

**Column E Explanation**

1. Registration Number
  - a. 51-F-0024
2. Number of animals used in this study.
  - a. Column E = 36
3. Species of animals used in this study
  - a. Guinea Pig
4. Explain procedure producing pain and/or distress.
  - a. Guinea pigs will be inoculated with a mixture of Diphtheria Toxin (one concentration) and Diphtheria Antitoxin (four concentrations) in order to measure the amount of neutralization conferred by the antitoxin. Guinea pigs inoculated with lower amounts of antitoxin in the Toxin/Antitoxin mixtures will begin to display signs of Diphtheria Toxin related toxicity between 24-96 hours post inoculation.
5. Attach or include an explanation with the reason/s for why anesthetics, analgesics and tranquilizers could not be used.
  - a. The onset of symptoms related to Diphtheria Toxin toxicity is used to determine the potency of the antitoxin. As such, anesthetics, analgesics and tranquilizers cannot be used as they might mask the onset of the clinical symptoms thereby rendering the assay useless.
6. What, if any, federal regulations require this procedure?
  - a. FDA/NIH Minimum Requirements for Diphtheria Antitoxin and Diphtheria Toxoid.