Ethidium Bromide Safety Procedure

Ethidium Bromide (EtBr) is a red cationic fluorescent dye that may be used for, among other things, to visualize DNA and RNA in electrophoresis gels and as a protein synthesis inhibitor. EtBr is a dark red, crystalline, solid, moderately soluble in water, and that fluoresces readily with a reddish-brown color when exposed to ultraviolet (UV) light. Due to EtBr’s hazardous properties, special safety precautions are required. For safety assistance, contact EH&S at x3347.

Health Hazards
EtBr is a potent mutagen (may cause genetic damage), and moderately toxic after an acute exposure. It can be absorbed through skin, so it is important to avoid any direct contact with the chemical. EtBr is an irritant to the skin, eyes, mouth, and upper respiratory tract.

Safety Precautions
• Consider using a less toxic chemical (e.g., SYBR Safe™ DNA gel stain) to reduce potential hazardous exposures and amounts of hazardous waste generated. Contact EH&S for assistance.
• EtBr work areas must be posted: Ethidium Bromide – Caution: Mutagen and Carcinogen. Both the size of these areas and the amount of EtBr in storage and use should be minimized.
• Before work begins, all lab personnel must review the Material Safety Data Sheet (MSDS) and be trained on the hazards and exposure controls for EtBr. It is recommended that lab-specific procedures be developed and followed.
• To prevent inhalation exposure, work with EtBr powder or crystals in a fume hood or work with premixed EtBr solutions or tablets to avoid handling the powder directly.
• When using EtBr outside of your work area or lab (e.g., transporting samples), put on clean gloves to prevent cross-contamination of doorknobs and other surfaces. When transporting, carry EtBr in a shatter-proof, leak-proof secondary container.
• Wipe down work area with soap and water when work is completed each day and properly dispose of contaminated towels, rags, and wipes. See disposal requirements below.
• All EtBr users are responsible for maintaining a clean lab area. Any area of suspected contamination should be promptly cleaned. Use a UV light to check for contamination. Remember to wear safety glasses or goggles when using a UV lamp and to protect your skin from exposure.
• Do not use bleach solutions to clean up EtBr contamination, as it is not effective and can be explosive under certain conditions.

Personal Protective Equipment (PPE)
• Protective Clothing: Wear a fully-buttoned lab coat, long pants and closed-toe shoes.
• Eye Protection: Wear safety glasses with side shields or chemical splash goggles when there is a splash hazard.
• Gloves: Use disposable nitrile gloves and change frequently to minimize cross-contamination. Wash hands thoroughly after removing gloves.
Emergency Exposure Procedures
- Seek immediate medical attention after a suspected EtBr exposure.
- **Eye Contact:** Immediately irrigate. Hold eyes open and irrigate for 15 minutes.
- **Skin Contact:** Remove contaminated clothing. Immediately wash affected areas with soap and water.
- **If swallowed or inhaled:** In the case of ingestion, obtain medical attention immediately. If dust is inhaled, move the patient to a source of fresh air and seek medical attention, x2911.

Spill Procedures
- For **hazardous spills**, evacuate and restrict access to the laboratory. Call x2911 for assistance.

  - **Incidental spills** (small, low hazard) of EtBr solutions may be cleaned by laboratory staff. Wear appropriate PPE at all times. Use paper towels to absorb any liquid. For solids (crystal or powder), place wet paper towels over the top to prevent generation of dust, wipe up, and properly dispose. Once liquid or solid material is cleaned up, the area must be decontaminated and surveyed. Use a UV light to locate any remaining EtBr and then follow one of the decontamination procedures below or an equivalent method.

  **Method 1:**
  - Scrub the area with clean wet paper towels and soap and water.
  - Repeat this step six times to remove contamination. (The process of scrubbing six times represents the average number of times to reduce contamination to undetectable levels. This is an accepted practice in laboratories across the United States.)
  - Properly dispose of all paper towels and contaminated PPE.
  - Use a UV lamp to check the area for any remaining contamination. If contamination remains, repeat the process until the area is free of detectable contamination.

  **Method 2:**
  - Just prior to using EtBr, prepare a decontamination solution of 4.2 g of sodium nitrite and 20 ml of hypophosphorous acid (50 percent) in 300 ml of water.
  - Wash the area with a paper towel soaked in decontamination solution. Rinse the area five times with paper towels soaked in tap water, using a fresh towel each time.
  - Use a UV lamp to check the area for any remaining contamination. If contamination remains, repeat the process until the area is free of detectable contamination.
  - Soak all the towels in decontamination solution for one hour; double-bag along with contaminated gloves and other items and properly dispose.

Disposal
- All EtBr wastes (liquid, solid and debris) must be managed as hazardous waste.
  - Collect all EtBr solutions in plastic containers and secure lid.
  - Collect all solid materials (gels, gloves, towels) in a plastic lined pail and secure lid.
  - Double bag all wetted towels or absorbent materials before placing in a plastic lined pail.
- Label all waste containers with a red hazardous waste tag and submit a Waste Pickup request using the online form on the EHS website. Check toxic/poison hazard box on tag.
- Contaminated sharps (needles, syringes, slides, broken glass, etc.) shall be discarded in an infectious waste sharps container labeled “MUTAGEN/CARCINOGEN SHARPS-DO NOT AUTOCLAVE”.
Acknowledgement

- PIs/Supervisors are responsible for providing lab specific training, reviewing emergency specific procedures with lab personnel, and implementing safe lab practices and engineering controls. PIs/Supervisors and lab personnel must sign below that they have reviewed and understand this procedure.

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