collection of information unless it displays a valid OMB control number. The valid OMB control num 0579-0036. The time required to complete this information collection is estimated to average ,5 hou	irs per response, including the time for	0579-0036	
reviewing instructions, searching existing data sources, gathering and maintaining the data needed collection of information	I, and completing and reviewing the	Interagency Report Control No. 0180-DOA-AN	
		Fiscal year: 2022	
(TYPE	ALTH INSPECTION of Research Faci Explanation	SERVICE lity	
This information is required by law (7 U.S.C. 2143 and 9 C.F.R. §2.36). Failure to report according to the regulations can result in an order to cease and desist.			
1. REGISTRATION NUMBER	2. Research Facil	lity Headquarters address	
74-R-0081		mp Bowie Blvd orth,Texas 76107	
<b>3. Number of animals used in the study.</b> 27	4. Species (common name) of animals used in the study. Hamster		
<b>5. Explain the procedure producing pain and c</b> Animals associated with this model may experience pain or distress as a molten do not manifest any signs of pain or distress prior to death, and 755 have been euthanized to prevent prolonged suffering. Signs that indicate tetharmy acuta/chronic diarrhea humothermin >15% docraase in body with the sufference of the sufferenc	% of the animals in those stud a severe disease state as a res	ies have either remained healthy and normal or ult of Clostridioides difficile infection include	
Animals associated with this model may experience pain or distress as a soften do not manifest any signs of pain or distress prior to death, and 759 have been euthanized to prevent prolonged suffering. Signs that indicate lethargy, acute/chronic diarrhea, hypothermia, >15% decrease in body w possible effects related to the infection, treatment with test articles (comparticle related effects include acute paralysis, increased respiratory/heart evaluated in this model are pre-screened for potential toxicity by the spor for evaluating other test articles in this model. All animals associated with this protocol will be monitored for severe dis clinical observation sheet for each day of a study will be maintained that (day and time) and personnel initials. Using the guidelines associated wi	% of the animals in those stud e a severe disease state as a rest eight, reduced water/food inta pounds) could also induce neg rates, and a rough coat. To pr nsors and selected dosing regi ease every 6 to 8 hours throug will include recording observa- th the IACUC 'Pain and Disco	ies have either remained healthy and normal or ult of Clostridioides difficile infection include ake, and swollen peritoneum. In addition to the pative effects in an animal as well. Signs of test revent treatment related effects, all compounds mens will be based on what has been safely used whout the duration of a study (up to 37 days). A ations and animal disposition, including the dat comfort' (document 013) and 'Humane	
Animals associated with this model may experience pain or distress as a soften do not manifest any signs of pain or distress prior to death, and 755 have been euthanized to prevent prolonged suffering. Signs that indicate lethargy, acute/chronic diarrhea, hypothermia, >15% decrease in body we possible effects related to the infection, treatment with test articles (comparticle related effects include acute paralysis, increased respiratory/heart evaluated in this model are pre-screened for potential toxicity by the spor for evaluating other test articles in this model. All animals associated with this protocol will be monitored for severe disclinical observation sheet for each day of a study will be maintained that (day and time) and personnel initials. Using the guidelines associated wite Endpoints' (document 008) protocols, any animal that appears to be more <b>6. Provide the scientific justification for not protocol second seco</b>	% of the animals in those stud a severe disease state as a resi- eight, reduced water/food inta- bounds) could also induce neg- rates, and a rough coat. To pri- nsors and selected dosing regi- ease every 6 to 8 hours throug will include recording observa- th the IACUC 'Pain and Disco- ibund will be immediately rer <b>oviding the approp</b> <b>re the animal exper</b> ossible drug interactions have not been fu- in this model. For example, buyenenphin t include macrolides (clarithromycin), ri- ig, which can directly impact their effica- vio-2C9) pathway, which also metabolical v can impact the efficacy of each drug by y intestinal infections. Given these facts a Gudin J et al., 2012; Preissner S et al., 2012 and/or prophylactic efficacy of a new inv for treating life-threatening Clostridioide . Dec 1; 44(6): S4-S14.	ies have either remained healthy and normal or ult of Clostridioides difficile infection include ake, and swollen peritoneum. In addition to the gative effects in an animal as well. Signs of test revent treatment related effects, all compounds mens will be based on what has been safely used shout the duration of a study (up to 37 days). A ations and animal disposition, including the date omfort' (document 013) and 'Humane moved from the study and euthanized. <b>Driate anesthetics, analgesics,</b> <b>rienced accompanying pain or</b> hely determined. If another drug is used to relieve pain and/or ne (analgesic) is metabolized by the cytochrome 450-3A4 famycins (rifampin), and fluoroquinolones (ciprofloxacin), y within the host (animal or human). Additionally, several ly interacts with the antibiotics metronidazole and rifampin. As is allering their metabolism and pharmacokinetics in the host. In about documented drug interactions within critical metabolic 2), it is imperative that non-study associated drugs used to relieve restigational compound can be accurately evaluated. If a new s difficile-associated diarrhea (CDAD) in humans, References:	
Animals associated with this model may experience pain or distress as a soften do not manifest any signs of pain or distress prior to death, and 755 have been euthanized to prevent prolonged suffering. Signs that indicate lethargy, acute/chronic diarrhea, hypothermia, >15% decrease in body w possible effects related to the infection, treatment with test articles (comparticle related effects include acute paralysis, increased respiratory/heart evaluated in this model are pre-screened for potential toxicity by the spor for evaluating other test articles in this model. All animals associated with this protocol will be monitored for severe dis clinical observation sheet for each day of a study will be maintained that (day and time) and personnel initials. Using the guidelines associated with Endpoints' (document 008) protocols, any animal that appears to be more <b>6. Provide the scientific justification for not pr</b>	% of the animals in those stud e a severe disease state as a resi- eight, reduced water/food inta- bounds) could also induce neg- rates, and a rough coat. To pri- nsors and selected dosing regi- ease every 6 to 8 hours throug will include recording observa- th the IACUC 'Pain and Disco- ibund will be immediately rer <b>oviding the approp</b> <b>re the animal exper</b> ossible drug interactions have not been fu- tion directly impact their efficas 50-2C9) pathway, which also metabolical van impact the efficacy of each drug by y intestinal infections. Given these facts a Gudin J et al., 2012; Preissner S et al., 2012 and/or prophylactic efficacy of a new inv for treating life-threatening Clostridioide Dec 1; 44(6): S4-S14. lysis of Common Combinations of Antib <b>S procedure? Cite 1</b>	ies have either remained healthy and normal or ult of Clostridioides difficile infection include ake, and swollen peritoneum. In addition to the gative effects in an animal as well. Signs of test revent treatment related effects, all compounds mens will be based on what has been safely used whout the duration of a study (up to 37 days). A ations and animal disposition, including the date omfort' (document 013) and 'Humane moved from the study and euthanized. <b>Driate anesthetics, analgesics,</b> <b>rienced accompanying pain or</b> hely determined. If another drug is used to relieve pain and/or ne (analgesic) is metabolized by the cytochrome 450-3A4 famycins (rifampio), and fluoroquinolones (ciprofloxacin). y within the host (animal or human). Additionally, several ly interacts with the antibiotics metronidazole and rifampin. As is altering their metabolism and pharmacokinetics in the host. In about documented drug interactions within critical metabolic 2), it is imperative that non-study associated drugs used to relieve vestigational compound can be accurately evaluated. If a new s difficile-associated diarrhea (CDAD) in humans, References: itoics and Pain Relieving Drugs. J Drug Metab Toxicol 2012, 3:5.	

Column E Explanation Registration Number: 74-R-0081 Fiscal Year 2022

### 1. Study One:

Number of animals used in this study: 27

# Species used in this study: Hamster

### Explanation of procedure producing pain and/or distress:

Animals associated with this model may experience pain or distress as a result of infection and/or treatment with a compound. Historically, animals often do not manifest any signs of pain or distress prior to death, and 75% of the animals in those studies have either remained healthy and normal or have been euthanized to prevent prolonged suffering. Signs that indicate a severe disease state as a result of Clostridioides difficile infection include lethargy, acute/chronic diarrhea, hypothermia, >15% decrease in body weight, reduced water/food intake, and swollen peritoneum. In addition to the possible effects related to the infection, treatment with test articles (compounds) could also induce negative effects in an animal as well. Signs of test article related effects include acute paralysis, increased respiratory/heart rates, and a rough coat. To prevent treatment related effects, all compounds evaluated in this model are pre-screened for potential toxicity by the sponsors and selected dosing regimens will be based on what has been safely used for evaluating other test articles in this model.

All animals associated with this protocol will be monitored for severe disease every 6 to 8 hours throughout the duration of a study (up to 37 days). A clinical observation sheet for each day of a study will be maintained that will include recording observations and animal disposition, including the date (day and time) and personnel initials. Using the guidelines associated with the IACUC 'Pain and Discomfort' (document 013) and 'Humane Endpoints' (document 008) protocols, any animal that appears to be moribund will be immediately removed from the study and euthanized. Explanation with reason(s) for why anesthetics, analgesics and tranquilizers could not be used: Most of the test articles that will be evaluated in this model are new investigational compounds and possible drug interactions have not been fully determined. If another drug is used to relieve pain and/or distress, this may interfere with the possible therapeutic/prophylactic efficacy a test article may have in this model. For example, buprenorphine (analgesic) is metabolized by the cytochrome 450-3A4 (CYP450-3A4) pathway. This same metabolic pathway interacts with 3 different antibiotic classes that include macrolides (clarithromycin), rifamycins (rifampin), and fluoroquinolones (ciprofloxacin). Interactions of these drugs in this pathway have been shown to alter the pharmacokinetics of each drug, which can directly impact their efficacy within the host (animal or human). Additionally, several nonsteroidal anti-inflammatory drugs (NSAIDS) are metabolized by the cytochrome 450-2C9 (CYP450-2C9) pathway, which also metabolically interacts with the antibiotics metronidazole and rifampin. As is the case with buprenorphine, the interaction of NSAIDS and other drugs in the CYP450-2C9 pathway can impact the efficacy of each drug by altering their metabolism and pharmacokinetics in the host. In addition to metabolic interference, NSAID use has been shown to clinically aggravate colitis caused by intestinal infections. Given these facts about documented drug interactions within critical metabolic pathways and how these interactions can interfere with drug efficacy or disease outcome in the host (Gudin J et al., 2012; Preissner S et al. 2012) it is imperative that non-study associated drugs used to

relieve pain and/or distress not be administered in animals associated with this model so that the therapeutic and/or prophylactic efficacy of a new investigational compound can be accurately evaluated. If a new compound is efficacious in this animal model, then these results would be critical to its development for treating life-threatening Clostridioides difficile-associated diarrhea (CDAD) in humans.

## **References:**

Gudin J. Opiod Therapies and Cytochrome P450 Interactions. J of Pain and Symp Management, 2012 Dec 1; 44(6): S4-S14.

Preissner S, Kuzman D, Pischon N. Drug Interactions Involving the Cytochrome P450 Enzymes: Analysis of Common Combinations of Antibiotics and Pain Relieving Drugs. J Drug Metab Toxicol 2012, 3:5.

According to the Paperwork Reduction Act of 1995, an agency may not conduct or sponsor, and a person is not required to respond to, a collection of information unless it displays a valid OMB control number. The valid OMB control number for this information collection is 05/20.0002. The time period to a subject the displays a valid of the		OMB APPROVED 0579-0036	
0579-0036. The time required to complete this information collection is estimated to average .5 hours per response, including the time for reviewing instructions, searching existing data sources, gathering and maintaining the data needed, and completing and reviewing the collection of information.		Interagency Report Control No. 0180-DOA-AN	
		Fiscal year: 2022	
UNITED STATES DEPARTMENT OF AGRICULTURE			
ANIMAL AND PLANT HEALTH INSPECTION SERVICE			
Appual Papart of Paparah Essility			
Annual Report of Research Facility Column E Explanation			
(TYPE OR PRINT)			
This information is required by law (7 U.S.C. 2143 and 9 C.F.R. §2.36). Failure to report according to the regulations can result in an order to cease and desist.			
1. REGISTRATION NUMBER	2. Research Faci	lity Headquarters address	
74-R-0081	3500 Camp Bowie Blvd Fort Worth, Texas 76107		
3. Number of animals used in the study.	4. Species (com	non name) of animals used in	
6		Rabbit	
5. Explain the procedure producing pain and d			
Animals associated with this model may experience pain or distress as a result of infection and/or treatment with a compound. Historically, animals often do not manifest any signs of pain or distress prior to death, and 99% of the animals in those studies have either remained healthy and normal or have been euthanized to prevent prolonged suffering. Signs that indicate a severe disease state as a result of a bacterial pneumonia infection include lethargy, hypothermia, hunched posture, emaciated state, and labored breathing. In addition to the possible effects related to the infection, treatment with test articles (compounds) could also induce negative effects in an animal as well. Signs of test article related effects include acute paralysis, increased respiratory/heart rates, and a rough coat. To prevent treatment related effects, all compounds evaluated in this model are pre-screened for potential toxicity by the sponsors and selected dosing regimens will be based on what has been safely used for evaluating other test articles in this model. All animals associated with this protocol will be monitored for severe disease every 6 to 8 hours throughout the duration of a study (up to 21 days). A clinical observation sheet for each day of a study will be maintained that will include recording observations and animal disposition, including the date (day and time) and personnel initials. Using the guidelines associated with the IACUC 'Pain and Discomfort' (document 013) and 'Humane Endpoints' (document 008) protocols, any animal that appears to be moribund will be immediately removed from the study and euthanized.			
6. Provide the scientific justification for not providing the appropriate anesthetics, analgesics,			
or tranquilizing drugs during procedures where the animal experienced accompanying pain or distress greater than momentary or slight.			
Most of the test articles that will be evaluated in this model are new investigational compounds and possible drug interactions have not been fully determined. If another drug is used to relieve pain and/or distress, this may interfere with the possible threapeutic/prophylactic efficacy a test article may have in this model. For example, buprenorphine (analgesic) is metabolized by the cytochrome 450-3A4 (CYP450-3A4) pathway. This same metabolic pathway interacts with 3 different antibiotic classes that include macrolides (clarithromycin), rifernycins (rifampin), and fluoroquinolones (ciprofloxacin). Interactions of these drugs in this pathway have been shown to alter the pharmacokinetics of each drug, which can directly impact their efficacy within the host (animal or human). Additionally, several nonsteroidal anti-inflammatory drugs (INSAIDS) are metabolized by the cytochrome 450-203 (DYP450-203) pathway, which also metabolically interacts with the antibiotics metronidazole and rifampin. As is the case with buprenorphine, the interaction of NSAIDS and other drugs in the CYP450-209 pathway can impact the efficacy of each drug by altering their metabolism and pharmacokinetics in the host. In addition to metabolic interference, NSAID use has been shown to clinically aggravate colitis caused by intestinal infections. Given these facts about documented drug interactions within critical metabolic and/or prophylactic efficacy of a new investigational compound can be accurately evaluated. If a new compound is efficacious in this animal model, then these results would be critical to its development for treating life-threatening respiratory infections in humans. References: Gudin J, Opiod Therapies and Cytochrome P450 Interactions, J of Pain and Symp Management, 2012 Dec 1; 44(6): S4-S14, Preissner S, Kuzman D, Pischon N, Drug Interactions Involving the Cytochrome P450 Enzymes: Analysis of Common Combinations of Antibiotics and Pain Relieving Drugs. J Drug Metab Toxicol 2012, 3:5.			
7. 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		CFR	

Column E Explanation Registration Number: 74-R-0081 Fiscal Year 2022

### 1. Study One:

### Number of animals used in this study: 6

#### Species used in this study: Rabbit

## Explanation of procedure producing pain and/or distress:

Animals associated with this model may experience pain or distress as a result of infection and/or treatment with a compound. Historically, animals often do not manifest any signs of pain or distress prior to death, and ~99% of the animals in those studies have either remained healthy and normal or have been euthanized to prevent prolonged suffering. Signs that indicate a severe disease state as a result of bacterial pneumonia infection include lethargy, hypothermia, hunched posture, emaciated state, and labored breathing. In addition to the possible effects related to the infection, treatment with test articles (compounds) could also induce negative effects in an animal as well. Signs of test article related effects include acute paralysis, increased respiratory/heart rates, and a rough coat. To prevent treatment related effects, all compounds evaluated in this model are pre-screened for potential toxicity by the sponsors and selected dosing regimens will be based on what has been safely used for evaluating other test articles in this model.

All animals associated with this protocol will be monitored for severe disease every 6 to 8 hours throughout the duration of a study (up to 21 days). A clinical observation sheet for each day of a study will be maintained that will include recording observations and animal disposition, including the date (day and time) and personnel initials. Using the guidelines associated with the IACUC 'Pain and Discomfort' (document 013) and 'Humane Endpoints' (document 008) protocols, any animal that appears to be moribund will be immediately removed from the study and euthanized.

Explanation with reason(s) for why anesthetics, analgesics and tranquilizers could not be used: Most of the test articles that will be evaluated in this model are new investigational compounds and possible drug interactions have not been fully determined. If another drug is used to relieve pain and/or distress, this may interfere with the possible therapeutic/prophylactic efficacy a test article may have in this model. For example, buprenorphine (analgesic) is metabolized by the cytochrome 450-3A4 (CYP450-3A4) pathway. This same metabolic pathway interacts with 3 different antibiotic classes that include macrolides (clarithromycin), rifamycins (rifampin), and fluoroquinolones (ciprofloxacin). Interactions of these drugs in this pathway have been shown to alter the pharmacokinetics of each drug, which can directly impact their efficacy within the host (animal or human). Additionally, several nonsteroidal anti-inflammatory drugs (NSAIDS) are metabolized by the cytochrome 450-2C9 (CYP450-2C9) pathway, which also metabolically interacts with the antibiotics metronidazole and rifampin. As is the case with buprenorphine, the interaction of NSAIDS and other drugs in the CYP450-2C9 pathway can impact the efficacy of each drug by altering their metabolism and pharmacokinetics in the host. In addition to metabolic interference, NSAID use has been shown to clinically aggravate colitis caused by intestinal infections. Given these facts about documented drug interactions within critical metabolic pathways and how these interactions can interfere with drug efficacy or disease outcome in the host (Gudin J et al., 2012; Preissner S et al. 2012), it is imperative that non-study associated drugs used to relieve pain and/or distress not be administered in animals associated with this model so that the

therapeutic and/or prophylactic efficacy of a new investigational compound can be accurately evaluated. If a new compound is efficacious in this animal model, then these results would be critical to its development for treating life-threatening respiratory infections in humans.

**References:** 

Gudin J. Opiod Therapies and Cytochrome P450 Interactions. J of Pain and Symp Management, 2012 Dec 1; 44(6): S4-S14.

Preissner S, Kuzman D, Pischon N. Drug Interactions Involving the Cytochrome P450 Enzymes: Analysis of Common Combinations of Antibiotics and Pain Relieving Drugs. J Drug Metab Toxicol 2012, 3:5.