

## **Ethidium Bromide Waste Disposal**

Ethidium bromide is commonly used in molecular biology laboratories for DNA identification in electrophoresis. Ethidium bromide has mutagenic properties that may present a hazard if it is poured down the drain untreated or placed in the trash. All ethidium bromide waste must be disposed of as directed in this guide or contact Risk Management & Safety (593-1662) with any questions.

### 1. When Possible Use Safer Alternatives

SYBR Safe DNA gel stain has been developed as a safer alternative to ethidium bromide. Product information is available here: <a href="http://www.invitrogen.com">http://www.invitrogen.com</a>. Read the instructions closely, as SYBR Safe may not be suitable for all applications. MIT conducted an evaluation of SYBR Safe, that information is available here: <a href="http://ehs.mit.edu/site/sites/default/files/files/SYBR.pdf">http://ehs.mit.edu/site/sites/default/files/files/SYBR.pdf</a>, note that the specific MIT programs are not applicable to Ohio University.

# 2. Solutions Containing Ethidium Bromide

Any solution that contains ethidium bromide AND is a hazardous chemical waste (flammable, toxic, reactive, corrosive, etc.) must be treated as hazardous chemical waste. Examples of hazardous chemical wastes include organic solvents or solutions containing heavy metals, cyanides or sulfides. Collect the waste in a sealable bottle; place a red EHS hazardous waste sticker on the bottle when the bottle is started and write the start date on the bottle. When the bottle is full, complete the list of constituents on the label and dispose of the waste through the chemical waste program, see section 6.

Aqueous solutions containing  $<10~\mu g/ml$  ethidium bromide may be poured down the drain to the sanitary sewer; run plenty of water to flush the solution. Each laboratory room may dispose of up to four (4) liters per day. Waste solutions may not be diluted to meet this criterion.

Aqueous solutions containing  $\geq 10~\mu g/ml$  ethidium bromide may be filtered for disposal as outlined below or they may be treated as chemical waste. To have the material treated as chemical waste, collect the liquid in a sealable bottle. When the bottle is full, label the bottle as "Ethidium Bromide Contaminated Liquid" and dispose of it through the chemical waste program, see section 6.

## 3. Filtering of Aqueous Solutions that Do Not Contain Other Hazardous Chemicals

Several options are available to filter aqueous ethidium bromide solutions through activated charcoal. The activated charcoal will collect the ethidium bromide. The remaining liquid can be pour down the drain to the sanitary sewer. After use, the ethidium bromide contaminated charcoal device must be bagged, labeled and disposed of as solid ethidium bromide waste, see section 4. When selecting a filter option, note how much ethidium bromide can be collected by each device and use the device according to the manufacturer's directions.

**Funnel Filter Options** 

Extractor Waste Reduction System	Clontech BondEX Detoxification Cartridge
from VWR, product #28165-500	from BD Biosciences, product #740701
http://www.vwrsp.com	http://www.clontech.com

"Tea Bag" Options

1 th 2 hg   0 p 1 th 1 h	
Green Bag	Destaining Bag
from Qbiogene, product #2350-200	from Amresco Inc., product # E732
http://www.qbiogene.com	http://www.amresco-inc.com/



### 4. Solid Wastes Contaminated with Ethidium Bromide

Electrophoresis Gels: Gels with trace amounts,  $\leq 0.1\%$ , of ethidium bromide may be placed into the regular trash. Wrap the gels in plastic before placing in the trash.

All other solid wastes contaminated with ethidium bromide must be collected and treated as chemical waste; this includes electrophoresis gels with >0.1% ethidium bromide, gloves, centrifuge tubes, towels, etc. The waste may be collected in a plastic container/pail that is lined with a plastic bag or the waste may be collected in plastic zip-locking bags. When the container/pail is full, tie the liner bag shut, or when the plastic bag is full, double bag it into a second zip-locking bag. Label the outside of the container as "Ethidium Bromide Contaminated Solids". The waste must be disposed of through the chemical waste program, see section 6.

Any sharp items, such as needles, syringes, slides, broken glass, etc., that are contaminated with ethidium bromide must be collected in a sharps container. Needles and syringes need to be collected in a puncture proof sharps container, one example is found here: <a href="VWR Sharps Container">VWR Sharps Container</a>. Other sharps may be collected in any hard-walled puncture resistant container. Label the container as "Ethidium Bromide Contaminated Sharps", when the containers are ¾ of the way full dispose of the container through the chemical waste program, see section 6.

5. Materials that are Contaminated with Ethidium Bromide and are also Regulated Infectious Waste or Non-Regulated Biohazardous Waste

See the Biosafety Manual for definitions of regulated infectious and non-regulated biohazardous wastes.

Regulated Infectious Waste: These wastes must be disposed of through the Infectious Waste program. The waste must be collected into the infectious waste boxes; instructions for waste collection are included in the Biosafety Manual. These boxes MUST have the option of "Chemotherapy Waste" checked on the outside of the box and the user must write "Incinerate Only" on the outside of the box. (Autoclaving does not destroy ethidium bromide, so incineration must be specified because university waste is typically autoclaved). For additional information or to join the infectious waste program, contact the Biosafety Officer at 593-1662.

Non-Regulated Biohazardous Wastes: These wastes may be treated as infectious waste according to the instructions above. Biohazardous plant waste that is not infectious to human or animals may be treated as follows. Collect the waste as indicated in the liquid or solid waste sections. When the containers are full label the containers as "Ethidium Bromide Contaminated Solid/Liquid – Plant Biohazard". You may include a biohazard sticker, but you must write "Plant Biohazard". Then the waste may be disposed of through the chemical waste program, see section 6.

# 6. Chemical Waste Disposal

Full instructions for handling chemical waste (<a href="http://www.ohio.edu/ehs/hazmat/chemical.htm">http://www.ohio.edu/ehs/hazmat/chemical.htm</a>) can be found online or by contacting the Hazardous Materials Manager at 593-1663. The basic procedure is as follows.

- 1. Collect and label the waste as described above.
- 2. Fill out the Hazardous Waste Disposal Form (<a href="http://www.ohiou.edu/ehs/docs/chemwaste\_form.xls">http://www.ohiou.edu/ehs/docs/chemwaste\_form.xls</a>), use one line per container, completing as much of the information as you have.
- 3. Email the disposal form to the Hazardous Materials Manager at hazwaste@ohio.edu.
- 4. The Hazardous Materials Manager will pick up the waste from your lab and store it prior to final disposal.