Pfizer Animal Health
Technical Bulletin

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# *CattleMaster*® *GOLD*™ induces significant fetal protection against IBR-induced abortion

**PfizerAnimal Health**
**Pfizer Inc**
**New York, NY 10017**

## Key Points

- An efficacy study was conducted to evaluate the ability of prebreeding vaccination with *CattleMaster*® *GOLD*™ *FP*® *5* to protect pregnant heifers against abortion and stillbirths caused by intravenous challenge with virulent IBR virus at approximately 6 months of gestation.[1]
- Laboratory diagnostic tests demonstrated microbiologic and/or histopathologic evidence of IBR virus-induced abortion in 92% of fetuses from control heifers and in only 8% of fetuses from heifers vaccinated with *CattleMaster GOLD.*
- The temperature-sensitive strain of IBR virus in *CattleMaster GOLD* has the safety characteristics of inactivated IBR vaccinal virus with the immunogenicity of modified live IBR vaccinal virus.

Reproductive problems caused by the infectious bovine rhinotracheitis (IBR) virus include temporary infertility, abortion, and reproductive tract infections in both male and female cattle.[2] The greatest economic impact comes from losses resulting from IBR abortions, which occur chiefly during the last half of gestation, often without evidence of other clinical signs.[3] The estimated 25% of susceptible cows aborting due to IBR virus demonstrate significant loss, in terms of both valuable genetic potential as well as market value.[4] In beef herds, abortions usually occur during a 3- to 4-month period when 50% or more of the cows may abort.[5] In dairy herds, IBR virus-induced abortion storms can last up to a year or more, affecting profits through calf death loss, loss of genetic potential, and failure to reach peak lactations.[5]

Introduced in July 2004, *CattleMaster GOLD* is the first killed BVD vaccine licensed in the U.S. that has label claims for use as an aid in the prevention of IBR abortion and BVDV persistent infection (Types 1 and 2). Presented here is a summary of the *CattleMaster GOLD* efficacy study demonstrating fetal protection against IBR abortion.

## Study Overview: IBR Fetal Protection

### *Test Vaccine*

Immune responses to the IBR virus were stimulated by a vaccine

(*CattleMaster GOLD FP 5)* formulated to contain only the minimum immunizing dose (MID) of the IBR vaccine antigen. MID levels are established prior to licensing of a vaccine and reflect an antigen level that would likely be present at product expiration. By using MID levels for challenged antigens, investigators put the challenge antigen at its maximum potential disadvantage. When vaccine withstands challenge under these trying circumstances, it will likely be at least as effective when antigen level is at a normal release level. Commercial vaccine lots have substantially higher release titers to ensure potency throughout the product shelf life.

### *Study Design*

A test population of IBR virus seronegative heifers (approximately 16 months of age) were vaccinated either with *CattleMaster GOLD FP 5* or a placebo at 5 and 2 weeks prior to breeding (Days 0 and 21, respectively). Following estrus synchronization, all test heifers were bred by artificial insemination with certified IBR and BVD virus-negative semen. At approximately 60 days of gestation, pregnancy status was confirmed by means of rectal palpation. Following the first 6 months of gestation, 25 heifers confirmed to be pregnant were selected for challenge. At 216 days after administration of the first vaccine dose, a 2 mL challenge dose of IBR virus (Cooper strain) was administered intravenously to each pregnant heifer (12 controls, 13 vaccinates). Tissues from aborted and stillborn calves and calves dying neonatally were collected for laboratory evaluation for potential causes of abortion including IBR virus.

### *Data Analysis*

Criteria for a satisfactory challenge were demonstrated by a clinically significant number of controls that aborted, had stillborn calves, or delivered weak calves attributable to IBR virus infection. Attribution of the cause of abortion, stillbirth, or weak calves was made by investigators at the University of Nebraska Veterinary Diagnostic Center, Lincoln, Nebraska. Demonstration of immunity resulting from vaccination with *CattleMaster GOLD FP 5* was based on a significant reduction in the frequency of abortions, stillbirths, and/or weak calves attributable to IBR virus infection in vaccinates as compared to controls.

Pfizer Biometrics and Data Management was responsible for all data summaries and analyses. Clinical observations and rates of abortion, stillbirths, and weak calves were summarized in frequency tables. Visual analog scales scoring calf weakness were summarized and mean and standard deviation calculated. Rectal temperatures, virus isolations, and serum virus neutralization end point titers were summarized. Rates of abortion, stillbirths, and weak calves were analyzed and means were compared using Fisher's Exact Test. Rectal temperatures and end point titers for heifers were analyzed using a mixed linear model with repeated measures. *A priori* contrasts were constructed to compare treatment means. The 5% level of significance ($P \leq 0.05$) was used to assess statistical differences.

## Results

### *Serology*

As Figure 1 shows, *CattleMaster GOLD* vaccinates had significantly ($P \leq 0.05$) higher IBR virus serum neutralizing (SVN) geometric mean titers (GMTs) at all sampling days after vaccination. All placebo controls remained seronegative until challenge day

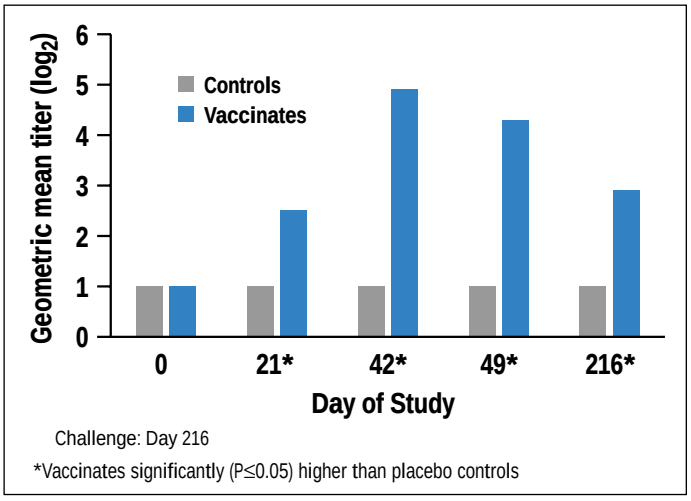



**Figure 1 –** Postvaccination IBR virus serum virus neutralization titers

(Day 216). On Day 230 (14 days after challenge), the IBR virus SVN GMTs in all treatments increased over prechallenge titers, indicating a serologic response to challenge (data not shown).

### *Clinical and Laboratory Observations*

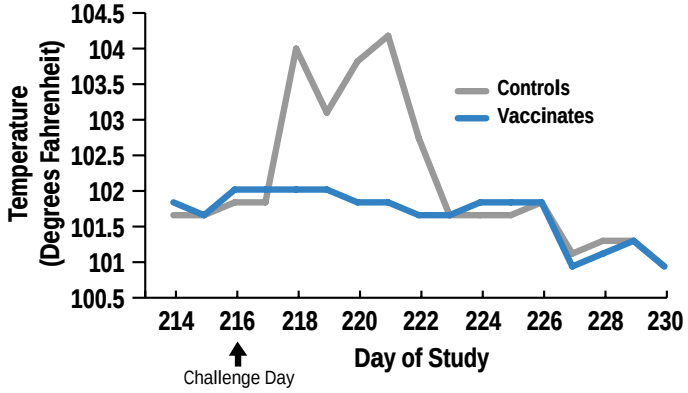
Clinical observations revealed a greater numerical frequency of nasal and ocular discharge in controls and a slightly greater nonsignificant incidence of coughing in vaccinates (Table 1). Controls had significantly ($P \leq 0.05$) higher rectal temperatures than vaccinates on five consecutive days (Days 218-222) following challenge on Day 216 (Figure 2).

Summarized in Tables 2 and 3 are additional clinical, microbiologic, and histopathologic findings recorded after IBR virus challenge. No heifer deaths occurred during the study. There was a significantly ($P \leq 0.05$) lower frequency of abortions and stillbirths attributed to IBR in vaccinated (8.0%) compared to control heifers (92.0%). Eleven of 12 pregnant control heifers aborted between 19 and 110 days after IBR virus challenge and all 11 showed fetal microbiologic and/or histopathologic evidence of IBR virus. Abortions in controls occurred in two peaks, the first at 19 to 28 days after challenge and the second at 57 to 60 days after challenge. Only 1 of 13 vaccinated heifers aborted after challenge (Day 237), and only this aborted fetus yielded IBR virus on virus isolation. One additional vaccinated heifer produced a stillborn calf as a result of dystocia. Diagnostic investigation of this calf demonstrated no evidence of IBR virus. All laboratory tests for other abortifacients (BVDV, *Leptospira*, *Campylobacter fetus*, *Neospora*, bacterial agents (nonspecific), and fungal agents (nonspecific) from aborted and stillborn calves, or calves dying neonatally, were negative (data not shown).

The comparison of weak calves between treatments did not have a sufficient sample size to allow analysis because only one viable control calf was available for assessment.

## Conclusion and Discussion

Efficacy of prebreeding vaccination with *CattleMaster GOLD FP 5* was confirmed on the basis of a significant ($P \leq 0.05$) reduction in the number of aborted, stillborn and/or weak calves attributable to IBR virus




*Controls had significantly ($P \leq 0.05$) higher rectal temperatures than vaccinates on Days 218-222.

**Figure 2 –** Least squares mean rectal temperatures

**Table 1–Clinical observations (percent positive for specific clinical signs)**

| Vaccine | No. Animals | Nasal Discharge | Ocular Discharge | Abnormal Respiration | Coughing | Vaginal Discharge |
|---|---|---|---|---|---|---|
| Placebo | 12 | 3.4% | 0.5% | 0% | 0.5% | 0% |
| *CattleMaster GOLD* | 13 | 0.9% | 0% | 0% | 0.9% | 0% |

**Table 2–Clinical responses in vaccinates and controls following challenge with IBR virus**

| | Reproductive Outcome—No. Calves | | | |
|---|---|---|---|---|
| Vaccine | Normal* | Aborted† | Stillborn | Weak‡ |
| Placebo | 1/12 | 11/12 | 0/12 | ND§ |
| *CattleMaster GOLD FP 5* | 11/13 | 1/13 | 1/13** | 0/11 |

*Normal = calves not stillborn or aborted
†Aborted = heifers giving birth to fetuses
‡Weak = calves considered weak on clinical assessment
§Not determined; sample size (1 viable calf born to a control heifer) was insufficient to allow statistical analysis.
**Non IBR virus-associated stillbirth (dystocia) not counted as an IBR-induced abortion.

**Table 3–Summary of laboratory responses in vaccinates and controls following challenge with IBR virus at 216 days after vaccination**

| | Laboratory Outcome—No. Calves | | | | |
|---|---|---|---|---|---|
| Vaccine | IBR virus positive | Histopath lesions | IBR virus abortion* | Clinical IBR abortion | IBR abortion, stillbirth weak calf |
| Placebo | 5/12 | 11/12 | 11/12 | 11/12 | 11/12 (92.0%) |
| *CattleMaster GOLD FP 5* | 1/13 | 1/13 | 1/13 | 1/13[†] | 1/13 (8.0%[a]) |

*Based on laboratory results only
[†]Based on laboratory results.
[a]Significant ($P < 0.05$) difference versus control group.

infection in vaccinates as compared to controls. Vaccination protected 92.0% of IBR seronegative heifers against abortion and stillbirth caused by a virulent IBR virus challenge (at 216 days postvaccination) that induced abortion in 92.0% of placebo-vaccinated control heifers. Calf weakness was not analyzed because of the low number of viable calves born to control heifers.

Results of the study reported here corroborate those of an earlier study in which the same IBR component was administered prior to artificial insemination of 10 IBR seronegative heifers.[6] Between 177 and 187 days of gestation, the 10 vaccinates plus 10 seronegative nonvaccinated pregnant heifers were challenged intravenously with $10^{6.0}$ $TCID_{50}$ Cooper strain IBR virus. The difference in number of abortions or stillbirths between vaccinates (1/10) and controls (10/10) was significant ($P < 0.05$).

Currently, no other IBR vaccine containing killed BVD antigen licensed in the U.S. has label claims for protecting pregnant cows and heifers against IBR-induced abortion. Induction of fetal protection with inactivated IBR virus stimulates inadequate cell-mediated immune responses, which are thought to play the pivotal role in stopping cell-to-cell spread of IBR virus.[7] The temperature sensitive (ts) modified live virus (MLV) strain of IBR virus used in *CattleMaster GOLD FP 5* circumvents this shortcoming. The ts IBR strain undergoes limited replication following administration, and thus shares the following immunogenic properties attributed to other attenuated vaccine viruses:[8-10]

• Stimulation of interferon production that inhibits viral replication[9]

• Activation of both CMI and humoral antibody responses[8-10]

• Generation of protective levels of memory T-cells[8]

Protection against the consequences of IBR virus infection involves complex interaction between the humoral and CMI branches of the immune system, which are both activated by vaccination. The CMI responses, however, are of prime importance as the IBR virus spreads from cell to cell by way of intracellular bridges and is not exposed to antibody circulating in the bloodstream.[7] With IBR virus, quick, strong CMI responses to disease challenge help avoid costly clinical signs and abortions in cows.

The restricted growth temperature of the ts IBR virus also assures the vaccine strain's safety for use in pregnant cows and heifers. In *in utero* testing, the vaccinal virus was inoculated directly into fetuses and the amniotic fluid of pregnant cows. Of 9 fetuses, only one responded serologically to the IBR vaccine strain and was aborted 3 weeks after inoculation. Virus was not isolated from any fetus.[11] Additional studies confirmed that the IBR strain did not shed or spread following administration[12-14] nor produce latency or re-excretion of virus following immunosuppression.[15,16]

Considered together, the results of safety and efficacy studies conducted with the ts IBR virus used in *CattleMaster GOLD* confirm that the vaccinal strain has the safety of inactivated IBR antigens while retaining the immunogenicity characteristic of MLV antigens.

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