

Guidelines for Use of Experimental Animals During Training Simulations Utilizing Flight Research Animal Housing Units, Animal Transportation Modules, Gloveboxes or Work Stations, Chamber Simulations (Closed Environments), and Space Flight

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This guideline summarizes the JSC CPHS current requirements and recommendations regarding subject experimental animal standards and procedures as viewed in the context of past advisory group meeting on this and related topics.

1. All animal holding facilities and/or breeding colonies will generally adhere to the guidelines and recommendations of the current National Research Council's Guide for Care and Use of Laboratory Animals, National Academy Press (currently 1996), and the Association for Assessment and Accreditation of Laboratory Animal Care, International (AAALAC, Int.).
2. Only NASA Flight Quality (NFQ) rodents and monkeys shall be utilized for crewmember training and flight activities. The NFQ criteria for rats and mice are given in Tables 1 and 2 respectively. Table 3, describes the NFQ criteria for squirrel monkeys and Table 4 for macaques. Table 5 describes the NFQ criteria for Xenopus. Fish shall be obtained from colony-bred sources. The use of feral animals for flight or training is discouraged.
3. Other animal species proposed for flight experiments shall be considered by the JSC CPHS on an individual basis. Other animal species will be considered by the JSC CPHS on an individual basis. Animals which are not colony or captive reared, may carry a greater variety of pathogens some of which may have undetermined zoonotic potential. The use of feral or non-colony born animals is discouraged.
4. The following general guidelines shall be followed where applicable:
 - A. STANDARD MICROBIOLOGICAL PRACTICES
 1. Work Surfaces shall be decontaminated with a suitable disinfectant before and after use.

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2. All waste liquids, solids, tissues, syringes and needles shall be placed in durable, leak proof, puncture-resistant, sealed containers for eventual autoclaving, incineration, or other appropriate decontamination/disposal procedure post-training, post-simulation or postflight. Such materials will not be transported between the animal investigation area and crew living quarters.
3. Hypodermic needles and syringes shall be used only for the parenteral injection or aspiration of fluids from laboratory animals and diaphragm bottles. Only needle-locking syringes or disposable needle syringe units (i.e., the needle is integral to the syringe) are to be used for the injection or aspiration of fluids. Needles should not be bent, sheared or removed from the syringe following use except if an aspirate is to be transported within a syringe. The needle shall be removed and appropriately discarded and the syringe tip shall be appropriately capped. Needles should not be replaced in the plastic sheath or guard prior to disposal. Needle and syringe should be promptly placed in puncture-proof container for eventual decontamination, preferably by autoclaving, before final discard.
4. Personnel shall use appropriate antiseptic wet wipes or other available means for cleaning hands after handling animals, when departing the laboratory, and especially before eating.
5. Street clothing, a laboratory coat (or equivalent) and appropriate protective gloves shall be worn when animals are handled. Facilities may have more stringent requirements for attire when working with or around laboratory animals; these standards shall apply within those facilities. Shorts, sandals or opened toed shoes may not be worn under a laboratory coat in the Animal Care Facility.

B. ANIMAL CERTIFICATION

1. Animals will be certified NFQ by the supplier for the proscribed organisms listed in Tables 1-6. Rodents will be housed appropriately in filtered cages. A minimum of 5% of animal populations destined for potential crew contact or actual space flight will be sampled for microbial culture screening by oral swab and fecal sample for those organisms on the SPF list upon receipt and again no more than 11 days prior to crew contact. Presumptive results must be available in 24 hours and definitive results in 72 hours. The crew will not be exposed to animals if the

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sampled animals cultures are positive for a proscribe organism. Rodent viral serology will be completed two weeks prior to crew exposure according to established protocols.

2. Monkeys shall be screened for proscribed organisms at six-month intervals. The flight animals selected will have viral serology screening completed one month before use; will be cultured for proscribed bacteria and undergo intra palpebral tuberculin testing using mammalian old tuberculin (MOT) testing 96 hours prior to crew contact. All microbiological and tuberculin skin test results will be forwarded to the JSC IRB as part of the Operational Readiness Review (ORR).
3. NFQ certified squirrel monkeys will at all times be housed in isolation apart from other non-certified non-human primates. The isolation quarters will be provided with a nonrecirculating type ventilation system to preclude contamination from other animals. Room entry will require shoe covers in addition to the standard outerwear (scrub suits, lab coat, mask and gloves as defined by CDC/NIOSH).

C. IN-FLIGHT GUIDELINES FOR ANIMAL HOUSING UNITS, ANIMAL TRANSPORTATION MODULES AND GLOVEBOXES OR WORK STATIONS

1. With the improved integrity of animal enclosures and associated flight procedures, THE ROUTINE USE OF LABORATORY ATTIRE IS NOT REQUIRED.

If anomalous situations should develop which produce free contaminants, all crewmembers will use suitable protective measures (viz., NIOSH-approved respirator) until the particular experiment or procedure is terminated and the contaminant is satisfactorily removed from the spacecraft. This precaution is necessary in the closed microgravity environment, since contamination does not remain localized in the continuous atmosphere of spacecraft.

Particular care should be exercised during the following procedures:

- a. Rats/Mice: Waste tray and food canister changeout; cage removal; condensate bottle changeout; Glovebox/Work Station operations involving animals.

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- b. Squirrel monkeys: Waste tray changeout; urine canister changeout; food canister changeout; blood sample collection.
2. Animal Housing Units and Glovebox/Work Stations will be designed to filter particulate matter and keep it from exhausting into the spacecraft.
3. Biological samples from animals shall not contaminate the spacecraft or crew at any time during collection, transport and storage procedure.
4. Animals transported between Animal Housing Units and Glovebox/Work Stations must be enclosed in a carrier.
5. Equipment and procedures for the housing, transport, and experimental protocol must preclude any possibility of animal escape into the spacecraft.

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TABLE 1
SPECIFIC PATHOGEN FREE (SPF) CRITERIA FOR RATS

MICROORGANISM	CULTURE SITE/MATERIAL
BACTERIA:	
Streptobacillis moniliformis	Oropharyngeal
Spirillum minus	Blood (Serology)
Streptococcus pneumoniae	Oropharyngeal
Streptococcus pyogenes	Oropharyngeal
Bacillus piliformis	Liver (invoke with Cortisone)
Cornybacterium kutscheri	Fecal, Oropharyngeal
Salmonella sp.	Fecal
Pasteurella pneumotropica	Oropharyngeal
Leptospira sp.	Urine
Campylobacter sp.	Fecal
VIRUSES:	
Lymphocytic Choriomeningitis virus (LCM)	Blood (Serology)
Rat paroviruses (TH1, KRV)	Blood (Serology)
Rat coronavirus (RCV)	Blood (Serology)
Sialodacryadenitis virus (SDAV)	Blood (Serology)
Sendal virus (SEN)	Blood (Serology)
FUNGI:	
All Pathogenic Dermatophytes	Skin
ECTOPARASITES:	Skin, Hair
ENDOPARASITES:	Fecal, Caecal Contents 1

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TABLE 2
SPECIFIC PATHOGEN FREE (SPF) CRITERIA FOR MICE

MICROORGANISM	CULTURE SITE/MATERIAL
BACTERIA/MYCOPLASMAS:	
Streptobacillis moniliformis	Oropharyngeal
Spirillum minus	Blood (Serology)
Streptococcus pneumoniae	Oropharyngeal
Streptococcus pyogenes	Oropharyngeal
Bacillus piliformis	Liver (invoke with Cortisone)
Cornybacterium kutscheri	Fecal, Oropharyngeal
Salmonella sp.	Fecal
Pasteurella pneumotropica	Oropharyngeal
Leptospira sp.	Urine
Campylobacter sp.	Fecal
Mycoplasma pulmonis (MYCO)	Blood (Serology)
VIRUSES:	
Pneumonia Virus of Mice (PVM)	Blood (Serology)
Mouse parovirus (MVN)	Blood (Serology)
Mouse coronavirus (MHV)	Blood (Serology)
Sendal virus (SEN)	Blood (Serology)
FUNGI:	
All Pathogenic Dermatophytes	Skin
ECTOPARASITES:	Skin, Hair
ENDOPARASITES:	Fecal

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TABLE 3
SPECIFIC PATHOGEN FREE (SPF) CRITERIA FOR SQUIRREL MONKEYS

MICROORGANISM	CULTURE SITE/MATERIAL
BACTERIA:	
Shigella sp.	Fecal
Salmonella sp.	Fecal
Streptococcus pneumoniae	Oral, Fecal
Mycobacterium tuberculosis	Skin test/chest X-ray
Pasteurella multocida	Nasal, Fecal
Campylobacter sp.	Fecal
Streptococcus pyogenes	Oral, Nasal
VIRUSES:	
Lymphocytic choriomeningitis virus	Blood (Serology)
Herpes tamarinus	Blood (Serology)
Herpesvirus saimiri	Blood (Serology)
ENDOPARASITES:	
Trichomonas	Oral
Acanthocephalans	Feces
Strongyloides	Feces
Entamoeba histolytica	Feces
Hemoprotozoa	Blood
SPIROCHETES:	
Leptospira sp.	Urine culture
FUNGI:	
All Pathogenic Dermatophytes	Skin

NOTE: Table 3 above is from the Rev A version. For Rev C, the following changes were incorporated: Microsporum sp., and Trichophyton sp. were ADDED and Streptococcus pneumoniae, Pasteurella multocida, Leptospira sp., Streptococcus pyogenes and Acanthocephalans were REMOVED.

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TABLE 4
SPECIFIC PATHOGEN FREE (SPF) CRITERIA FOR MACACA MULLATA

MICROORGANISM	CULTURE SITE/MATERIAL
BACTERIA:	
<i>Mycobacterium tuberculosis</i>	Skin test/chest X-ray (Cohost group must be negative on 3 successive tuberculin skin tests conducted at 2-week intervals)
<i>Shigella</i> sp.	Fecal
<i>Salmonella</i> sp.	Fecal
<i>Pasteurella multocida</i>	
<i>Yersinia pseudotuberculosis</i>	Fecal
<i>Yersinia enterocolitica</i>	Fecal
<i>Streptococcus pneumoniae</i>	
<i>Campylobacter jejuni</i>	Fecal
VIRUSES:	
Herpesvirus simiae (B virus)	Blood (Serology)
Yaba	Blood (Serology)
Tanapox virus group	Blood (Serology)
Monkeypox	Blood (Serology)
Rubeola (Measles)	Blood (Serology)
Lymphocytic choriomeningitis	Blood (Serology)
Rabies	Blood (Serology)
SRV-1, SRV-2	Blood (Serology)
HIV, SIV	Blood (Serology)
STLV-1	Blood (Serology)
PARASITES:	
<i>Rodenolepis nana</i>	Microscopic exam; fecal
<i>Entamoeba histolytica</i>	Microscopic exam; fecal
<i>Giardia</i> sp.	Microscopic exam; fecal
<i>Balantidium coli</i>	Microscopic exam; fecal
<i>Trichomonas hominis</i>	Microscopic exam; fecal
<i>Ascaris lumbricoides</i>	Microscopic exam; fecal
<i>Strongyloides</i> sp.	Microscopic exam; fecal

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Enterobius hominis	Microscopic exam; fecal
Trichuris sp.	Microscopic exam; fecal
SPIROCHETES:	
Leptospira sp.	Urine culture
FUNGI:	
All Pathogenic Dermatophytes	Skin

NOTE: Table 4 above is from the Rev A version. For Rev C, the following changes were incorporated: Ebolavirus, Hymenolepis nana, Microsporium sp., and Trichophyton sp. were ADDED and Streptococcus pneumoniae, Pasteurella multocida, HIV, STLV-1 and Rodenolepis nana were REMOVED.

TABLE 5
SPECIFIC PATHOGEN FREE (SPF) CRITERIA FOR *Xenopus laevis*

MICROORGANISM	CULTURE SITE/MATERIAL
<i>Aeromonas hydrophila</i>	Culture of lesions or water sample
<i>Alaria</i> sp.	Fecal floatation in 10% sucrose solution; Histopathology of representative animals
<i>Salmonella</i> sp.	Fecal culture; culture water
<i>Mycobacterium</i> group IV (<i>M. xenopi</i> , <i>M. marinum</i>)	Culture water; selective media and conditions
<i>Chlamydia psittaci</i>	Necropsy and liver histopathology of representative animals

NOTE: There were no SPF requirements for *Xenopus* in the Rev A version. For Rev C, the crew training and flight requirements were completely different. Table 5 above is the flight requirements with the crew training requirements added.

TABLE 6
SPECIFIC PATHOGEN FREE (SPF) CRITERIA
(NASA Requested for Unique Basis)

MICROORGANISM	CULTURE SITE/MATERIAL
<i>Staphylococcus aureus</i>	Nasopharyngeal culture
<i>Klebsiella oxytoca</i>	Culture of cecal contents/feces