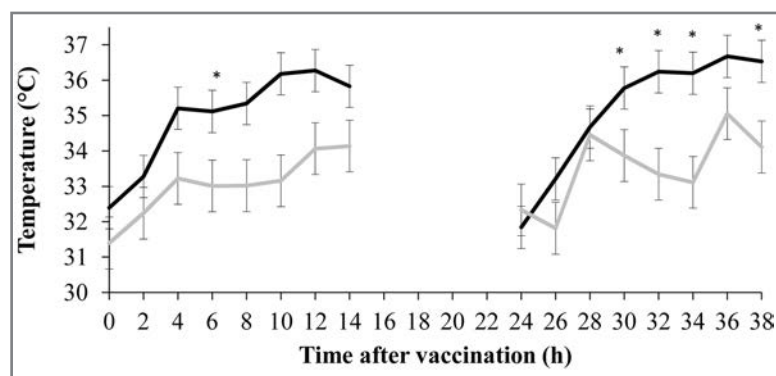


Figure 2—Mean  $\pm$  SD nasal mucosal temperatures for the 38 hours immediately after administration of an IN modified-live IBR-PI3 vaccine to weaned black beef calves that were housed outdoors during a period of high ( $> 32.2^\circ\text{C}$ ) ambient temperatures (HAT;  $n = 10$ ; black line) or housed indoors in a controlled moderate-temperature (approx  $21^\circ\text{C}$ ) environment (MAT; 10; gray line). Temperatures were not measured between 14 and 24 hours after vaccine administration (hour 0; day 0). Although there were 10 calves in the MAT group, only 7 calves had biotermal chips implanted in the nasal mucosa, and nasal mucosal temperature data for 1 calf in the MAT group that had a CIS  $> 1$  and rectal temperature  $> 40^\circ\text{C}$  on day 1 were excluded from this analysis. Therefore, values for the MAT group represent the means for 6 calves. \*Within an hour, mean values differ significantly ( $P < 0.01$ ) between the HAT and MAT groups.

nation (day 0), 2 calves (1 each in the HAT and MAC groups) developed BRD and were treated and removed from the study and replaced with the appropriately acclimated substitute calves.

On day 14, a calf in the MAC group was treated for frothy bloat. It recovered without any complications and remained in the study. The data from that calf were included in all analyses.

Following vaccination, 4 calves (MAT [ $n = 3$ ; 1 on day 1 and 2 on day 9]; HAT [1; day 12]) were assigned a CIS  $> 1$  and had a rectal temperature  $> 40^\circ\text{C}$ . All calves were determined to have BRD on the basis of physical examination results and were treated with florfenicol<sup>l</sup> (40 mg/kg, SC). The calves were retained in their respective treatment groups. Temperature data for the calf treated on day 1 were not included in the analyses; however, temperature data for the calves treated on days 9 and 12 were included in the analyses. Virus isolation and serology data remained in the analysis for the calf treated on day 1. The calf with BRD in the HAT group died on day 15, and necropsy findings included bronchopneumonia and hepatic lipidosis. All data from that calf prior to its death were included in the analyses.

**Environmental temperature**—The maximum outdoor temperature (Figure 1) and relative humidity were  $38.7^\circ\text{C}$  and 49%, respectively, on day 0 and  $42.1^\circ\text{C}$  and 66%, respectively, on day 1. The mean  $\pm$  SD outdoor relative humidity was  $40.0 \pm 12.7\%$  for days 1 and 2. The mean  $\pm$  SD indoor temperature and relative humidity were  $19.9^\circ \pm 0.28^\circ\text{C}$  and  $70.9 \pm 5.1\%$  on days 0 and 1.

**Treatment group comparisons**—Nasal mucosal temperature was significantly ( $P < 0.01$ ) associated with treatment group. The nasal mucosal temperature increased throughout the day for all calves during the intensive measurement period on days 0 and 1 (Figure 2), and during that period, the maximum nasal mucosal temperature was  $36.6^\circ\text{C}$  for calves in the HAT group and  $35.1^\circ\text{C}$  for calves in the MAT group. The mean nasal mucosal temperature for calves in the HAT group was significantly higher than that for calves in the MAT group at 6, 30, 32, 34, and 38 hours after vaccination.

The interaction between treatment group and the time of sample collection relative to vaccine administration (time) was significantly ( $P < 0.01$ ) associated with rectal temperature. Mean rectal temperature did not differ significantly at any time between the 2 groups of vaccinated calves (HAT and MAT). However, the mean rectal temperature for the calves in the MAC group was significantly lower, compared with that for calves in the HAT, HAC, and MAT groups at 5, 6, 7, 8, and 14 hours after vaccination (Figure 3).



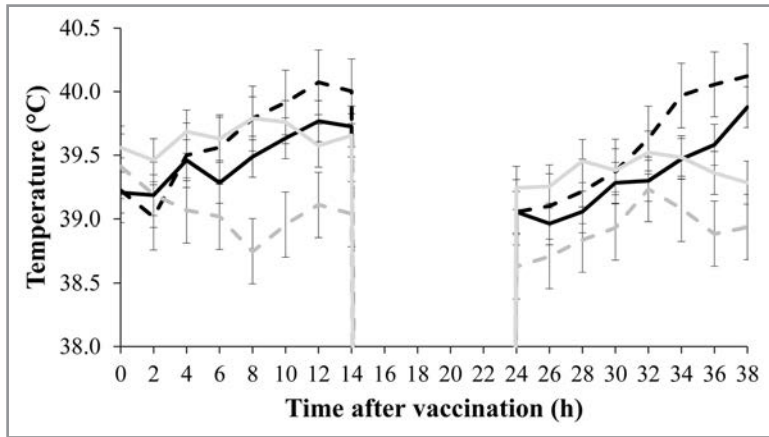


Figure 3—Mean  $\pm$  SD rectal temperatures of weaned black beef calves that were housed outdoors during a period of high ( $> 32.2^{\circ}\text{C}$ ) ambient temperatures and were (HAT;  $n = 10$ ; solid black line) or were not (HAC; 4; dashed black line) administered an IN modified-live IBR-PI3 vaccine and of calves that were housed indoors in a controlled moderate-temperature (approx  $21^{\circ}\text{C}$ ) environment and were (MAT; 10; solid gray line) or were not (MAC; 4; dashed gray line) administered the same vaccine during the 38 hours immediately after vaccine administration (hour 0; day 0). Temperature data for 1 calf in the MAT group that had a CIS  $> 1$  and rectal temperature  $> 40^{\circ}\text{C}$  on day 1 were excluded from this analysis. Therefore, values for the MAT group represent the means for 9 calves. See Figure 2 for remainder of key.

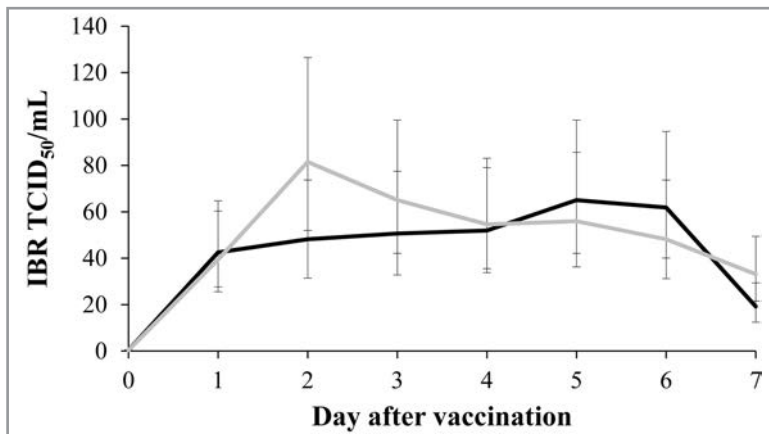


Figure 4—Model-estimated IBR TCID<sub>50</sub>  $\pm$  95% confidence intervals for calves in the HAT ( $n = 10$ ; black line) and MAT (10; gray line) groups of Figure 2 on days 0 through 7. See Figure 2 for remainder of key.

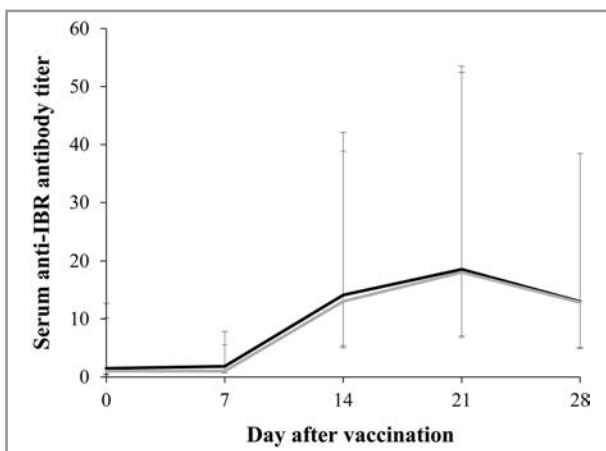


Figure 5—Model-estimated least squares mean  $\pm$  95% confidence interval serum anti-IBR antibody titers for calves in the HAT ( $n = 10$ ; black line) and MAT (10; gray line) groups of Figure 2 on days 0, 7, 14, 21, and 28. One calf in the HAT group developed BRD and died on day 15; therefore, the values for the HAT group on days 21 and 28 represent the mean for only 9 calves. See Figure 2 for remainder of key.

For all calves, nasal swab specimens obtained on day 0 yielded negative results for IBR as determined by virus isolation and PCR assay. Day 0 nasal swab specimens obtained from 2 calves (1 each in the HAT and MAC groups) yielded positive results for PI3 on both virus isolation and PCR assay.

Virus isolation results for IBR were negative for all calves in the control groups (HAC and MAC) during the first week of the study (days 0 to 7). Infectious bovine rhinotracheitis virus was isolated from all calves in the vaccinated groups (HAT and MAT) on days 1 through 6 and from all calves except 1 calf in the HAT group on day 7. The interaction between vaccinated calves and time was significantly ( $P < 0.01$ ) associated with the TCID<sub>50</sub>; however, the TCID<sub>50</sub> did not vary significantly between calves in the HAT and MAT groups at any time during days 0 through 7 (Figure 4).

All calves were seronegative for anti-IBR antibodies (SN titer,  $< 1:4$ ) on day 0, and all calves in the control groups (HAC and MAC) remained seronegative for anti-IBR antibodies throughout the duration of the study. All calves in the HAT and MAT groups were seropositive for anti-IBR antibodies by day 28. Although the interaction between treatment group and time was significantly ( $P < 0.01$ ) associated with the serum anti-IBR antibody titer, the mean titer did not differ significantly between calves in the HAT and MAT groups at any time during the study (Figure 5).

## Discussion

Results of the present study indicated that the extent of viral replication and the mean serum anti-IBR antibody titer did not differ significantly between beef calves that were vaccinated with an IN modified-live IBR-PI3 vaccine during a period when the ambient temperature was  $> 32.2^{\circ}\text{C}$  and those that were vaccinated while housed in a controlled moderate temperature (mean temperature,  $19.9^{\circ}\text{C}$ ) environment. The purpose of this study was to evaluate the effect of high environmental temperatures on the ability of the vaccine viruses to replicate and induce a serologic immune response in weaned beef calves. Although the study calves were processed and vaccinated during a period of high ambient temperature and high relative humidity, the study was conducted in accordance with a protocol that was approved by the university's institutional animal care and use committee, and these results do not imply that cattle should be processed or vaccinated during periods of extreme heat. Handling cattle during periods of high temperature and humidity should be done with care in accordance with industry-recommended guidelines to minimize heat stress.<sup>13,14</sup>

In the present study, the maximum nasal mucosal temperature did not exceed 39°C for any calf, a finding that contradicted the findings of other studies<sup>9,15</sup> in which calves housed in high ambient temperatures occasionally had nasal temperatures > 39°C.<sup>9</sup> The apparent contradiction in findings for maximum nasal mucosal temperature between the present study and those other studies<sup>9,15</sup> might be attributable to other environmental factors such as relative humidity. For example, in one of those studies,<sup>9</sup> the relative humidity ranged from 64% to 89%, whereas the relative humidity for the 2 days immediately after vaccination (days 0 and 1) in the present study ranged from 49% and 66%. The low relative humidity during the present study might have contributed to the observation of lower nasal mucosal temperatures observed in the present study, compared with those observed in that other study.<sup>9</sup>

Interestingly, the rectal temperature trend for the calves that were vaccinated and maintained in a moderate-temperature environment (MAT) was similar to that for calves that were housed outdoors in a high-temperature environment (HAT and HAC). Also, the mean rectal temperatures for calves in the HAT, HAC, and MAT groups were significantly higher than those for control calves that were housed in the moderate-temperature environment (MAC) at several points during the 48 hours immediately after vaccination. The reason calves in the MAT group had significantly higher rectal temperatures than did the calves in the MAC group is unknown, but could be associated with the presence of subclinical BRD or a vaccine-induced immunologic response in the calves of the MAT group. The calves in the HAT group were housed outdoors in extreme environmental conditions, and it is likely that those calves had nearly reached their maximum physiologic capabilities, which limited their potential temperature response to vaccination.<sup>15</sup>

Because we discovered evidence that PI3 was circulating among the study calves immediately prior to vaccination, we did not evaluate the effect of vaccination on the extent of PI3 replication and induction of serum anti-PI3 antibodies. The vaccine-induced anti-IBR antibody response was similar for calves in the HAT and MAT groups, with all calves in both groups seroconverting (serum anti-IBR antibody titer, > 1:4) by 28 days after vaccination. This finding suggested that high ambient temperature had no effect on the calves' response to vaccine administration. Similar to results of another study,<sup>16</sup> the serum anti-IBR antibody titers for the calves vaccinated in the present study peaked 21 days after administration of an IN modified-live IBR vaccine.

For the first week (days 1 through 6) after vaccine administration, IBR was isolated from all calves in the HAT and MAT groups, and the mean TCID<sub>50</sub> for IBR did not differ significantly between the 2 groups at any time. This suggested that high ambient temperature had a minimal effect on the viability of the vaccine virus after administration. In a similar study,<sup>16</sup> IBR was isolated from vaccinated calves up to 8 days after administration of an IN modified-live IBR vaccine, with peak IBR recovery occurring 4 days after vaccination. In the present study, IBR was still detectable in the nasal swab specimens of 9 of 10 calves in the HAT group and all

calves in the MAT group on day 7, and determination of the cessation of vaccine-induced IBR replication would have required a longer observation period, which was beyond the scope of the study design.

One of the limitations of the present study was that pen location and the number of calves in each pen prevented the study observers from being blinded to the treatment group assignments of individual calves. This lack of observer blinding likely resulted in minimal bias because the primary outcomes of interest were the recovery of viable virus and serum anti-IBR antibody titer, which were determined by personnel at the diagnostic laboratory, who were blinded to the treatment group assignments of the individual calves.

Another limitation of the present study was that biothermal chips could not be implanted in all of the calves because of a lack of availability of a sufficient number of chips. Consequently, biothermal chips were not implanted in any of the calves of the control groups, and we were unable to compare the nasal mucosal temperature between vaccinated and control calves. Also, the presence of PI3 in calves prior to vaccination prevented us from evaluating the vaccine-induced immune response for that virus.

In the present study, administration of an IN modified-live IBR-PI3 vaccine stimulated an IBR immune response in calves housed in a high-ambient temperature environment (> 32.2°C) that was similar to that in calves housed in a controlled moderate-temperature environment (mean temperature, 19.9°C). Viable IBR was recovered from all vaccinated calves on days 1 through 6 after vaccination, and all vaccinated calves developed serum antibodies against IBR by 28 days after vaccination. These findings indicated that IN administration of this particular vaccine to weaned beef calves during periods of high ambient temperatures had minimal effect on the vaccine-induced immune response. However, this does not mean that calves should be vaccinated during periods of high ambient temperature, and caution should always be used when cattle are processed during periods of extreme heat.

- a. Excede, Pfizer Animal Health, New York, NY.
- b. Microsoft Office Excel 2007, Microsoft, Redmond, Wash.
- c. Key Feeds, Clay Center, Kan.
- d. Biothermal RFID Chip, Destron Technologies, Round Rock, Tex.
- e. Nasalgen IP, Merck Animal Health, Summit, NJ.
- f. Wireless weather station, La Crosse Technology Ltd, La Crosse, Wis.
- g. HOBO U12 Temperature/Relative Humidity/Light/External Data Logger, Onset, Bourne, Mass.
- h. Springfield 91551 Daily Digital Humidity and Temperature Monitor, Taylor Precision Products, Oak Brook, Ill.
- i. JMP, SAS Institute Inc, Cary, NC.
- j. Nufloor, Merck Animal Health, Summit, NJ.

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