

Tel-Aviv University –Safety Unit

Standard Operating Procedure for Working with *Human Metapneumovirus (hMPV)* in Animals.

1. Health hazards	<p>Human metapneumovirus (hMPV) is an enveloped, negative single-stranded RNA virus of the family Paramyxoviridae .</p> <p>hMPV causes upper and lower respiratory tract infection, in humans of all ages.</p> <p>The infection can cause more severe disease in infants, children, elderly people, and immunocompromised individuals.</p> <p>hMPV infections range from mild respiratory illness to severe cough, bronchiolitis, pneumonia and exacerbation of preexisting respiratory conditions. The presentation is similar to respiratory syncytial virus infection and may also include high fever, myalgia, rhinorrhea, dyspnea, tachypnea, and wheezing.</p> <p>Co-infection with hMPV and respiratory syncytial virus can occur, and is generally assume to associated with worse disease.</p> <p>Currently there are no vaccines or antiviral drugs against this virus.</p> <p>MODE OF TRANSMISSION:</p> <p>hMPV is spread by contact with respiratory secretions of infected persons by aerosols and droplets (from coughing and sneezing) or contaminated objects/surfaces/fomites. The incubation and contagious periods are not well defined but are probably similar to those of respiratory syncytial virus.</p> <p>HOST RANGE:</p> <p>Humans; however, various animal species can be experimentally infected with hMPV including cotton rats, mice, ferrets, guinea pigs, hamsters, marmosets, lambs, and nonhuman primates.</p> <p>hMPV is closely related to the avian metapneumovirus (AMPV) subgroup C.</p> <p>ZOONOSIS: None.</p> <p>VECTORS: None.</p> <p>*Staff member with common cold/flu, should not work with hMPV.</p> <p>*Researchers are asked to be vaccinated (against flu) before they begin working with the virus.</p>
2. Designated	ABSL-2 facility.

Area	
3.Training	Practical experience with animal care/maintenance, as well as general biosafety, is required.
4. Personal Protective Equipment (PPE)	Gloves, Eyes safety goggles, Lab coat, Disposable shoe covers and Animal handling gown.
5.General . Precautions for Animal Use	Inhalation of virus from aerosols, generated when aspirating, dispensing, or mixing virus-infected samples (tissues, feces, secretions) from infected animals. Laboratory infection can also occur from direct inoculation of mucous membranes via virus contaminated gloves following the handling of tissues, feces and/or secretions from infected animals.
6. Environmental / Ventilation Controls	<p>Work should be conducted in ABSL-2 facility, over absorbent pads in a class II type A1 or A2 biological cabinet.</p> <p>SURVIVAL OUTSIDE HOST: hMPV is suspected to be vulnerable to environmental changes, particularly temperature and humidity. It is sensitive to high and low temperature, and to drying; i.e., low humidity levels. It may survive for about 3 to 30 hours on nonporous surfaces at room temperature.</p>
7. Animal handling practices	<ol style="list-style-type: none"> 1. Animals must be housed in filter top cages marked as biohazards (including the name of the pathogen/biohazard). Handling the cages (including bedding) will be done only by the researchers. 2. Use a class II Biological Safety Cabinet at all times (especially during injection or any surgical procedure), when performing work on these animals and/or when moving animals from dirty to clean cages. 3. Infected animals may shed hMPV after treatment; take precautions to avoid the creation of aerosols when changing or washing cages, or cleaning the room. 4. Dead animals must be placed in primary plastic bags, which are then placed in biosafety bags for infectious waste incineration. 5. All surfaces and racks that may be contaminated will be decontaminated with 0.5% bleach (or virusolve), ASAP. 6. When changing cages, use a standard microisolator technique: <ul style="list-style-type: none"> • place the cage containing the animals, under the biological safety cabinet and transfer the animals into a clean cage.

	<ul style="list-style-type: none"> • spray the dirty cage with 0.5% bleach (or virusolve), remove from the safety cabinet and place on a transfer rack . • when all cages have been changed, spray the dirty cages and rack again with 0.5% bleach, and cover the rack. Put on a pair of new gloves and bring the rack directly to the autoclave in the dirty cage wash area. • immediately autoclave the dirty cages (1 hour at 121°C/250° F, 15psi of steam pressure). Once the autoclave cycle is completed, the cages can be emptied and the bedding disposed of in a normal fashion. <p>**In cases where the use of autoclave (within the animal facility) is not an option:</p> <ul style="list-style-type: none"> • the cages (bedding) must be emptied inside the BSL-2 cabinet, directly to a double biohazard bags. • Before closing the bags, carefully, add a small amount of water (250ml) to improve the sterilization process. <p><i>Do not close the bag completely/tightly (in order to aloud entering of steam during the sterilization process).</i></p> <ul style="list-style-type: none"> • Spray the dirty bag with 0.5% bleach or virusolve. • Remove from the safety cabinet and place on a transfer rack/container. • Put on a pair of new gloves and bring the rack/container, directly to the collection point of your department.
8.Decontamination	<p>SUSCEPTIBILITY TO DISINFECTANTS: hMPV susceptible to ether, chloroform, and a variety of detergents, including 0.1% sodium deoxycholate, sodium dodecyl sulphate, and Triton X-100. It may also be sensitive to hypochlorites (1% sodium hypochlorite), formaldehyde (18.5 g/L; 5% formalin in water), 2% glutaraldehyde, and iodophores (1% iodine).</p> <p>PHYSICAL INACTIVATION: RSV is sensitive to heating above 55 °C for 5 minutes (up to 90% decrease in infectivity)(1). It is also sensitive to freezing and thawing (~90% loss in infectivity following each freeze-thaw cycle). It is also sensitive to acidic media (pH<7).</p> <p>Susceptible to moist heat at 121°C for 20 minutes or dry heat at 170°C for 1 hour, 160°C for 2 hours, or 121°C for at least 16 hours.</p>
9. Spill and Accident Procedures	<ol style="list-style-type: none"> 1. Evacuate area, remove contaminated PPE and allow agents to settle for a minimum of 30 minutes. Initiate spill response procedure. 2. Cover the spill with absorbent material. Starting at the edges and work towards the center.

	<ol style="list-style-type: none"> 3. Carefully pour disinfectant over the absorbed spill, again starting at the edges. Saturate the area with disinfectant. 4. Allow sufficient contact period to inactivate the material in the spill. Non-viscous spills require 15-20 minutes: viscous spills require 30 minutes. 5. Use paper towels to wipe up the spill, working from the edge to center. Use tongs or forceps to pick up broken plastics, glass or other sharps that could puncture gloves. 6. Discard absorbent material in Chemical waste bags. 7. Clean the spill area with fresh paper towels soaked in disinfectant. Thoroughly wet the spill area, and wipe with towels. 8. Discard all cleanup materials in Chemical bag, along with any contaminated PPE (pay special attention to gloves and shoe covers). Close and secure the bag. 9. Place bag in a second Chemical bag, secure and dispose as chemical waste. 10. Discard contaminated PPE (with biohazard materials) in biohazard bag. Place bag in a second biohazard bag, secure and disinfect by autoclaving. <p><u>Exposure:</u></p> <ol style="list-style-type: none"> 1. In case of skin contact or injection with hMPV, wash the affected area with soap and water for at least 15 minutes. 2. For eye exposure, flush with water for at least 15 minutes. Consult with Employee Health Center,. Report incident to supervisor. Supervisor reports the accident/injury to the Biosafety Unit.
10. Waste Disposal	Autoclave all waste (1 hour at 121°C/250°F, 15psi of steam pressure).
I hereby confirm that I have read the SOP (Standard Operating Procedure) for Working with hMPV in Animals, and agree to follow these procedures.	
Name:	Title:
Signature:	Date:

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