

**Column E explanation****1. Registration Number**

21-R-0119

**2. Number of animals used in this study**

74

**3. Species (common name) of animals used in this study***Mustela furo* (ferret)**4. Explain the procedure producing pain and distress.**

Animals receive intranasal inoculation of SARS-CoV-2 virus in order to establish a respiratory model of SARS-CoV-2 infection in ferrets. Minimum infectious dose and pathogenesis studies are conducted in which the infection runs its course unless various intervention criteria are reached and/or animals are euthanized for histology and other ex-vivo analysis. All animals that are infected with SARS-CoV-2 on this protocol are placed in category E because there is a chance that animals may develop a fever or other adverse symptoms between daily checks.

**5. Provide a 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 overall goal of this study is to develop mustelid models for COVID-19 and to establish clinical, virological, and immune-pathological parameters associated with acute SARS-CoV-2 infection in this animal model. Additionally, the pathogenesis of SARS-CoV-2 will be compared in ferrets and mink. Ferrets have proven to be effective models in prior human virology and immunology studies and mink have been shown to catch and develop COVID-19 symptoms on mink farms. The study of COVID-19 in animal models of the human immune system is of great importance due to the loss of life, economic, social, and educational impact of SARS-CoV-2 pandemic over the past ~2 years. Since the goal of this study is to investigate the pathogenesis of SARS-CoV-2 in ferrets with the goal of determining disease onset and duration, no clinical intervention will be performed in the animals. Humane euthanasia will, however, be performed should the intervention points listed above be noted in the animals.

**Column E explanation****1. Registration Number**

21-R-0119

**2. Number of animals used in this study**

14

**3. Species (common name) of animals used in this study***Neogale vison* (mink)**4. Explain the procedure producing pain and distress.**

Animals receive intranasal inoculation of SARS-CoV-2 virus in order to establish a respiratory model of SARS-CoV-2 infection in mink. Minimum infectious dose and pathogenesis studies are conducted in which the infection runs its course unless various intervention criteria are reached and/or animals are euthanized for histology and other ex-vivo analysis. All animals that are infected with SARS-CoV-2 on this protocol are placed in category E because there is a chance that animals may develop a fever or other adverse symptoms between daily checks.

**5. Provide a 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 overall goal of this study is to develop mustelid models for COVID-19 and to establish clinical, virological, and immune-pathological parameters associated with acute SARS-CoV-2 infection in this animal model. Additionally, the pathogenesis of SARS-CoV-2 will be compared in ferrets and mink. Ferrets have proven to be effective models in prior human virology and immunology studies and mink have been shown to catch and develop COVID-19 symptoms on mink farms. The fact that the related mink species shows natural ability to contract SARS-CoV-2 suggests that it may be a viable model as well, if not only for comparative studies. The study of COVID-19 in animal models of the human immune system is of great importance due to the loss of life, economic, social, and educational impact of SARS-CoV-2 pandemic over the past ~2 years. Since the goal of this study is to investigate the pathogenesis of SARS-CoV-2 in mink with the goal of determining disease onset and duration, no clinical intervention will be performed in the animals. Humane euthanasia will, however, be performed should the intervention points listed above be noted in the animals.