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EH&S FACT SHEET

Environment, Health and Safety Information for the Berkeley Campus

Ethidium Bromide
Hazards and Precautions

Ethidium bromide (EtBr) is commonly used as a non-radioactive marker for identifying and visualizing nucleic acid bands in electrophoresis and in other methods of nucleic acid separation. EtBr is a dark red, crystalline, non-volatile solid, moderately soluble in water, which fluoresces readily with a reddish-brown color when exposed to ultraviolet (UV) light. Although it is an effective tool, its hazardous properties require special safe handling and disposal procedures.

Health hazards

EtBr is a potent mutagen (may cause genetic damage), and moderately toxic after an acute exposure. EtBr 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. It should be stored away from strong oxidizing agents in a cool, dry place, and the container must be kept undamaged and tightly closed.

Safety precautions

Good work practices can help reduce hazardous exposures.

- 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.
- To prevent skin contact when working with liquid solutions, wear protective gloves, a laboratory coat, and chemical goggles. Change gloves frequently.
- Provide EtBr users with safety training on EtBr hazards, use, and proper cleanup procedures. Document training by signing the laboratory's Chemical Hygiene Plan (CHP, yellow flip chart). Include this fact sheet in the CHP as a standard operating procedure (SOP).
- Review an EtBr Material Safety Data Sheet (MSDS) and this EH&S fact sheet before handling the material.
- Wear eye protection and ensure that there is unobstructed access to an eyewash/shower unit in the work area.
- As with any chemical, to avoid ingestion do not eat or drink where EtBr is handled, processed, or stored.
- Always wash hands thoroughly after handling EtBr, even if gloves are used.
- Wear UV-blocking eyewear or work behind a UV shielding glass when using ultraviolet light to visualize EtBr.

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Emergency exposure procedures

Eye care: If EtBr comes in contact with the eyes, immediately flush them with copious amounts of cold or cool water for at least 15 minutes, preferably in an emergency eyewash.

Skin care: In the event of skin exposure, remove contaminated clothing and immediately wash the affected area with soap and copious amounts of cold or cool water for 15 minutes.

If swallowed or inhaled: In the case of EtBr ingestion, obtain medical attention immediately. If EtBr dust is inhaled, move the victim to a source of fresh air.

Note: After any exposure to EtBr (via skin, inhalation, ingestion, or eye contact), the victim should immediately seek a medical evaluation from Tang Center or, if the exposure occurs while Tang Center is closed, from Alta Bates Medical Center (2450 Ashby Avenue, Berkeley).

Spill procedures

If a spill of any size of EtBr enters a sink or floor drain, immediately notify EH&S (642-3073) during normal business hours or UCPD (642-6760) after hours. In the event of a large spill, notify all others in the area that the spill has occurred. Evacuate the room or immediate area and call EH&S (or UCPD, after hours) for assistance with the cleanup. Post signs warning others of the spill, and prevent unnecessary entry into the area until the EH&S response team arrives. Provide any assistance and information you can to the spill responders.

Small spills that do not enter drains can be cleaned up by laboratory personnel who are aware of the hazards of EtBr, have been trained on proper cleanup procedures below, and have access to appropriate safety and cleanup equipment. If you do not have appropriate training or equipment, contact EH&S for assistance.

Decontamination procedure[1]

Always wear protective clothing (as described in "Safety precautions," above) when cleaning up a small spill of EtBr. Use a UV light source to locate the spill; EtBr's fluorescence is easy to see. If the spill is **powder,** carefully wipe it up with wet paper towels and follow the decontamination procedure below. If the EtBr spill is **liquid,** absorb freestanding liquid with dry paper towels. Use UV light to locate any remaining EtBr, then follow the decontamination procedure below.

Wear full protective equipment (lab coat, gloves, and goggles) when preparing or using the decontamination solution. Just prior to using EtBr, prepare a decontamination solution of 4.2 g of sodium nitrite ($NaNO_2$, CAS # 7362-00-0) and 20 ml of hypophosphorous acid (50 percent) (H_3PO_2, CAS # 6303-21-5) in 300 ml of water.

1) 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.

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[1] Lunn, G. & Sansone, E.B., *Destruction of Hazardous Chemicals in the Laboratory*, John Wiley & Sons, New York, 1990, p. 119-120.

2) Using a UV light, check the area to ensure that all the EtBr has been removed. Repeat decontamination procedure as necessary. If the acid could damage the contaminated surface, use a few additional rinses with paper towels soaked in tap water.

3) Soak all the towels in decontamination solution for one hour. Then remove them, gently wring out excess solution back into the decontaminant container, and double-bag along with contaminated gloves and other contaminated items.

4) Using chemical names, clearly label the container of decontamination solution and bag of cleanup materials (e.g., "Laboratory debris contaminated with ethidium bromide"), and contact EH&S or follow the disposal procedures below.

Specific guidance for disposal of various EtBr waste streams (buffer solutions, filters, etc.) is provided in the table below. EtBr solutions destined for drain disposal must first be checked for fluorescence using UV light. If fluorescence is detected, the solution(s) must be treated (or re-treated) prior to drain disposal. Two treatment methods are recommended for dilute solutions of EtBr:

1) **Filtration,** using granular charcoal filtration or an EtBr extractor. Examples include the Schleicher & Schuell Extractor Ethidium Bromide Waste Reduction System and the EMD Biosciences Ethidium Bromide Adsorber, both available through VWR; or the Fluka Ethidium Bromide/SYBR®Green Detoxification Kit or Sigma Extractor for Ethidium Bromide Decontamination, both available through Sigma-Aldrich. It is important not to exceed the extractor's capacity, as described in the product instructions. Once they have been exhausted, the filters or extractors must be discarded through EH&S.

or

2) **Absorption,** using "tea bag" style products specifically designed for EtBr decontamination, such as the Qbiogene EtBr GreenBag™ Disposal Kit or Amresco Destaining Bags. Follow the product instructions, ensuring that enough tea bags are used to absorb the amount of EtBr in solution. Once they have been exhausted, the tea bags must be discarded through EH&S.

Disposal

Your lab's procedures for treating EtBr solutions should be documented and incorporated into the lab's standard operating procedures (SOPs). For complete instructions regarding proper disposal of items through EH&S, please refer to Fact Sheet #52, "Unwanted Hazardous Materials."

DISPOSAL GUIDELINES FOR ETHIDIUM BROMIDE

Waste Stream	Description	Waste Disposal Procedure
Buffer solutions	Typically contain very small concentrations of EtBr (<0.5 mg/L)	EH&S strongly recommends that buffer solutions be run through a filter or treated with tea bags prior to drain disposal. Users are required to verify that buffer solutions do not fluoresce prior to drain disposal.
Stock solutions, cesium chloride solutions	Typically contain higher concentrations of EtBr (1–10 mg/ml)	The high concentration of EtBr in most of these solutions makes filtration/absorption impractical. Dispose of them through EH&S.*
Used filters and tea bags	Contaminated with EtBr from filtration/absorption process	Place items in clear, labeled bags. Dispose of used filters and tea bags through EH&S.
Gels	Typically contain lower concentrations of EtBr (3–5 mg/L)	Allow gels to dry out, then place in clear, labeled bags. Dried gels may be bagged with EtBr-contaminated debris. Dispose of gels through EH&S.
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. Dispose of contaminated sharps and debris through EH&S.
Crystals and powders	Typically pure or concentrated EtBr	Dispose of EtBr crystals and powders through EH&S.

* You may drain-dispose of these solutions yourself only if your treatment method effectively removes all traces of EtBr. (Verify with fluorescence.)

EtBr Alternatives

Consider switching to less-toxic alternatives to EtBr (SYBR Safe™ DNA gel stain, for example) to reduce potential hazardous exposures in the lab. Disposal of most EtBr alternatives must be managed by EH&S, in a manner similar to that described above for EtBr. Contact EH&S for specific guidance if you have questions regarding proper disposal of EtBr alternatives.