Category E Explanation
Registration Number: 33-R-0008
Customer No. 694

In a preclinical study to evaluate the efficacy of antiviral agents, a total of 18 ferrets received influenza virus and experienced flu-like symptoms. Symptoms included fever, inappetence, weight loss, vomiting, and/or diarrhea. Ferrets are observed at least twice daily to identify flu-like symptoms and to identify animals that should be euthanized due to the development of a moribund state (as defined by Standard Operating Procedures). Analgesic agents cannot be administered in these studies for the following reasons:

- Narcotic analgesics can cause histamine release (Soma, 1983, Nemzek, *et al.*, 2008) and induce respiratory depression that could alter the pathogenic and clinical response to infection. These effects of narcotic analgesics could interfere with (and may invalidate) the evaluation of antiviral activity.
- Nonsteroidal anti-inflammatory drugs can also alter the pathogenesis of infection and clinical responses to vaccines or anti-infective agents. Such effects could also invalidate evaluations of vaccine or anti-viral potency.

Prior to the initiation of these studies, a literature search was conducted to identify possible alternative models for the testing of antiviral agents. No suitable models were identified that could replace the current *in vivo* antiviral/vaccine efficacy study that includes clinical evidence of disease and possible moribundity as critical experimental endpoints.

Category E Explanation
Registration Number: 33-R-0008
Customer No. 694

In a preclinical toxicology study of a novel small molecule therapeutic drug, thirty rabbits demonstrated clinical evidence of toxicity as a result of exposure to the test article. Toxic effects included weight loss, hypoactivity and/or neurologic signs. Rabbits were observed at least twice daily throughout the studies to identify toxic effects and to identify animals that should be euthanized due to the development of a moribund state (as defined by institutional Standard Operating Procedures). All rabbits underwent a complete necropsy with tissue collection; a full set of tissues from each animal was evaluated histopathologically. Data from these studies were critical parameters supporting the selection of appropriate doses of these therapeutic agents for use in clinical trials, and in identifying sensitive target organs that should be monitored in those trials.

Analgesic agents cannot be administered in such studies for the following reasons:

- Many common analgesics (e.g., non-steroidal anti-inflammatory drugs) can modulate the activity of enzymes involved in drug metabolism, with resulting effects on agent Absorption, Distribution, Metabolism, and/or Excretion (ADME). For this reason, co-administration of analgesic agents in a preclinical toxicology study may have a significant impact on agent toxicity with resulting influences on study results.
- Administration of analgesics may mask clinical signs of toxicity, and thereby reduce the sensitivity of the test system to identify toxicologic effects of mild to moderate severity.

The animal species and group numbers used in these studies were selected to meet published FDA and ICH standards for the design of preclinical safety assessments of novel therapeutic agents. The use of non-rodent test systems is mandated by U.S. and international regulatory agencies, and no *in vitro* alternatives are accepted by these agencies. Furthermore, literature searches performed prior to the initiation of these studies were not successful in identifying alternate model systems that are both (a) scientifically rigorous and (b) acceptable to the FDA and other regulatory agencies.

Category E Explanation
Registration Number: 33-R-0008
Customer No. 694

In a preclinical toxicology study of a novel small molecule therapeutic drug, three dogs demonstrated clinical evidence of toxicity as a result of exposure to the test article. Toxic effects included weight loss, vomiting, decrease in appetite, and/or neurologic signs. Dogs were observed at least twice daily throughout the studies to identify toxic effects and to identify animals that should be euthanized due to the development of a moribund state (as defined by institutional Standard Operating Procedures). All dogs underwent a complete necropsy with tissue collection; a full set of tissues from each animal was evaluated histopathologically. Data from these studies were critical parameters supporting the selection of appropriate doses of these therapeutic agents for use in clinical trials, and in identifying sensitive target organs that should be monitored in those trials.

Analgesic agents cannot be administered in such studies for the following reasons:

- Many common analgesics (e.g., non-steroidal anti-inflammatory drugs) can modulate the activity of enzymes involved in drug metabolism, with resulting effects on agent Absorption, Distribution, Metabolism, and/or Excretion (ADME). For this reason, co-administration of analgesic agents in a preclinical toxicology study may have a significant impact on agent toxicity with resulting influences on study results.
- Administration of analgesics may mask clinical signs of toxicity, and thereby reduce the sensitivity of the test system to identify toxicologic effects of mild to moderate severity.

The animal species and group numbers used in these studies were selected to meet published FDA and ICH standards for the design of preclinical safety assessments of novel therapeutic agents. The use of non-rodent test systems is mandated by U.S. and international regulatory agencies, and no *in vitro* alternatives are accepted by these agencies. Furthermore, literature searches performed prior to the initiation of these studies were not successful in identifying alternate model systems that are both (a) scientifically rigorous and (b) acceptable to the FDA and other regulatory agencies.