Tel-Aviv University –Safety Unit

Standard Operating Procedure for 6-Hydroxy dopamine (6-OHDA) in Animals.	
1. Health hazards	6-Hydroxy dopamine (6-OHDA) is neurotoxin that destroys catecholamine terminals and can cause Parkinsonism, ataxia, and other motor disturbances in humans if it enters the blood stream in significant amounts. At low concentrations 6-OHDA can cause irritations to the respiratory tract, digestive tract, the eyes and skin. **Hazard Statements:** Harmful if swallowed Harmful in contact with skin Causes skin irritation Causes serious eye irritation Harmful if inhaled May cause respiratory irritation. There has been concern that this material can cause cancer or mutations, but there is not enough data to make an assessment. **Chronic exposure** to phenethylamines excite the central nervous system and induce tolerance; in extreme cases they produce amphetamine-like responses including personality changes, compulsive and stereotyped behavior and may induce psychosis with auditory and visual hallucinations and paranoid delusions. * Pregnant and lactating women should avoid exposure to 6-OHDA and animals that have been administered 6-OHDA, **or** use additional PPE (respirator). ***I**mmunocompromised individuals should also use extreme caution when handling 6 -OHDA.
2. Designated Area	ABSL-2 facility.
3.Training	Hazardous chemical training and training on this SOP is required before working with 6-OHDA. This should include but is not limited to reviewing the MSDS, training on the physical hazards of the chemicals, symptoms of exposure, appropriate work practices, and proper use of PPE.
4. Personal	Nitrile gloves, Chemical safety goggles, Lab coat and mask. Appropriate PPE should

Protective Equipment (PPE)	also be used for lower arms such as sleeve covers or securing gloves over the sleeves of laboratory coat. ***Personnel should not work with 6-OHDA if skin is cut or scratched.***
5.General . Precautions for Animal Use	Tools (as, syringe, blades and safety needles where possible) should be adapted for BSL-2. Have a sharps container in close vicinity. Animals should be restrained or anesthetized during injection.
6. Environmental / Ventilation Controls	The preparation of 6-OHDA including reconstitution, weighing, and diluting should be performed in a fume hood or biological safety cabinet (class II Type B). Work should be done over absorbent pads. Work should be conducted in ABSL-2 facility, over absorbent pads in a class II type A1 or A2 biological cabinet.
7. Special Handling Procedures & Storage Requirements	Handling : 6-OHDA should be handled in containment and done over absorbent pads. Utilize safe sharps procedures (i.e. sharps container in the immediate vicinity, Leurlock syringes are recommended). The fume hood or other approved containment must be cleaned upon completion of tasks. When transporting 6-OHDA, the vials should be placed in secondary, sealed, plastic, labeled, non-breakable containers. Storage: Store in a well-ventilated place. Keep container in a dry place, tightly closed. Do not store next to strong oxidizing agents or strong bases.
8. Precautions for Animal Use	No recapping needles. Have a sharps container in close vicinity. Animals should be restrained or anesthetized during injection. **Once 6-OHDA is injected, animals , animal waste and cages are considered hazardous.**
9. Animal handling practices	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. **Injecting animals with 6-OHDA:** Animals will be injected IP with 6-OHDA within Class II Biosafety cabinet or designated chemical fume hood. All needles will be disposed of in sharps container – do not recap or bend needles. 4. Infected animals considered hazardous after each administration of 6-OHDA; take precautions to avoid the creation of aerosols when changing or washing cages, or

	cleaning the room. A respirator is recommended for personnel that are immunocompromised or pregnant and for healthy personnel if work is done outside the ventilated cabinet . 5. Care should be taken to avoid exposure to bedding dust when handling exposed animals and their waste materials during this time. 6. Dead animals must be placed in primary plastic bags, which are then placed in biosafety bags for infectious waste incineration. 7. All surfaces and racks that may be contaminated will be decontaminated with detergent solution followed by water ASAP. 8. The bedding is considered contaminated and requires special handling. When changing cages, use the following technique: • Transfer the animals to clean cages . • Insert the used cages in a plastic bag . • Twist the ends of full bags, and seal with tape. Label with wide tape or other type of label marked “toxin-6-OHDA ”. • Transport the bags of cages to a HEPA filtered dumping station that draws air away from the use. .(it is recommended to use a mask) or fume hood. • All contaminated bedding will be labeled as hazardous materials and handled accordingly : incinerated or placed in chemical waste bags for disposal. • Animal carcasses will be incinerated or handled as Chemical waste. • The cages should then be put in plastic bags (marked “toxin-6-OHDA) and sealed for transport to the washroom. • In the washroom ,cages should be unloaded from the bags with the appropriate PPE as mentioned above and run through the cage wash in the conventional manner. Note- cage wash personnel that meet the criteria for extra precautions above (pregnant exc.) should take extra precautions (additional PPE) when handling cages that may have 6-OHDA contamination.
10. Spill and Accident Procedures	1. Spills must be cleaned immediately by properly protected trained personnel. Do not let product enter drains. 2. **Liquid Spills:** should be cleaned immediately by personnel wearing a gown, goggles, two pairs of gloves (nitrile). Use absorbent pads to wipe liquid. The spill

	area should then be cleaned thoroughly with a detergent solution followed by clean water. Place waste in plastic bag and then in the chemical waste container. 3. **Powder Spills**: should be cleaned immediately by personnel wearing a gown, goggles, two pairs of gloves (nitrile). For powder spills outside of a fume hood or approved containment, non-essential personnel should be instructed to leave the laboratory and entrance should be restricted. In addition to the above specified PPE, A respirator should also be worn. The spill area should then be cleaned thoroughly with a detergent solution followed by clean water. Place waste in a plastic bag and then in the chemical waste container. **Exposure:** 4. In case of skin contact or injection with 6-OHDA, wash the affected area with soap and water for at least 15 minutes. Consult with Employee Health Center. 5. 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.
11. Waste Disposal	Dispose all waste material in the appropriate chemical waste container. Any unused reagent , any mixtures of 6-OHDA , and any spill clean-up debris should be managed as chemical waste.
I hereby confirm that I have read the SOP (Standard Operating Procedure) for Working with 6-Hydroxy dopamine (6-OHDA) in Animals, and agree to follow these procedures.	
Name:	Title:
Signature:	Date:

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