

Column E Explanation

This form is intended as an aid to completing the column E explanation. It is not an official form and its use is voluntary. Names, addresses, protocol, 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: 23-R-0021
2. Number of Category E animals: 79
3. Species: Ferret

4. **Explain the procedure producing pain and or distress:** Ferrets in this study were inoculated with wild-type or recombinant wild-type Influenza viruses. Animals inoculated with these viruses will display clinical signs including weight loss and inactivity.

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.** Treatment of these animals with analgesics or anti-inflammatory agents will mask clinical signs, alter behavior, and interfere with viral replication and transmission; therefore, it is not possible to relieve pain or distress resulting from influenza infection. However, the investigator, in consultation with the veterinarian and with the approval of the IACUC, have established humane endpoints intended to relieve pain and/or distress of animal that become severely ill.

6. **What, if any, federal regulations require this procedure?** This procedure is the focus of the PI's research and not required by law.

1. Registration Number: 23-R-0021
2. Number of Category E animals: 20
3. Species: Ferrets

4. **Explain the procedure producing pain and or distress:** Ferrets in this study were inoculated with wild-type wild-type viruses or exposed to inoculated ferrets in standard cages or transmission cages. At this time, we do not know if SARS-CoV-2 will cause clinical illness in ferrets. However, given that this virus can cause severe disease in humans, clinical disease could include weight loss, lethargy, fever, labored breathing, and animals could develop severe disease that meets endpoint criteria.

5. **Provide scientific justification why pain and/or distress could not be**

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relieved. State methods or means used to determine that pain and/or distress relief would interfere with test results. Treatment of these animals with analgesics or anti-inflammatory agents will mask clinical signs, alter behavior, and interfere with viral replication and transmission; therefore, it is not possible to relieve pain or distress resulting from virus infection.

6. **What, if any, federal regulations require this procedure?** This procedure is the focus of the PI's research and not required by law.

1. Registration Number: 23-R-0021
2. Number of Category E animals: 80
3. Species: Hamsters

4. **Explain the procedure producing pain and or distress:** Hamsters in this study are inoculated with wild-type influenza viruses or exposed to inoculated hamsters in standard cages. Depending upon the virus strain, influenza viruses can cause clinical disease in hamsters with weight loss and ruffled fur.

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.** Treatment of these animals with analgesics or anti-inflammatory agents will mask clinical signs, alter behavior, and interfere with viral replication; therefore, it is not possible to relieve pain or distress resulting from virus infection.

6. **What, if any, federal regulations require this procedure?** This procedure is the focus of the PI's research and not required by law.

1. Registration Number: 23-R-0021
2. Number of Category E animals: 16
3. Species: Hamsters

4. **Explain the procedure producing pain and or distress:** Hamsters in this study will be inoculated with wild-type viruses or exposed to inoculated hamsters in standard cages. SARS-CoV-2 does cause clinical disease in hamsters with weight loss and ruffled fur.

5. **Provide scientific justification why pain and/or distress could not be relieved. State methods or means used to determine that pain and/or**

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distress relief would interfere with test results. Treatment of these animals with analgesics or anti-inflammatory agents will mask clinical signs, alter behavior, and interfere with viral replication and transmission; therefore, it is not possible to relieve pain or distress resulting from virus infection.

6. **What, if any, federal regulations require this procedure?** This procedure is the focus of the PI's research and not required by law.

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