Infectious Bovine Rhinotracheitis, Bovine Viral Diarrhea and Parainfluenza-3 Viral Antibodies in Louisiana Cattle.

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Introduction

Bovine respiratory diseases (BRD) represent significant disease problems to the cattle industry. The infectious etiologies including viruses, bacteria, mycoplasmas, and chlamydia have been isolated from cattle with respiratory infections (12). The role of various viruses in BRD has been extensively reviewed (12,13). Three viruses commonly associated with BRD are: bovine herpesvirus-1 (BHV-1), also known as infectious bovine rhinotracheitis virus (IBR virus), bovine viral diarrhea (BVD), and parainfluenza-3 (PI-3) viruses. There are no published prevalence studies using serologic studies for these virus infections in Louisiana cattle. This paper describes the prevalence of IBR, BVD, and PI-3 viral infections in Louisiana cattle as based on serologic tests for viral antibodies. These prevalence studies also present information on which management recommendations concerning vaccinations can be assessed.

Materials and Methods

Sera: Sera were from cattle in five Louisiana Agricultural Experiment Station research herds. These cattle were apparently healthy when blood samples were taken. No IBR, BVD, or PI-3 vaccines had been administered to these cattle nor was there a history of prior illness due to IBR, BVD, or PI-3 viruses in these herds. Animals selected for bleeding were adult (>2 years old) breeding cattle. Upon receipt of the blood samples, the blood was centrifuged and the serum decanted and stored at -20C until assayed for viral antibodies.

Cell Cultures and Viruses: Monolayer cultures used in the study included bovine turbinate (BT) and Crandell feline kidney (CRFK) cells. The source of these cultures, growth conditions, and mediums have been described (4,8).

Viruses used included: IBR, Colorado 1 vaccinal strain (5); BVD virus, Singer strain (8); and the SF-4 strain of PI-3 virus obtained from the USDA, NVSL, Ames, IA. The viral stocks were propagated in CRFK cells (IBR) and BT cells (BVD virus and PI-3 virus). When maximum cytopathic effect (CPE) was nearly complete, the cultures were processed and stored as described (4). IBR and BVD viruses were titered in 96-well microtitration plates using CRFK cells for IBR virus assay and BT cells for BVD virus assay. PI-3 virus was titered for hemagglutination activity by a microtitration procedure.

Virus Neutralization Test A microtitration virus neutralization test (VNT) in 96-well microtitration plates was used to quantitate IBR and BVD viral neutralizing antibodies.(6) Plates were incubated at 37 C for 4 days (IBR) and 7 days (BVD) respectively. Each serum was tested in duplicate rows. The VNT titers were expressed as the final dilution of serum, (0.025 ml serum dilution and 0.025 virus dilution) prior to adding 0.05 ml cell suspension, that completely inhibited viral CPE of 100 $TCID_{50}$ in both wells per dilution. Thus for IBR and BVD serologic tests, 1:2 (undiluted serum and virus dilution) was the lowest dilution tested. Antibody titers of 1:2 or higher were considered positive in this study. For each group of serums tested, serum and virus controls were included. Also included were serums proven positive or negative for the respective viral antibodies.

Hemagglutination - Inhibition Test A microtitration hemagglutination-inhibition (HI) test was used to detect PI-3 viral antibodies. All serums were heat inactivated as described above. For each serum tested, 0.025 ml of

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phospate buffered saline (PBS) was added to each well in 96-well "V" bottom plates. To the first well, 0.025 ml of a 1:5 serum dilution in PBS was added with subsequent twofold dilutions made with microdiluting loops. Thus the lowest dilution prior to addition of antigen was 1:10. Antigen was added to each well in 0.025 ml aliquots and contained 4 hemagglutinating (HA) units. The plates were agitated to mix the components and incubated for 30 minutes at room temperature. Chicken red blood cells (RBC), 0.75% suspension, were added to each well in 0.05 aliquots. The plates were then incubated for 1 hour at room temperature for RBC sedimentation and HI activity. The HI titer was expressed as the dilution of serum, (0.025 ml serum dilution) prior to addition of antigen and RBC, which completely inhibited hemagglutination. Antibody titers of 1:10 (the lowest dilution tested or higher were considered positive for this study. Controls included serum, RBC and virus control. Also known positive and negative serums for PI-3 viral antibodies were included.

Statistical analysis: Statistical evaluation was performed using a chi square test.

Results

The results of IBR, BVD, and PI-3 serologic testing are summarized in Table 1. IBR, BVD, and PI-3 viral anitibodies were present in each of the five herds tested. The prevalence rate in each herd for IBR ranged from 18.8% to 60%; for BVD, 50.0% to 100%; and for PI-3, 85.0 to 98.2%. There were statistically significant differences ($p<0.01$) in the prevalence rates of seropositive animals for IBR among the five herds. Similar statistically significant differences ($P<0.01$) were observed for BVD and PI-3 viruses among the five herds.

Discussion

The results of this serosurvey indicate that IBR, BVD and PI-3 viral infections are common in Louisiana cattle. Also, the viruses appear widespread in the state and cause subclinical infections for the herds tested. The IBR, BVD, and PI-3 viruses are considered widespread in cattle populations. (2,7,9,10,11,14)

Numerous serologic surveys have been reported and cited in the above references. Antibody prevalence studies indicate the widespread nature of these viruses. Kahrs (10) reported serosurveys indicating 10% to 96% of cattle tested had IBR antibodies. However, those prevalence studies may reflect natural infections and/or vaccination. Kahrs (11) reported that antibody prevalence studies indicated that 0% to 100% of adult cattle had BVD viral antibodies with averages around 50% for North American cattle. Gillespie and Timoney (7) cited previous serolgic survey results of 53% to 73% for BVD viral antibodies. Potgeiter (13) reported that approximately 85% of cattle have PI-3 viral antibodies. Abinanti (1) reported that PI-3 viral antibody distribution ranged from 53% to 85% in beef cattle tested.

Table 1. Prevalence of IBR, BVD, and PI-3 viral antibodies in selected Louisiana cattle herds, 1976.

	Virus		
Herd	IBR	BVD	PI-3
A	39/110*(35.4%)†	77/110 (70.0%)	108/110 (98.2%)
B	30/160 (18.8%)	80/160 (50.0%)	136/160 (85.0%)
C	45/75 (60.0%)	61/75 (81.3%)	70/75 (93.3%)
D	16/48 (33.3%)	33/48 (68.8%)	44/48 (91.7%)
E	26/51 (51.0%)	51/51 (100.0%)	50/51 (98.0%)
Total	155/444 (34.9%)	302/444 (68.0%)	408/444 (91.9%)

*Number of cattle with antibodies/number of cattle tested.
†Percent of cattle with antibodies.

An example of a later serosurvey of U.S. cattle was reported by Crandell and Melloh (3). In that study of Illinois cattle tested which had not received IBR, BVD, or PI-3 vaccines: 39.9% were seropositive to IBR; 59.2% to BVD; and 60.1% to PI-3. Thus, it would appear that for the present study the antibody prevalence rates of 34.9% for IBR, 68.0% for BVD, and 91.9% for PI-3 are similar to other surveys.

Each of the herds tested had seropositive animals for each virus. These seropositive animals represented subclinical infections as these cattle had not received viral vaccines nor had specific disease caused by each virus been identified in each herd. There were significant differences between the herds for antibody prevalence rates to each virus. Possibly these differences reflect management such as animal concentration affecting transmission between animals or the genetic differences of the viruses affecting their ability to infect various cattle. In particular, 2 herds (C and E) had greater than half of the animals tested (60.0% and 51.0%) with IBR antibodies. This indicates that this virus in certain herds may infect a high percentage of the herd without causing illness.

A major consideration regarding these results is their use for vaccination recommendations. In IBR infections, neutralizing antibody in the serum (humoral response) is probably not the most accurate measurement of immunity (11). However, as cell-mediated immunity is not readily measured, serum antibody will be most likely used for IBR immunity considerations (11). Thus 65.1% of the cattle tested in this study were seronegative. Therefore, vaccination of these cattle would be indicated to prevent the major economic losses such as IBR respiratory disease and/or abortions.

Similarly, 32.0% of the cattle tested in this survey were seronegative for BVD. In one herd 50.0% of the cattle were seronegative. Thus, in these breeding herds considerations should be given to vaccinations to prevent BVD induced losses such as abortions and/or other fetal infections.

It would appear that a "herd immunity" to PI-3 resulting from natural infections exists in each herd tested with greater than 85.0% of the animals with PI-3 viral antibodies. Relatively few animals appear susceptible to PI-3 virus as indicated by the seronegative animals. Thus, losses due to respiratory and/or fetal infections would be minimal in this unvaccinated population.

Summary

Serums from five cattle herds in Louisiana were tested for antibodies to infectious bovine rhinotracheitis (IBR), bovine viral diarrhea (BVD), and parainfluenza-3 (PI-3) viruses. These cattle were free of obvious signs of illness and had not received IBR, BVD, or PI-3 viral vaccines previously. Of the 444 bovine serums tested; 155/444 (34.9%) had antibodies to IBR virus; 302/444 (68.0%) to BVD virus; and 404/444 (91.9%) to PI-3 virus.

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