46R0002

The time course required for the development of disease is sufficiently long that factors that adversely affect animal appetite, mobility and other animal behaviors such as anesthetics, analgesic and tranquilizers would adversely affect experimental outcomes and would likely decrease animal survival from the infectious challenge. This is particularly true with regard to this study as the pigs are new-borns and must learn to drink from feed bowls. Medication that reduces alertness will attenuate the learning process and adversely affect the animal as well as the study. Medications that reduce intestinal distress physiologically would make experimental outcomes non-interpretable.

46R0002

Column E Explanation

1. **Registration Number:** 96-027 - [redacted] (b)(6), (b)(7)(C)
2. **Number of animals used in this study:** 2
3. **Species of animals used in this study:** Bovine - calves
4. **Explain the procedure producing pain and/or distress.**

Calves were orally inoculated with a virulent strain of bovine coronavirus. This virus replicates in the cells lining the small intestine and colon of the calf. The result of virus replication is the destruction of these cells and the induction of a malabsorptive diarrhea. The diarrhea develops within 48 hours after inoculation and persists for up to 7 days. Calves are monitored daily of signs of depression, fever, dehydration and loss of bodily flesh. Calves that appear to becoming severely dehydrated are treated with an oral electrolyte solution to reduce potential of death.

5. **Provide scientific justification why pain and/or distress could not be relieved. State methods or means used to determine that pain and/or distress relief would interfere with test results.**

The calves were allowed to experience the typical disease induced by inoculation with the calf coronaviruses. Our goal was to induce diarrhea in these calves, which will also result in the shedding of coronavirus particles in the feces. This was necessary to provide the investigators with fecal samples from a known calf infected with coronavirus. These known samples are then used as positive controls in the development of a new diagnostic assay for the detection of bovine coronavirus. We could not interfere with the development of the disease because this would eliminate the shedding of virus in the fecal samples. We did not intend for calves to become moribund as the result of this disease, only to develop diarrhea for a limited period of time to allow for the collection of coronavirus contaminated feces.