Tufts University Standard Operating Procedure (SOP) for: Ethidium Bromide

CAS # 1239-45-8
Synonyms: EtBr, 3.8-Diamino-5-ethyl-6-phenylphenanthridinum bromide, Homidium Bromide
NFPA 4, HMIS 3

Toxicity

Ethidium bromide (EtBr) is commonly used in molecular biology laboratories for visualizing nucleic acids using electrophoresis and other gel-based nucleic acid separation methods. Ethidium bromide fluoresces when exposed to ultraviolet light and exhibits a vivid red-orange color.

Ethidium bromide is a powerful mutagen and is a skin, eye and respiratory irritant. It can be readily absorbed through the skin. Avoid direct skin contact. Inflammation and discoloration of the skin may occur after contact. Contact will stain the skin purple. Lesser exposures may cause coughing or sneezing, and irritation of the skin and of the mucous membranes of the eyes and respiratory system.

Ethidium bromide can be purchased as powder or in solution. The crystal or powder form is odorless and appears dark red in color. The powder form is considered an irritant to the upper respiratory tract, eyes, and skin. Handling powders are more hazardous than handling premade solutions. Thus, it is recommended that EtBr be purchased as a solution in an effort to reduce contamination and exposures. Ethidium bromide is strongly mutagenic, causing living cell mutations. While there is no evidence, at this time, of human carcinogenicity or teratogenicity, this material is considered by Tufts University Policy as a possible carcinogen or teratogen.

Good Practices and Personal Protective Equipment (PPE)

Ethidium Bromide Powder: If it is necessary to use EtBre in crystal form, lab workers shall do the following (laboratory personnel are strongly encouraged to purchase aqueous stock solutions).

- Wear gloves, standard laboratory clothing; a fully-buttoned lab coat, long pants, closed-toe shoes, safety glasses and nitrile gloves.
- If there is a risk of splash, wear a full face shield.
- Ensure that the entire hand and arm area is covered.
- Use a laboratory chemical fume hood to measure out the ethidium bromide
- Prepare your solutions in the laboratory chemical fume hood
- Scan for contamination using the appropriate UV wavelength

Pouring gels and post electrophoresis

- Cover the bench area with bench paper
- Wear appropriate PPE (gloves, eye protection, lab coats) when setting up, pouring, and handling ethidium bromide gels/solutions.
- Change gloves after handling ethidium bromide.

- Scan for contamination using the appropriate UV wavelength*

**Caution is required when using and ultraviolet light source for work with EtBr. As a general rule, avoid exposing unprotected skin and eyes to intense UV sources for prolonged work close to UV light boxes or other intense sources. UV protective eyewear shall be worn when visualizing EtBr (face shield). Wash hands thoroughly before and after removing gloves and scan for contamination with a UV source.*

Incompatibilities
EtBr is stable under ordinary use and storage conditions. Toxic gases and vapors may be released if involved in a fire. EtBr is incompatible with strong oxidizers (such as Bleach) and dust formation should be avoided.

Spills
If you spill a small amount (minor spill) of EtBr notify your supervisor for cleanup assistance. A major spill is any amount of chemical that the lab staff cannot easily and safely clean up without outside assistance. In this case, vacate the lab and call your supervisor and TUPD at 6-6911.

Small liquid spill:
Use ultraviolet light to locate spill, absorb freestanding liquid with dry paper towels or absorbent material and place into appropriate waste containers for disposal. Wet the surface with ethanol (dissolves EtBr), after ensuring no ignition sources are present. Dispose of in a plastic bag for disposal as hazardous waste. Use ultraviolet light to verify cleanup.

Small dry spill:
Don appropriate PPE. If potential respiratory hazard exists, contact TEHS.
If the powder is spilled, wet paper towels and carefully lay on top of the powder to avoid creation of airborne dust when cleaning the spill and place into appropriate waste containers for disposal. Once recovered wet the surface with ethanol and wipe the area from the outer edge to the center of the spill with dry paper towels and dispose of in a plastic bag for disposal as hazardous waste. Use ultraviolet light to verify cleanup.

Accident/Exposures
If case of skin contact, immediately remove any contaminated clothing and wash the area with soap and water for 15 min. **Do not use disinfecting chemicals** such as ethanol or bleach as this could actually increase skins permeability to EtBr. In case of eye contact, rinse the eyes with large amounts of water for a minimum of 15 min. and seek medical attention. If inhaled, move the person to fresh air and seek medical attention. In the event of ingestion, call your supervisor and X66911 and seek medical attention immediately. Do not induce vomiting unless directed to do so by medical personnel. If spill is on equipment, use ultraviolet light to locate spill (if can be done safely), then decontaminate.
As with all teratogenic or suspect teratogenic substances: use by pregnant persons warrants careful consideration between you and your supervisor. Contact TEHS for additional information.

Disposal:
General housekeeping measures including proper storage, use of disposable bench paper, and cleaning of contaminated work area(s) and equipment at the conclusion of work.

All EtBr waste should be contained in a sealable container, labeled with a completed chemical waste label, and placed in the nearest SAA. Contact TEHS for waste pick up at 6-3615.

Waste Stream	Description	Waste Disposal Procedure
Buffers	Typically contain very small concentrations of EtBr (<0.5 mg/L)	Dispose of via sanitary sewer (laboratory sink disposal)
Stock solutions	Typically contain higher concentrations of EtBr (1–10 mg/ml)	Dispose as hazardous waste. Place the container in the labs SAA and contact EH&S for a waste pick-up.
Gels	Typically contain lower concentrations of EtBr (3–5 mg/L)	Place gels in a single lined black poly five (5) gallon pail. Contact EH&S for a waste pick-up.
Contaminated Debris	Gloves, spill cleanup materials, and other lab supplies contaminated with EtBr	Broken glassware and sharps must be placed in puncture-resistant containers. Other debris may be placed in clear, labeled bags. Place the bags/containers in the labs SAA and contact EH&S for a waste pick-up.
Crystals and powders	Typically pure or concentrated EtBr	Dispose of EtBr crystals and powders as hazardous waste. Place the container in the labs SAA and contact EH&S for a waste pick-up.

References:

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