Optional Column E Explanation Form 55R0005

This form is intended as an aid to completing the Column E explanation. It is not an official form and its use is voluntary. Name, addresses, protocols, veterinary care programs, and the like, are not required as part of an explanation. A Column E explanation must be written so as to be understood by lay persons as well as scientists.

1. Registration Number: **55-R-0005**

2. Number _____99_____of animals used in this study

3. Species (common name)_____Rabbit_____of animals used in this study

4. Explain the procedure producing pain and/or distress.

Under proparacaine-induced topical anesthesia, one eye of rabbits is intravitreally injected with a test reagent (LPS; 10ng in 10-50 µl) approximately 3 mm posterior to the limbus, an area containing very few nerve endings. Injections are made with a 30-guage needle attached to a glass Hamilton syringe. In some cases, at the time an experimental agent may be instilled or intravitreally injected (total volume of 50µl). Animals are monitored for evidence of inflammation at various time intervals with a slit lamp (scores based on iridal hyperemia and aqueous flare) to determine the appropriate time for termination (up to 48 hours). Animals are terminated by exposure to a saturated CO_2 atmosphere, eyes removed, ocular fluids aspirated, and tissues processed for histologic/molecular analyses.

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 (For Federally mandated testing, see question 6 below)

The use of opioid analgesics might alter the course of inflammation that we are studying. The use of additional animals to evaluate these substances would need to be included as controls. Investigators in the literature utilizing this same animal model do not use opiod analgesics in their studies. The use of anti-inflammatory drugs such as corticosteriods and nonsteroidal drugs would negate our studies, since the purpose of our research is to study the inflammatory response in the eye. Also, the dose of endotoxin that we inject (10 ng) is low compared to most reports in the literature (10-100mg), and induces a moderate inflammatory response. The animals continue to eat and do not become photophobic.

6. What, if any, Federal regulations require this procedure? Cite the agency, the Code of Federal Regulations (CFR) title number and the specific section number (e.g., APHIS, 9 CFR 113.102):

Agency_____N/A_____CFR____N/A_____

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1. Registration Number: **55-R-0005**

2. Number _____4_____of animals used in this study

3. Species (common name)_____Pigs_____of animals used in this study

4. Explain the procedure producing pain and/or distress.

4 pigs were experimentally infected with either Salmonella typhi or Salmonella choleraesuis. Those infected with S. choleraesuis (3) developed signs of salmonellosis. Those infected with S. typhi showed no signs of distress.

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 (For Federally mandated testing, see question 6 below)

Experimental infections were employed to better understand a disease process in a normal animal. This included signs of infection and disease. Consequently we could not alleviate pain or distress without jeopardizing understanding. Also, the action of some analgesics would alter the normal course of the actual infection. By examining relatively few animals for signs of infection, we were able to draw very confident conclusions as to the sustability of pigs for modeling disease caused by S. typhi.

6. What, if any, Federal regulations require this procedure? Cite the agency, the Code of Federal Regulations (CFR) title number and the specific section number (e.g., APHIS, 9 CFR 113.102):

Agency_____N/A_____CFR_____N/A_____