

Division of Research Comparative Medicine

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SOP # 204	Rodent Quarantine Non-approved Vendors
SCOPE:	This SOP is applicable to all Comparative Medicine (CM) personnel responsible for the receipt, housing and processing of incoming shipments of rodents into quarantine.
SOP OWNER:	Attending Veterinarian (AV)
PURPOSE:	To prevent the introduction of rodent pathogens into established rodent colonies.
LOCATION:	Vivarium 35B, specific rooms in other vivaria as approved by AV
LOCATION:	rodent colonies.

I. RESPONSIBILITIES

- A. Attending Veterinarian (AV) and Director
 - 1. The veterinarian oversees all aspects of animal health including quarantine of animals from non-approved vendors.
 - 2. Reviews health information from approved and non-approved vendors/institutions and discusses options for import of rodents from non-approved vendor(s), anticipated lengths of quarantine, necessary testing of animals and other relevant topics with appropriate research personnel.

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- 3. Interprets test results and determines disposition of quarantine animals.
- 4. Directs CM personnel regarding personal protective equipment (PPE), quarantine husbandry procedures, collection of specimen for testing, possible treatments to be administered and time point of animal release into regular housing.
- 5. Assures that CM personnel is appropriately trained and adhere to procedures as outlined in this SOP.
- B. Coordinator, Administrative Services (CAS)
 - 1. Receives animal orders from research personnel.
 - 2. Receives Incoming Shipment Request Form from the Principal Investigator (PI) and contacts corresponding institution. Coordinates communication between non-approved vendor institution, FAU research personnel and FAU veterinarian.
 - 3. Coordinates shipment of animals and provides Facility Manager with appropriate Rodent Quarantine Record (i.e. CM013 or CM014) to include expected arrival date for each shipment.
- C. Facilities Manager (FM)
 - 1. Coordinates location of quarantine with AV depending on availability.
 - 2. Assigns responsible animal care technician for each animal shipment and notifies AV if health reports of approved vendor(s) indicate that animals are positive for murine pathogens that are not already known and/or accepted.
 - 3. Assures limited access to quarantine area.
 - 4. Oversees receipt of animals into quarantine housing, initial health evaluation, care/husbandry procedures within quarantine location, possible treatments, collection of test samples, shipping to commercial laboratory and in-house testing as directed by AV and outlined in this SOP.
 - 5. Inform AV immediately about any equivocal or positive results and abnormal findings and/or observations.
 - 6. Assures that responsible Animal Care Technicians are appropriately trained and implement procedures as delineated in this SOP.
- D. Animal Care Technician (ACT)
 - Adhere to procedures as outlined in this SOP or as directed by Facility Manager and/or AV.
 - 2. Document all activities as outlined below in section Recordkeeping.

II. GENERAL

- A. **Rodents**, as defined for the purpose of this SOP, are considered various strains/lines of both rats of the species *Rattus norvegicus* and mice of the species *Mus musculus*.
- B. Rodents from approved commercial vendors of animals from SPF/VAF colonies are housed without a period of quarantine. The AV establishes the list of approved vendors and publishes it on the CM website available to all stakeholders of the FAU Animal Care and Use Program. The Coordinator, Administrative Services is

- responsible for ensuring that requested animals will be delivered from SPF/VAF colonies. All animals should be acclimated as per IACUC Policy.
- C. Health status of approved vendors are posted at their respective websites and reviewed regularly by the AV but no less than every 6 months. If vendors send health reports with each animal delivery, these will be filed by vendor and calendar year after review by the receiving person. If health reports indicate animals being positive for an excluded murine pathogen, the AV will be notified immediately before moving the animals into the housing room.
- D. Rodents from non-approved sources including commercial vendors and other academic institutions, regardless of written health assurances are quarantined at the discretion of the AV and evaluated as described below.
- E. Access to quarantine rooms/facilities is limited to essential personnel. Research personnel are not allowed in quarantine areas unless explicitly requested, permitted by the AV and accompanied by CM staff. No experimental procedures are routinely to be performed on quarantine animals. Breeding might be permitted and overseen by CM personnel as depending on circumstances and permitted by the AV.
- F. Just before entering the quarantine room, don appropriate personal protective clothing (PPE), which is posted at the outside of the quarantine room door. Animal Care personnel have to wear dedicated scrubs in addition to indicated PPE when performing husbandry procedures other than daily health checks only, which cannot be worn into any other rodent housing or procedural area. These scrubs have to be separately collected and laundered using bleach. Any person entering a quarantine room has to don disposable gown, shoe-covers, bouffant cap, surgical mask and gloves or Tyvek® coveralls as directed by the AV. When performing procedures that include opening cages Tyvek sleeves have to be donned. Tyvek sleeves have to be sprayed with a disinfectant after use and can be reused while left in the room. Those need to be discarded at least once a week or more often if torn. All other disposable protective clothing is removed after the completion of all duties and disposed of in the trash container upon exiting the room. Shoes are sprayed with a disinfectant approved by the AV before leaving the area and hands are sanitized immediately upon exiting.
- G. Every piece of equipment or supply item, either dirty or still clean, removed from the quarantine room has to be bagged, sprayed with a disinfectant and then autoclaved as per SOP. If it is an item that cannot be autoclaved a different sterilization technique has to be employed as directed by the AV.
- H. Quarantined animals are housed in either a ventilated cage/rack system or autoclavable microisolator cages, in a separate room from non-quarantined animals. Only if no separate room is available and with special consideration and approval by the AV, semi-rigid isolators can be used for housing quarantined animals, within a regular housing room.
- Quarantine areas are always serviced last in the day, by a designated technician. If several shipments are quarantined at the same time service earliest shipment first, the latest shipment last, etc. unless otherwise specified by the AV. If under special

circumstances the designated technician or anybody else having visited the quarantine area, must enter a regular rodent housing/procedure area, the individual has to take a whole body shower including washing hair and don clean scrubs/PPE.

- J. The following murine pathogens (i.e. rats and mice) are excluded for all animal housing facilities of FAU's animal care and use program:
 - 1. Parasites:
 - i. Fur mites (Myobia, Myocoptes, Radfordia)
 - ii. Pinworms (Aspicularis, Syphacia)
 - iii. Spironucleus muris
 - iv. Entamoeba
 - v. Giardia
 - 2. Viruses:
 - i. Parvovirus (MPV, MVM or RPV, KRV, RMV)
 - ii. Coronavirus (MHV or SDAV)
 - iii. Poliovirus (TMEV –GDVII or RTV)
 - iv. Parainfluenzavirus type I (Sendai)
 - v. Paramyxovirus (PVM)
 - vi. Reovirus Types 1&3 (Reo)
 - vii. Artevirus (LCMV)
 - viii. Adenovirus (MAV)
 - ix. Hantavirus
 - x. Rotavirus (EDIM or IDIR)
 - xi. Lactate Dehydrogenase Virus (LDH)
 - xii. Papovavirus (Poly and K virus) mice
 - xiii. Mouse Cytomegalovirus (MCMV) mice
 - xiv. Poxvirus (Ectromelia) mice
 - **xv.** Mouse T Lymphocytic Virus (MTLV) mice
 - 3. Bacteria:
 - i. CAR bacillus
 - ii. Mycoplasma pulmonis
 - iii. Citrobacter rodentium
 - iv. Corynebacterium kutscheri and bovis
 - v. Streptobacillus moniliformis
 - vi. Streptococcus pneumoniae
 - 4. Fungi:
 - i. Enzephalitozoon cuniculi
 - ii. Pneumocystis spp. rat
- K. In addition, any *Helicobacter* spp. and Mouse Norovirus (Calicivirus, MNV) are excluded for the Boca Raton housing facilities.

III. PROCEDURE

A. Preparation

- Animal orders submitted by research personnel are checked against the approved vendor list by the CAS. Non-approved vendors include particular commercial vendors and other research institutions such as universities and private non-for-profit research institutions. Non-approved vendors can be domestic or international.
- 2. Health status records of animals from non-approved commercial vendors will be reviewed by the AV, animals ordered by the CAS and received into guarantine.
- 3. The CAS secures the Incoming Shipment Request Form for animals from other research institutions if not already provided by the research personnel. The outlined responsible person at the other institution will be contacted to request the following documents/information:
 - i. Health records for the housing facility for at least the previous 12 months including test methods used such as PCR, serology, necropsy/histopathology, in-house CM. commercial laboratory testing
 - **ii.** Information about specific disease outbreaks within any of the facilities belonging to the animal care unit of the institution
 - **iii.** Brief description of housing conditions, husbandry and care procedures, and work practices of the animal care unit such as work flow within particular facility and between facilities, sharing of ACTs between facilities of equal or differing health status, and biosecurity status of the particular housing facility (e.g. barrier, conventional facility) including animal movement between housing facility and research labs.
- 4. The AV reviews all provided documents, requests additional information if necessary, decides whether animals can be imported and discusses decision and further options with the relevant PI.
- 5. The AV communicates specific testing/treatment options based on the health status, biosecurity level of the shipping facility, domestic or international shipment and any other relevant information to the CAS and FM.
- 6. The CAS arranges shipment with shipping institution and FAU PI. Shipment has to be paid either by the FAU PI or his/her collaborator.
- 7. The CAS provides the Facility Manager with the appropriate Rodent Quarantine Record (i.e. CM013 or CM014) and communicates the expected arrival date.

B. Receiving by FM or Designated ACT

- 1. Receive incoming rodent quarantine shipment at the appropriate receiving area, according to SOP Rodent Receiving (FAUSOP 406), especially check shipping container for any signs of damage, spray with approved disinfectant and verify information on shipping container matches shipping paperwork. Communicate discrepancies to relevant CM personnel (i.e. FM, CAS and/or AV).
- 2. Transport shipping container to quarantine area, spray wheels of cart with disinfectant and push cart into quarantine room, which is already stocked with appropriate caging and supplies.

Follow PPE rules as described above.

C. Husbandry and Care

- 1. Animals are checked daily as described in *SOP Rodent Husbandry and Care* (FAUSOP 402).
- Place appropriate numbers of clean caging supplies for incoming shipment(s) or cage changing on sanitized transport cart and push into quarantine room.
 Spray wheels of cart with disinfectant if coming from outside of facility (i.e. in building 35B).
- 3. Cage changing for scheduled changes or spot changes are performed as in regular housing. Please refer to *SOP Rodent Husbandry and Care* (FAUSOP 402).
- 4. Work surfaces are decontaminated with an approved disinfectant before and after **each** use.
- 5. Empty water bottles in the sink inside the room if possible. If not, leave the partially filled bottles inside the cage to be autoclaved.
- 6. Stack dirty cages in an autoclave bag and tape it shut using autoclave tape. Place a biological indicator in every second or third bag depending on their size. Mark the bag(s) containing a bio indicator with a sharpie and make sure each autoclave load contains one of those.
- 7. Place every item to be removed from quarantine room in an autoclavable biohazard bag if possible and spray with disinfectant. Observe contact time of particular disinfectant.
- 8. Move removed items to nearest autoclave (i.e. 35B to 35A autoclave and in other facilities to autoclave of respective vivarium) and sterilize as soon as possible.
- 9. If specific equipment or supplies cannot be autoclaved follow directions by AV for appropriate sterilization/sanitation procedure.
- 10. Trash from quarantine area has to be collected in heavy duty trash bags, closed and sprayed before removal from room.

D. Testing and Treatment of Quarantine Animals

- 1. Standard quarantine practices include
 - i. In-house testing immediately upon arrival for 1) ectoparasites via fur plucks, 2) pinworms via anal tape test and 3) endoparasites via fecal flotation
 - **ii.** Fecal matter and body swabs collection for PCR testing 4-7 days after arrival. At a minimum a PRIA Prevalent panel has to be requested from Charles River Laboratory (CRL).
- 2. Non-standard quarantine practices
 - i. Will be employed for
 - a. All international imports and
 - b. National shipments at the discretion of the AV.

ii. Involve

- a. 1) in-house testing as per D.1.i., 2) initial PCR testing 4-7 days after arrival and 3) serological testing
- b. PCR testing with PRIA Prevalent
- Blood samples collected for serology directly from imported animals (immunologically mature animals, i.e. ≥ 10 wks old) after 2-4 weeks of arrival to be tested with Assessment Plus panel
- 3. The PCR and serology panels listed in table 1 are for regular testing and can be changed by the AV on a case by case basis. In addition, sentinel animals might be used if necessary.
- 4. For small shipments (≤10 animals), each animal will be evaluated.
- 5. For medium size shipments (>10 ≤ 40 animals), representative animals from each cage are evaluated. The number of animals to be evaluated is determined based on the animal's health history, the health history of the originating facility, and the method housed, but no less than 25% of the arriving animals so as to achieve a 95% confidence that the individual results reflect the population's health status (assuming an infectivity rate of 15%).
- 6. The number of animals to be evaluated for large shipments of rodents (>40 animals) is determined on a case by case basis, and is based on the animal's health history, the health history of the originating facility, and the method housed, but no less than 10% of arriving animals so as to achieve a 95% confidence that the individual results reflects the population's health status (assuming an infectivity rate of 30%).
- 7. If sentinel animals are used one sentinel cage (2 mice per cage) per ≤10 cages will be placed. Contact animals might be deployed at the discretion of the AV in agreement with the PI. One animal per sentinel cage will be euthanized for serology testing no earlier than 10 weeks after placement. The second animal is kept until serology results are received and its disposition decided at this time.
- 8. Additional health evaluations are conducted at the discretion of the veterinarian, after consultation with the research staff.
- 9. If test results are positive the veterinarian will discuss with the PI and develop a plan that will ensure delivery of the rodent line/strain free of evidence of exposure to agents on the exclusion list.
- 10. Animals positive for Helicobacter and/or MNV might be approved for import into Boca Raton vivaria at the discretion of the AV. Also, animals might test unexpectedly positive for these agents. Those animals will be bred and pups rederived via cross fostering to clean dams to eliminate Helicobacter/MNV if possible. Additional testing will be necessary and the AV will discuss specifics with the particular researcher(s).
- 11. Necessity for any treatment is assessed by the AV. Treatment might include but is not limited to

- i. Endoparasite treatment (e.g., pinworms) by providing Harlan Teklad TD 01432 (a pelleted rodent diet containing fenbendazole) ad libitum for days 1-7 of quarantine and for alternating subsequent weeks 3 and 5 (7 days on, 7 days off) for a total of 21 days of medication
- ii. Ectoparasite treatment (e.g., fur mites), and nematode endoparasite treatment, by the application of Ivermectin (10 mg/ml) at a dose of 1μ I/5grams of body weight topically between the animals scapulae on day 1 and day 10 of quarantine. The micropipette tip application of Ivermectin should be against the grain of hair growth, directly on the skin, while the animal is restrained.
- 12. All treatments, evaluations, tests initiated/performed and test results are recorded on the relevant Rodent Quarantine Record.
- 13. Table 1: Sample Collection Schedule for Mice and Rats

Time	Protocol	Specimen Type			
Standard Quarantine for Unapproved Vendors					
Day 0-1	In-House Parasitology	Feces, Fur & Perianal			
	(endo/ectoparasites)	Tape			
Day 4-14	PRIA Prevalent (PCR)	Body Swab &Fecal			
		Pools			
Comprehensive Quarantine for Exotic Sources					
Day 0-1	In-House Parasitology	Feces, Fur & Perianal			
	(endo/ectoparasites)	Tape			
Day 4-14	PRIA Prevalent (PCR)	Body Swab &Fecal			
		Pools			
Day 14-28	Assessment Plus	Dried Blood Spot 2:1			
	Serology Panel				

E. Release of animals from Quarantine

- 1. After all test results have been gathered the AV reviews and makes a final decision, which is discussed with the PI. The decision is to whether
 - i. Release the animals to regular housing
 - ii. Require additional test(s) and/or treat the animals
 - iii. Euthanize animals
 - iv. Or any other reasonable disposition.
- Only after the Rodent Quarantine Record has been signed and release documented by the AV, the FM informs the PI via email and arranges transfer of the animals to the final housing area.
- 3. After transfer of the animals, the procedures for cleaning and/or sterilization of equipment/supplies left in the quarantine room are decided by the AV.

 The quarantine room has to be cleaned and disinfected.

IV. RECORDKEEPING

- A. Rodent Quarantine Record
 - 1. A shipment specific Rodent Quarantine Record has to be established by the CAS before arrival of the animals.
 - 2. The AV completes the rodent health history and indicates the evaluations and/or tests performed during quarantine.
 - 3. The prepared Rodent Quarantine Record is placed in the particular room log book by the FM.
 - 4. The assigned technician(s) completes the form as health evaluations are performed or tests required making sure dates and initials of the responsible person are entered.
 - 5. As results become available, the number of results per total number of tests requested are entered while negative results are entered as 0 (zero) and positive results as the number of positive results.
 - 6. After all testing has been completed, the form and the particular test results are presented to the AV for final review. Based on the test results the AV enters the final disposition of the animals.
 - 7. The FM completes the record including date of communication with the PI and transfer of animals to regular housing.
 - 8. Any relevant information not captured in the Rodent Quarantine Record, e.g. health conditions unrelated to quarantine such as fight wounds, are written into the progress notes located in the room log book.
- B. All paperwork related to a particular shipment from a non-approved vendor
 - 1. Are separately filed/archived per import/shipment
 - 2. Kept for at least three years following completion of quarantine
 - 3. Includes but is not limited to:
 - i. Any paperwork including health reports provided by the originating institution.
 - ii. Relevant email conversations.
 - **iii.** Shipping paperwork including health certificates
 - iv. Testing results and evaluations performed.
 - v. Rodent Quarantine Record
 - vi. Progress Note sheet(s) from the log book of the quarantine room.

Review Date	Revision Date	Revision Number	Description of Revision
01/06/16	01/06/16	204.1	Review of sent vendor health reports, addition of exclusion pathogen list, changed health status of Jupiter vivaria
7/26/16	7/26/16	204.2	Update of pathogen exclusion panel, update of testing of quarantine animals with addition of table for overview