Antibody Responses to IBR Vaccination in Feedlot Cattle

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Infectious Bovine Rhinotracheitis (IBR) is an important respiratory disease of cattle (3, 9, 13, 19). The disease has been especially critical in western feedlots and IBR virus infection has been associated with the shipping fever complex (8). Numerous commercial vaccines are in wide use to prevent this disease. Experimentally, these vaccines have been shown to be immunogenic and effective in preventing clinical IBR (10,23). A large western feedlot has routinely used a modified live virus IBR vaccine in all cattle entering the feedlot. Despite vaccination, the feedlot continued to experience significant morbidity and mortality associated with respiratory disease. A major portion of this respiratory disease was believed to be IBR virus infection (3).

This study was designed to study immunity to IBR virus of cattle as they enter the feedlot, and to measure the immune responses of feedlot cattle to intramuscular modified live virus IBR vaccines under field conditions. Immunity to bovine parainfluenza type 3 (PI_3) virus upon arrival at the feedlot and seroconversion to that virus as a result of natural infections was also studied.

Materials and Methods

Feeder cattle weighing approximately 350 kg were obtained from various western and midwestern sources. Routine management procedures within 2-5 days of entry included implantation in the ear of a growth stimulant, a total body dip in an organophosphate insecticide, and vaccination against IBR and leptospirosis. No other vaccines were given.

Twenty to 25 animals were randomly chosen from each of 30 incoming pens of cattle over a 5-month period from September to February. Each pen contained approximately 450 cattle. A blood sample for serum collection was taken from each subject after which it was vaccinated with a combination vaccine composed of an intramuscular modified live virus IBR and a killed *Leptospirosis pomona* bacterin. Three commercial vaccines [a-c] were randomly used. Two animals per pen were not vaccinated. Post vaccination blood samples were taken from 6 to 8 randomly chosen subjects and the two unvaccinated subjects per pen 21 or 35 days after vaccination. A total of 692 prevaccination and 88 post vaccination samples were obtained. The serums were stored at –20 C until tested.

Serum neutralizing (SN) antibodies against IBR virus were measured by the varying serum-constant virus method using the microtiter technique (16). Serums were heated at 56 C for 30 minutes. Serial 2-fold dilutions of serum were reacted with 25-30 $TCID_{50}$ of Colorado 1 IBR virus strain in plastic transfer plates and incubated 1 hour at room temperature. The mixture was then transferred to manolayers of bovine fetal spleen cells in microtiter tissue culture plates. The cell culture medium was supplemented by 10% lamb serum and antibiotics. The cells were examined for the presence of cytopathic effect 5 days after inoculation. Known positive and negative serums were included each time the test was done. Paired pre- and post vaccination serums were tested side by side on the same day to eliminate variation. Paired samples in which both serums had antibody titers less than 1/2 were retested with 9-10 $TCID_{50}$/0.025 ml of IBR virus.

Paired serum samples were also tested for hemagglutination inhibition (HI) antibodies against PI_3 virus. Heat-treated serums were diluted 1/2 and absorbed with kaolin to remove non-specific inhibitors. Two-fold dilutions of serum in V-shaped microtiter plates were reacted with 4 hemagglutinating units of PI_3 virus strain SP-4 (15) for one hour at room temperature. A 0.5% suspension of bovine red blood cells was added, and the test was read 3-4 hours later.

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[a] *IBR-Lepto, Bio Ceutic Laboratories, Inc., St. Joseph, MO.*
[b] *Rhino-Lep, Diamond Laboratories, Inc., Des Moines, IA.*
[c] *Jencine-IL, Jensen-Salsbery Laboratories, Kansas City, KA.*

Results

The results of SN antibody titers were grouped for comparison purposes into 4 relative classes: not demonstrable (<1/2), low (>1/2, <1/8), moderate (>1/8, <1/64, and high (>1/64). Prevaccination antibody titers against IBR virus were not demonstrable in 66% of the samples (455/692) (Figure 1). Twenty-six percent (184/692) had low titers. Only 8% (53/692) had moderate or high titers. Postvaccination antibody titers were generally much higher than prevaccination titers (Figure 2), but 20% (18/88) still had no demonstrable antibodies. Twenty-seven percent (24/88) had low, 31% (27/88) had moderate, and 22% (19/88) had high postvaccination titers.

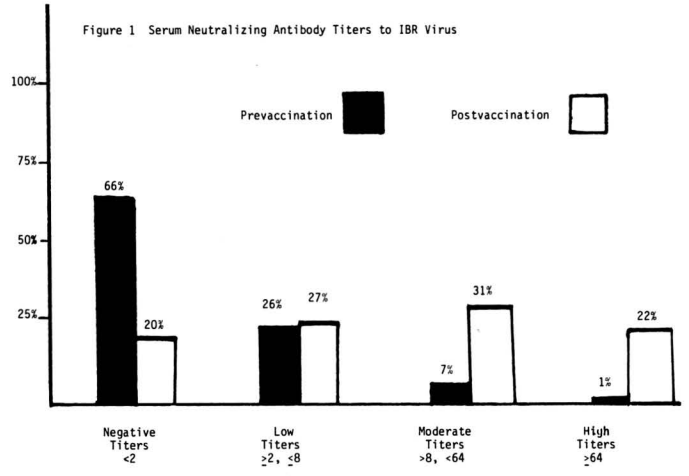

Figure 1 Serum Neutralizing Antibody Titers to IBR Virus

A distinct difference was noticed in antibody responses between animals that were seropositive and those that were seronegative prior to vaccination (Figure 2). Of the cattle with prevaccination titers > 1/2, 97% (36/37) had postvaccination increases in antibody titer. One animal had an antibody titer of 1/16 in both pre- and postvaccination serums. Some cattle with prevaccination antibody titers as high as 1/48 responded with even higher postvaccination titers. However, 35% (18/51) of the cattle with negative prevaccination titers did not develop demonstrable postvaccination SN antibody. There were no significant differences in antibody response among the three vaccines used.

Nine of 14 cattle that were not vaccinated had antibody titers to IBR virus. Twenty-one to 35 days later, 4 of 9 of these cattle had seroconverted to IBR virus and 5 of 9 remained negative. Of the 5 of 14 seropositive, unvaccinated cattle, 2 of 5 had 4-fold or greater increases in antibody titers and 3 of 5 remained unchanged in serums taken 21-35 days later.

Eighty-two percent (74/90) of incoming cattle had PI_3 HI antibody titers that did not increase during the sampling period. Seventeen percent (15/90) seroconverted to PI_3 virus. Only 1/90 animals had no HI antibody titer to PI_3 virus in serums taken 21-35 days after IBR vaccination.

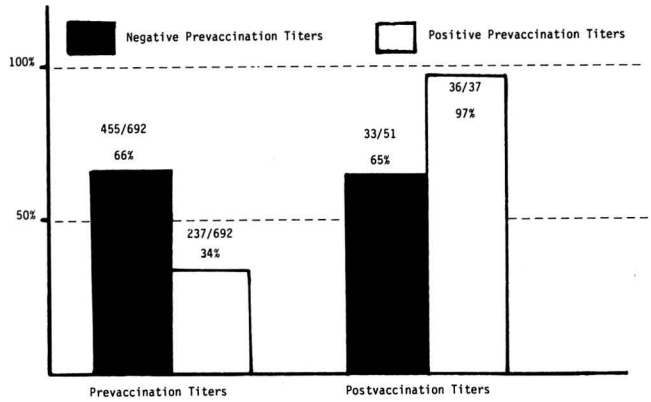

Figure 2 Relative Seroconversion: Positive vs. Negative Prevaccination Titers

Discussion

A relatively high percentage (35%) of seronegative cattle had no serological response to the IBR vaccines used as determined by the microtiter SN test. If extrapolated from the results of our test samples, 20% of the animals in the feedlot have no measureable antibody 21-35 days after vaccination. Various methods of measuring antibody to IBR virus include SN (16), indirect immunofluorescence (1), micro-modified direct complement fixation (5), microimmunodiffusion (12), indirect hemagglutination inhibition (11), plaque neutralization (7) and ELISA (14). The sensitivity of the SN test is considered high, but may be slightly less sensitive than indirect hemagglutination inhibition, plaque neutralization (4,20) or ELISA (14). We made our SN test as sensitive as possible by challenging negative serums with only 9-10 $TCID_{50}$ of IBR virus. Most systems used 25-100 $TCID_{50}$ (4).

Protection against clinical IBR cannot be assumed to be only antibody mediated. Recent studies have indicated that the cell-mediated immune response is extremely important in protection against IBR virus (17). We did not measure cell-mediated immune responses. However, antibody-dependent cell-mediated cytotoxicity has been shown to be a major defense mechanism against IBR virus (18,22).

The relatively poor serologic response to 3 IBR vaccines in this field study differs from studies done during vaccine development (10,23). The differences may not lie in the effectiveness of the vaccines, but in the time or condition of the animals at vaccination. It is important to note that nearly every animal (97%) with a positive prevaccination titer had increased postvaccination titers. This infers that the vaccine was both viable and immunogenic. The poor responses occurred only in seronegative animals. The stresses of shipping, con-comitant infections and other non-specific factors can render an animal temporarily immunologically unresponsive (21), increase interferon levels, or interfere in some way with the necessary replication of the modified live virus (6).

A large feedlot that acquires cattle from many sources has little choice but to vaccinate the cattle upon arrival even

though significant stress and concomitant disease factors are present at that time. Unless vaccination can occur in the absence of these stresses, we believe vaccination failures are important factors feedlot operators must consider. Presently there seems to be no general monetary or other incentive for the producers of feeder cattle to vaccinate calves at the appropriate time, then administer a booster 1 or 2 months prior to shipment to a feedlot. Our data indicated that many unvaccinated, seronegative cattle seroconverted to IBR virus soon after they entered the feedlot. Whether this seroconversion was due to virulent field virus or shedding vaccine virus is unknown. Clinical IBR was confirmed in vaccinated feedlot cattle in another study (2). The vast majority (82%) of animals were already immune to PI_3 virus when they entered the feedlot. Within 3-5 weeks, all but one susceptible test animal had seroconverted to this virus, indicating its high prevalence in feedlot cattle as well.

Summary

Prevaccination serums were obtained from 692 feeder cattle upon arrival at a major western feedlot and tested for antibodies to IBR virus. Sixty-six percent had no detectable serum neutralizing antibodies, 27% had low titers and 8% had moderate to high titers. The animals were vaccinated with an intramuscular modified live virus vacine, and serums were taken from 88 of these subjects, 3 or 5 weeks after vaccination and tested for serum neutralizing antibodies to IBR virus. Ninety-seven percent of the cattle which had positive titers to IBR virus prior to vaccination developed 4-fold or greater increases in antibody titers after vaccination. However, 35% of cattle with negative prevaccination titers did not develop demonstrable post vaccination antibodies. Of the 88 animals for which both prevaccination and post vaccination serums were compared, 20% remained negative, 27% had low, 31% had moderate and 22% had high serum neutralizing antibodies.

Seventy-four of 90 (82%) incoming cattle had positive antibody titers to PI_3 virus. Three or five weeks later all but 1 of the 16 susceptible cattle had seroconverted to PI_3 virus even though PI_3 vaccine had not been administrated.

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