

Work with MPTP & MPTP-Treated Animals

INTRODUCTION

MPTP (1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine) is a neurotoxin used to induce parkinsonism symptoms in experimental models of Parkinson's disease (PD). MPTP can produce irreversible neurological damage indistinguishable from PD in humans and animals. The inappropriate handling of MPTP may result in exposure and irreversible neurological damage to research staff.

MPTP is lipophilic which freely and rapidly crosses the blood-brain barrier. It is metabolized to MPDP+ which is oxidized to the active metabolite MPP+. Excretion of unmetabolized MPTP occurs mainly during first day post injection while MPTP metabolites are excreted up to 4 days post injection. Excretion is mainly in the urine and some has been found in bile. MPTP in urine is expected to be ionized and not volatile and be absorbed by the bedding.

Exposure to staff may occur via needlesticks and inhalation of vapors from MPTP solutions, or inhalation of aerosols from MPTP contaminated bedding. Exposure potential is greatest during preparation, administration, and in the bedding during the first 4 days after injection. The high risk period will be from preparation of MPTP solutions to 4 days post administration after the first cage change.

The purpose of this SP is to help ensure the safe work practices and appropriate medical surveillance for people working with MPTP. A lab specific SP must be approved by UW Safety prior to IACUC approval. For questions or assistance in implementing this SP, contact the UW Safety at 307-766-3277.

1. **PRIOR TO BEGINNING WORK WITH MPTP** (All of the following items are required for employees who plan to work with MPTP or enter MPTP work areas up to 4 days post administration)
 - A. Medical Management: Prior to initiating work with MPTP, the Principal Investigator (PI), and his/her staff must contact the Student Health Service (SHS) -UW Campus at 307-766-2130 to schedule an appointment for evaluation and to receive the post-exposure prophylaxis (PEP) of selegiline hydrochloride (Eldepryl). SHS will prescribe the selegiline.
 1. Selegiline is obtained prior to beginning work with MPTP. It must be kept in the white capped container labeled "FOR MPTP USE ONLY" and readily accessible to those working with MPTP. It must be regularly checked to ensure it is in its proper place and the use date has not expired.
 2. The PI must send all personnel who intend to work with MPTP to the SHS prior to beginning work and annually thereafter for evaluation and determination of their ability to successfully take selegiline. All personnel must visit SHS for Respiratory Protection medical evaluation (i.e. N95) Program enrollment as well.



- B. Respiratory Protection: Respirators are required when working with MPTP treated animals. You *MUST* be enrolled the University's Respiratory Protection Program prior to wearing a respirator. Contact the UW Respirator Program Administrator at UW Safety at 307-766-3277 to initiate this process.
- C. Employee Training and Notification:
1. The PI must train his/her staff on this Safety Procedure (SP) and hazards of MPTP prior to beginning work. This training must be documented. For assistance, contact UW Safety at 307-766-3277.
 2. If animals will be housed prior approval by the IACUC is required. This document be forwarded to UW Safety for review. Post the approved protocols in the animal housing areas two weeks prior to administering MPTP.

ACCIDENTS, INJURIES, AND POTENTIAL EXPOSURES

- A. In the event of a **recognized** percutaneous or mucous membrane exposure to MPTP:
1. Immediately take prescribed selegiline as directed by the SHS.
 2. Initiate first aid at the worksite:
 - a. Contaminated skin should be washed with copious amounts of soap and water for 15 minutes.
 - b. Contaminated eyes and mucous membranes should be irrigated for 15 minutes, using normal saline or water.
 3. Quickly contact the Student Health Service (SHS) on main campus at 307-766-2130 between 8:00 a.m. and 5:00 p.m. M-F. If the SHS is closed or not available, go directly to the Ivinson Memorial Hospital Emergency Room (ER). Be sure to disclose your name, your exact location, and that you were exposed to MPTP.
 4. Notify the PI as soon as possible. If he/she is not available, contact secondary people. Have these emergencies readily accessible in the laboratory.
 5. Report the incident to your supervisor who will fill out the required workers compensation forms.
- B. In the event of a **possible** exposure to MPTP:
1. Immediately initiate first aid at the worksite as outlined above.
 2. Quickly contact the SHS on main campus at 307-766-2130 between 8:00 a.m. and 5:00 p.m. M-F. If the Student Health Service is closed or not available, go directly to Ivinson Memorial Hospital Emergency Room (ER). Be sure to disclose your name, your exact location, and that you were exposed to MPTP.
 3. Notify the PI as soon as possible. If he/she is not available, contact secondary people. Have these emergencies readily accessible in the laboratory.
 4. Report the incident to your supervisor who will fill out the required workers compensation forms.
 5. In the event of a spill outside of a chemical fume hood, evacuate people from the room and close off the area. Contact the UW Safety - RMMC at 307-766-3698.



PERSONAL PROTECTIVE EQUIPMENT (PPE)

- A. Personal protective equipment must be worn whenever working with MPTP or MPTP-treated animals and their waste within 4 days of last administration. ***These activities include preparing MPTP solutions, administering MPTP to animals, entering MPTP-animal room within first 4 days of administration, first cage change (must be no earlier than 4 days after last administration):***
 - Laboratory coat
 - Rear closing disposable water proof gown
 - Sleeve covers (wrist guards)
 - Double nitrile gloves
 - Chemical safety goggles
 - N95 disposable respirator
- B. *Cage changing*: Must be done no earlier than 4 days after last administration. If emergency occurs and cages must be changed within 4 days of last administration, use the above PPE.
- C. Frequent glove changes are recommended. Change gloves immediately if their integrity has been compromised (e.g., soiled, punctured, torn).
- D. Protective clothing should be changed prior to entering non- MPTP designated areas.
- E. PPE must be removed in a specific order to prevent contamination. When removing PPE,
 1. remove outer gloves
 2. sleeve covers
 3. disposable gown
 4. chemical goggles
 5. N95 respirator
 6. Remove inner pair of gloves
 7. Put on a new pair of gloves, spray goggles with 1% bleach (wipe),
 8. Remove laboratory coat
 9. Remove the new gloves, wash hands, and exit the MPTP administration area.

MPTP ACCESS RESTRICTIONS AND PPE

Procedures	PPE	Restricted Access
MPTP Preparation and administration	Lab coat, rear closing water resistant gown, N95 respirator, double nitrile gloves, sleeves covers, chemical safety goggles	Room restricted to only those staff authorized to work with MPTP.
Animal housing for 1-4 days post last injection	Lab coat, N95, double-gloves, sleeves covers, chemical safety goggles	Room restricted to only those staff authorized to work with MPTP.
Animal housing after 4 days post injection	Standard laboratory PPE: Lab coat, gloves, chemical safety goggles	Room No restricted access



MPTP PREPARATION

- A. Order *PRE-WEIGHED MPTP* from the manufacturer. MPTP must be purchased in hydrochloride or tartrate salt form rather than as a free base.
- B. MPTP and MPTP-solutions must be stored in labeled, tightly capped containers. The container label must include the identity of the hazardous contents (i.e., MPTP) and the appropriate hazard warning (i.e., neurotoxin). The primary container for MPTP must be placed in a sealed, leak-proof, unbreakable secondary container, which must also be labeled as above.
- C. MPTP, as received from the manufacturer/vendor in its original undiluted form, must be stored in a clutter free certified chemical fume hood in a labeled, sealed, leakproof secondary container. ***The hood must be posted with the red MPTP sign (at end of document)***
- D. *During preparation and administration of MPTP*: ALL of Room is restricted to only those staff authorized to work with MPTP.
- E. The room where the MPTP fume hood is located is restricted to staff authorized to work with MPTP and MPTP-treated animals for a minimum of 4 days after last MPTP administration. All staff working with MPTP and MPTP-treated animals must follow all of the steps in the SP. PPE must be changed prior to exiting the room.
- F. If emergency maintenance must be done during the first 4 days after the injection, stop all work and decontaminate the area. Ensure that all concentrated and dilute concentrations of MPTP are closed and properly stored. Please escort the worker into the space and remain in the room during the entire maintenance. The sign should remain on the door and fume hood to inform other staff of the MPTP and MPTP treated animals. After the 4 days post injection, no extra PPE is required in this space and any requirements for escorts will be determined by lab staff.
- G. MPTP solutions must be prepared in a certified chemical fume hood. Appropriate personal protective equipment (PPE) must be worn, as noted in Section III. All work surfaces must be covered with absorbent, plastic-backed, disposable absorbent pads.
- H. ***The red MPTP sign (at end of document) must be posted on door and outside the MPTP fume hood is located for a minimum of 4 days after last MPTP administration.***
- I. Disposable lab-ware should be used when preparing MPTP solutions. If non-disposable glassware is used, it must be triple rinsed in 1% bleach prior to washing. The rinsate must be collected and disposed of as chemical waste (see Section G: MPTP Waste).
- J. When all work with MPTP is complete, carefully remove all absorbent pads and dispose as regular garbage. Wipe all surfaces with a 1% bleach solution and double bag the wipes or paper towels and discard with regular garbage.

ADMINISTERING MPTP

- A. Animals must be chemically or physically restrained prior to starting the MPTP administration.
- B. Administration of MPTP to rodents must be conducted in a certified chemical fume hood or other UW Safety approved containment. Call UW Safety at 307-766-3277 to evaluate containment other than a chemical fume hood. Both the personnel administering the MPTP and those in the immediate vicinity of the procedure must wear appropriate PPE (see section III).

- C. All work surfaces must be covered with absorbent, plastic-backed, disposable absorbent pads.
- D. Use only needle-locking syringes or disposable syringe-needle units (i.e., needle is integral to the syringe). Used disposable needles must not be bent, sheared, broken, recapped, removed from disposable syringes, or otherwise manipulated before disposal.
- E. After each injection, immediately place the syringe-needle unit in a sharps disposal container located in the fume hood or approved containment.

TRANSPORTING MPTP TREATED ANIMALS

- 1. Prepare animals for transport in a fume hood:
- 2. Secure the cage tops to the cage bottoms with tape.
- 3. Wipe down the cages with 1% bleach.
- 4. Pack up to 4 cages in the transport box.
- 5. Secure the transport lid using the snaps.
- 6. Carefully transport the animals to the new fume hood or animal housing room.
- 7. Ensure no cage has opened during transport.
- 8. Remove cages from the transport box into the chemical fume hood.

ANIMAL HOUSING

- A. Animals will be housed in room _____ where the fume hood is located.
- B. For the first 4 days post-injection, the door to each MPTP-animal room (the fume hood in room _____ is the designated location for preparing, administering, and housing animals) must be posted in such a manner that it is clear that the fume hood contains MPTP-treated animals. The fume hood may only be used for the MPTP work (preparing, administering MPTP and housing animals). **Post the sign (at end of this document) on the chemical fume hood and outside the room containing the chemical fume hood.**
- C. The animal cages must also be properly labeled.
- D. For the first 4 days post-injection, cover the cages with filter bonnets. Place cages in a certified chemical fume hood or ventilated rack. Ensure the lids are secured to the cage.
- E. Cage changes **should not** be performed for the first 4 days post-injection.
- F. 4 days after the final MPTP injection, the animals may be transferred to clean, standard caging and the MPTP door signs and cage tags may be removed.
 - 1. Cage change/dumping should be performed wearing appropriate PPE (section III) and in a chemical fume hood or other EHS approved containment. Work atop absorbent pads. This local exhaust ventilation should be wiped down with detergent and water after use.
 - 2. Dirty cages and racks should be wiped down with 1% bleach and covered with a full drape and moved to the cage wash for immediate cleaning.
 - 3. Reusable water bottles can be sprayed with 1% bleach and processed same as other water bottles.
- G. The use of special PPE is not required after the animals have been transferred to clean cages (beyond 4 days post-injection).



MPTP WASTE

1. Unused concentrated reagent (including empty vials) is managed as hazardous waste through UW Safety. See: <http://www.uwyo.edu/safety>
2. Spill cleanup materials, soiled PPE, and absorbent pads grossly contaminated with concentrated reagent should be double bagged and managed as hazardous waste through UW Safety as above.
3. Grossly contaminated solids can be decontaminated with 0.1 N hydrochloric acid or 1% bleach. After decontamination, the solids can be disposed with regular garbage or reused. The liquid waste containing dilute MPTP and decontaminant should be managed as chemical waste through UW Safety.
4. Mildly contaminated solids (including gowns and gloves believed to not have contact with MPTP) can be double bagged and discarded with regular garbage.
5. Animal bedding: Bedding from first change (4 days after last administration) must be incinerated via Stericycle. Complete the UW Safety - RMMC hazardous waste form at: <http://www.uwyo.edu/safety> to arrange for proper containers and pick-up. **Specify that pick-up is for incineration.** Bedding from subsequent cage changes can be double bagged and discarded through the regular waste stream.
6. Animal carcasses: Carcasses that have received MPTP within the last 4 days must be incinerated via Stericycle. Contact UW Safety - RMMC at 307-766-3698 to arrange for proper containers to be delivered and pick-up. **Specify that pick-up is for incineration.** Carcasses that have not received MPTP in the past 4 days can be double bagged and discarded through the regular waste stream.

SIGNAGE

POST ON FUME HOOD:



MPTP NEUROTOXIN

POST ON DOORS until 4 days post last administration:



MPTP NEUROTOXIN

ENTRY RESTRICTED TO AUTHORIZED*PERSONNEL

MPTP – TREATED LIVE ANIMALS NEUROTOXIN

Name (print) (Assessor):	Title:
Signature (Assessor):	Date:
Name (print) (PI, Lab Manager, or Unit Head):	Title:
Signature (PI, Lab Manager, or Unit Head):	Date:
Date Sent to UW Safety:	

References:



1. Kopin, I.J. 1987. MPTP: An industrial chemical and contaminant of illicit narcotics stimulates a new era in research on Parkinson's disease. *Environmental Health Perspectives*. **75**, 45-51.
2. National Institutes of Health (NIH), Office of Research Services, Division of Occupational Health and Safety: Procedures for Working with MPTP or MPTP-Treated Animals. [http://dohs.ors.od.nih.gov/pdf/Procedures for Working with MPTP or MPTP Treated Animals_july%202006.pdf](http://dohs.ors.od.nih.gov/pdf/Procedures_for_Working_with_MPTP_or_MPTP_Treated_Animals_july%202006.pdf)
3. Przedborski, Serge, Jackson-Lewis , V., Naini, A.b., Jakowec , M., Petzinger , G., Miller, R., Akram , M. 2001. The parkinsonian toxin 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine (MPTP): A technical review of its utility and safety. *Journal of Neurochemistry*. **76**, 1265-1274.
4. Yang, S.C., Markey, S.P., Bankiewicz, K.S., London, W.T., Lunn, G. 1988. Recommended safe practices for using the neurotoxin MPTP in animal experiments. *Laboratory Animal Science*. **38**, 563-567