

**APPENDIX B: Acrylamide; Chloroform; Ethidium bromide; Formaldehyde; and Trypan blue stain: UTHSCSA ROUTINE MOLECULAR LABORATORY PROCEDURAL USE  
AND  
EMERGENCY RESPONSE TO EXPOSURE INFORMATION**

**NOTE:** Only use any of these 5 hazardous chemicals after receiving safety training from your Principal Investigator / Supervisor. This training is to be documented on the **Laboratory / Area Hazardous Chemical Training Record - Texas Hazard Communication Act – Specific Training Requirements** form.

Unless otherwise specified, contact Environmental Health & Safety (210-567-2955) for all accidental spills of a hazardous material. **The South Texas Poison Center may be reached at 1-800-222-1222 for medical assessment and treatment recommendations at any time for acute exposures to any potential toxin or toxicant. For any immediate life-threatening exposure 9-1-1 should be called.**

**1. Acrylamide; CAS # 79-06-1:**

- a. **Synonyms:** Acrylic amide; Ethylenecarboxamide; 2-Propenamamide; Propenoic Acid Amide
- b. **Form:** Solid – white crystalline chunks.
- c. **Procedural use:** Used to prepare polyacrylamide gels for electrophoresis of proteins and nucleic acids. The typical expected quantity of use:  $\leq 100$  mls of an 8% solution per month.
- d. **Hazards:** Toxic. IARC Group 2A carcinogen. Acrylamide monomer is highly toxic by inhalation and via skin contact (can penetrate unbroken skin easily); also a suspected carcinogen. The polymer is not generally considered as toxic. Danger of serious damage to health by prolonged exposure through inhalation, contact with skin, or if swallowed. Acrylamide tends to sublime (go directly from solid to vapor form) which may lead to inhalation. Hazardous polymerization may occur from prolonged exposure to heat or light.
- e. **NFPA ratings:** Health = 2; Flammability = 2; Reactivity = 2; Special = TOX
- f. **Risk Control Measures:** Gloves should be used when potential for skin contact exists. Disposable nitrile, neoprene, natural rubber, PVC, or butyl gloves should be worn. Safety splash shield or goggles & mask should be worn, especially when weighing powder or transferring solution. Use of large quantities may warrant the use of a NIOSH approved respirator. A laboratory coat should be worn when working with chemicals.
  - i. **Engineering / Ventilation Controls:** Ensure access to a safety shower and eye wash in areas where acrylamide is used. Use engineering controls such as a chemical fume hood or weighing hood to reduce airborne acrylamide concentrations to as low as reasonably achievable.
- g. **Storage Requirements:** Powder easily becomes airborne and may result in personal exposure and area contamination. Use care to avoid dispersing dust. Keep container tightly closed to prevent acrylamide from subliming and entering the atmosphere. Store in cool place. Keep from contact with oxidizing materials, reducing agents, acids, bases, metal and contaminants. Acrylamide is light sensitive, air sensitive, and hygroscopic.
- h. **Waste Disposal:** RCRA waste # U007. Used gels are normally collected in a dedicated & labeled plastic pail with lid and a disposable plastic liner. Contact the EH&S Environmental Protection Division @ 567-2955 for proper waste disposal information, containers, or pick-up.
- i. **Emergency Response to Exposure**
  - i. **Signs & symptoms of overexposure:** Nervous system effects (neurotoxicity). These include increased sweating of the hands and feet, numbness, tingling, and weakness in the extremities, unsteady gait and decreased reflexes.
    1. **Eyes** will become irritated – overexposure causes neurotoxicity as above.

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2. **Skin** exposure may cause peeling and redness at the area of exposure. Sensitivity more likely to occur after repeat exposures.
3. **Ingestion** – harmful if swallowed – may exhibit neurotoxicity effects.
4. **Inhalation** – Acrylamide can be absorbed through the lungs and will produce the signs of neurotoxicity.
5. **Chronic** – Prolonged or repeated skin contact may cause dermatitis. May cause cancer or adverse reproductive effects according to animal studies. Prolonged exposure may cause nervous system disturbances.

**ii. First Aid and Treatment**

1. **Eyes** – In case of contact immediately flush eyes with plenty of water for at least 15 minutes. Get medical aid.
2. **Skin** – In case of contact immediately flush skin with plenty of water for at least 15 minutes while removing contaminated clothing and shoes. Get medical aid immediately. Wash contaminated clothing prior to reuse.
3. **Ingestion** – If swallowed, DO NOT INDUCE VOMITING unless directed to do so by medical personnel. Never give anything by mouth to an unconscious person. Get medical aid.
4. **Inhalation** – if inhaled, remove to fresh air. If not breathing, give artificial respiration using one way valve. If breathing is difficult, give oxygen. Get medical aid.
5. **Note** – Either acute or chronic exposure may lead to weak or absent reflexes. An early sign of toxic effects is peeling of the skin at the fingertips.
6. **Known Antidote** – Pyridoxine (vitamin B6), pyruvate, and N-acetylcysteine have been used to reduce the toxicity of acrylamide in experimental studies, but are unproven.

**2. Chloroform; CAS # 67-66-3**

- a. **Synonyms:** Formyl trichloride; Methane trichloride; Trichloroform; Trichloromethane; R-20 Refrigerant; Methyl Trichloride
- b. **Form:** Liquid
- c. **Procedural use:** Used as a solvent for organic compounds in the molecular extraction process. The typical expected quantity of use:  $\leq 100$  mls per month.
- d. **Hazards:** Animal carcinogen / possible human carcinogen. IARC Group 2B carcinogen. Do not breathe vapors and harmful if swallowed. Do not get in eyes, on skin, or clothing - irritating to the eyes and skin with possible risk of irreversible effects. Danger of serious damage to health by prolonged exposure through inhalation and if swallowed. Emits toxic fumes under fire conditions. Target organs include cardiovascular system, CNS, blood, liver, and kidneys. Hazardous polymerization will not occur.
- e. **NFPA ratings:** Health = 2; Flammability = 0; Reactivity = 0; Special = CA
- f. **Risk Control Measures:** Viton or heavy duty butyl rubber gloves should be used when potential for skin contact exists. Nitrile, neoprene and natural rubber exhibit poor resistance to chemical breakthrough. Safety splash shield or goggles should be used when splash potential exists. Use only in an approved chemical fume hood or ducted biological safety cabinet with adequate exhaust. Use of large quantities may warrant the use of a NIOSH approved respirator and protective apron. A laboratory coat should be worn when working with chemicals.
  - i. **Engineering / Ventilation Controls:** Ensure access to a safety shower and eye wash in areas where chloroform is used. Use engineering controls such as a

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chemical fume hood or weighing hood to reduce airborne chloroform concentrations to as low as reasonably achievable. . OSHA PEL = 50 ppm.

- g. Storage Requirements:** Keep container tightly closed to prevent chloroform from vaporizing to the atmosphere. Store in cool, dry, place. Keep from contact with strong oxidizing agents.
  - h. Waste Disposal:** RCRA waste # U044. Used chloroform is collected in a dedicated & labeled amber/brown glass or polycarbonate bottle with secondary containment – chloroform is sensitive to light and may decompose. Contact the EH&S Environmental Protection Division @ 567-2955 for proper waste disposal information, containers, or pick-up.
  - i. Emergency Response to Exposure**
    - i. Signs & symptoms of overexposure:** Irritating to the eyes, respiratory system and skin. May cause central nervous system effects (neurotoxicity). These include increased sweating of the hands and feet, numbness, tingling, and weakness in the extremities, unsteady gait and decreased reflexes. . May cause vomiting and gastrointestinal disturbances. Alcohol consumption may increase toxic effects.
      - 1. **Eyes** will become irritated.
      - 2. **Skin** exposure may cause irritation and redness at the area of exposure. . May be harmful if absorbed through the skin.
      - 3. **Ingestion** – Harmful if swallowed – may exhibit neurotoxicity effects.
      - 4. **Inhalation** – Chloroform may be irritating to mucous membranes and upper respiratory tract, can be absorbed through the lungs, and may produce the signs of neurotoxicity.
      - 5. **Chronic** – Prolonged or repeated skin contact may cause dermatitis. May cause cancer or adverse reproductive effects according to animal studies. Prolonged exposure may cause nervous system disturbances, and damage to other listed target organs.
    - ii. First Aid and Treatment**
      - 1. **Eyes** – In case of contact immediately flush eyes with plenty of water for at least 15 minutes. Get medical aid.
      - 2. **Skin** – In case of contact, immediately wash skin with plenty of soap and water. Remove contaminated clothing and shoes. Get medical aid immediately. Wash contaminated clothing prior to reuse.
      - 3. **Ingestion** – If swallowed, wash out mouth with water provided person is conscious - never give anything by mouth to an unconscious person. Get medical aid.
      - 4. **Inhalation** – If inhaled, remove to fresh air. If not breathing, give artificial respiration using one way valve. If breathing is difficult, get medical aid.
      - 5. **Known Antidote** – None listed.
- 3. Ethidium bromide; CAS # 1239-45-8**
- a. Synonyms:** 2,7 – Diamino-10-ethyl-9-phenylphenanthridinium bromide; 3, 8, - Diamino-5-ethyl-6- phenylphenanthridinium bromide; Dromilac; Homidium bromide
  - b. Form:** Dark red solid tablet
  - c. Procedural use:** Used to visualize (stain) nucleic acids in agarose gels in the molecular biology laboratory. The typical expected quantity of use: ≤ 2 milligrams per month.
  - d. Hazards:** Highly Toxic and potent mutagen. Ethidium bromide is highly toxic by inhalation and via skin contact (can penetrate unbroken skin easily). Irritating to the eyes, respiratory

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tract, and skin. Danger of serious damage to health by prolonged exposure through inhalation, contact with skin, or if swallowed. Do not breathe in dust – do not get in eyes, on skin, or on clothing. Hazardous polymerization will not occur.

- e. **NFPA ratings:** Health = 2; Flammability = 1; Reactivity = 0; Special = TOX
- f. **Risk Control Measures:** Gloves must be used when potential for skin contact exists. Disposable nitrile, neoprene, natural rubber, PVC, or butyl gloves should be worn. Safety splash shield or goggles & mask should be worn, especially when weighing powder or transferring solution. A laboratory coat should be worn when working with chemicals.
  - i. **Engineering / Ventilation Controls:** Ensure access to a safety shower and eye wash in areas where ethidium bromide is used. Use engineering controls such as an approved chemical fume hood and weighing hood to reduce airborne ethidium bromide concentrations to as low as reasonably achievable.
  - ii. **Spills:**
    - 1. **Small spills** (<25 ml) of ethidium bromide in solution: use UV light to locate spill, and neutralize (deactivate) with paper towel soaked in decontaminating solution consisting of 4.2 g of sodium nitrite dissolved in 20 ml of hypophosphorous acid and 300 ml water. Then wash area 5 times with a new wet paper towel each time, and soak all the towels in remaining decon solution for 1 hour. Confirm deactivation with UV light. Contact EH&S to pick up materials for disposal.
    - 2. **Large spills** - Notify others in area and evacuate area immediately. Secure area and notify EH&S personnel (7-2955) immediately.
- g. **Storage Requirements:** Use care to avoid dispersing dust. Keep container tightly closed when not in use. Store in cool, dry place. Keep from contact with oxidizing materials, reducing agents, acids, and bases - ethidium bromide is incompatible with strong oxidizing agents.
- h. **Waste Disposal:** RCRA – Not a regulated waste, however the mutagenic properties of this chemical may present a hazard if it is poured down the drain, or put in the regular trash. Concentrations of < 0.1 % of ethidium bromide in solution may be disposed of down the drain or, if mixed with biologic agents, in the regulated medical waste. Ethidium bromide in used gels is normally collected in a dedicated & labeled plastic pail with lid and a disposable plastic liner. Charcoal filtration methods are also available for aqueous solutions  $\geq 0.1\%$  of ethidium bromide. Contact the EH&S Environmental Protection Division @ 567-2955 for proper waste disposal information, containers, or pick-up.
- i. **Emergency Response to Exposure**
  - i. **Signs & symptoms of overexposure:** Nervous system effects (neurotoxicity). These include increased sweating of the hands and feet, numbness, tingling, and weakness in the extremities, unsteady gait and decreased reflexes.
    - 1. **Eyes** will become irritated – overexposure causes neurotoxicity as above.
    - 2. **Skin** exposure may cause redness at the area of exposure.
    - 3. **Ingestion** – harmful if swallowed – may exhibit neurotoxicity effects.
    - 4. **Inhalation** – Ethidium bromide can be absorbed through the lungs and will produce the signs of neurotoxicity.
    - 5. **Chronic** – Prolonged or repeated skin contact may cause dermatitis. May cause cancer or adverse reproductive effects. Prolonged exposure may cause nervous system disturbances.
  - ii. **First Aid and Treatment**
    - 1. **Eyes** – In case of contact immediately flush eyes with plenty of water for at least 15 minutes. Get medical aid.

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2. **Skin** – In case of contact immediately wash skin with plenty of soapsuds, and rinse with water while removing contaminated clothing and shoes. Get medical aid immediately. Wash contaminated clothing prior to reuse.
3. **Ingestion** – If swallowed, seek medical attention immediately. Contact South Texas Poison Center at 1-800-222-1222.
4. **Inhalation** – if inhaled, remove to fresh air. If not breathing, give artificial respiration using one way valve. If breathing is difficult, give oxygen. Get medical aid.
5. **Known Antidote** – No known antidote.

**4. Formaldehyde; CAS # 50-00-0**

- a. **Synonyms:** Formaldehyde – 37% in 12-15% methanol, Fannoform, Formalin, Formalin-40, Formalith, Formic aldehyde, Formol, Fyde, Lysoform, Methaldehyde, Methanal, Methyl aldehyde, methylene oxide.
- b. **Form:** Liquid, solution with 37% formaldehyde; 49.5% water; 13.5% methanol. Clear colorless, mobile liquid with strong, pungent characteristic odor.
- c. **Procedural use:** In the molecular laboratory, used as an ingredient in the staining solution for polyacrylamide gels. The typical expected quantity of use:  $\leq 10$  mls per month.
- d. **Hazards:** NTP, IARC, and OSHA regulated human carcinogen. May cause heritable genetic damage. Acute exposure to formaldehyde will cause severe eye burns, skin irritation, and respiratory irritation. Material is extremely destructive to the tissue of the mucous membranes and upper respiratory tract. Readily absorbed through the skin. Chronic exposure can cause blindness, skin hardening, tanning, and sensitization. Avoid contact with caustic soda, soda ash, other alkalis, sodium metal, potassium metal, and other alkali metals. Also avoid contact with amines, acids, oxygen, and hydrogen peroxide. Hazardous polymerization will not occur. Vapors are heavier than air.
- e. **NFPA ratings:** Health = 2; Flammability = 1; Reactivity = 0; Special = CA
- f. **Risk Control Measures:** Gloves should be used when potential for skin contact exists. Nitrile, Viton, or butyl rubber gloves should be worn. Neoprene or natural latex rubber gloves should not be worn as they rapidly degrade. Safety splash shield or goggles should be worn when splash hazard exists. Use of large quantities may warrant the use of a protective apron and NIOSH approved respirator. A laboratory coat should be worn when working with chemicals.
  - i. **Engineering / Ventilation Controls:** Ensure access to a safety shower and eyewash in areas where formaldehyde is used. Use engineering controls such as a chemical fume hood or weighing hood to reduce airborne formaldehyde concentrations to as low as reasonably achievable and below the OSHA PEL. OSHA TWA = 0.75 ppm. OSHA STEL = 2.0 ppm.
  - ii. **Spills:**
    1. **Small spills** of less than or equal to 25 mls can be neutralized with ammonium hydroxide or sodium sulfite.
    2. **Large spills** > 25 mls - Notify others in area and evacuate area immediately. Secure area and notify EH&S personnel (7-2955) immediately.
- g. **Storage Requirements:** Store in cool, dry, well-ventilated place. Keep from contact with oxidizing materials, reducing agents, acids, bases, metal and contaminants.
- h. **Waste Disposal:** RCRA waste # U122. Collect used formaldehyde in a dedicated & labeled plastic container with secondary containment. Keep capped when not transferring waste. Other neutralizing agents are available. Contact the EH&S Environmental

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Protection Division @ 567-2955 for proper waste disposal information, containers, or pick-up.

**i. Emergency Response to Exposure**

**i. Signs & symptoms of overexposure:** Most immediate effect will be obvious irritation of the eyes and respiratory tract.

1. **Eyes** will become very irritated with excessive tearing.
2. **Skin** exposure may cause redness or burns at the area of exposure. Sensitivity more likely to occur after repeat exposures.
3. **Ingestion** – harmful if swallowed – causes severe irritation and inflammation of the mouth, throat and stomach. Severe abdominal pains leading to unconsciousness.
4. **Inhalation** – Formaldehyde is highly irritating to the respiratory tract – causes inflammation of the nose and throat. May cause coughing, wheezing, laryngitis, shortness of breath, headache, nausea and vomiting. Inhalation into the lungs may result in chemical pneumonitis or pulmonary edema.
5. **Chronic** – Prolonged or repeated skin contact may cause dermatitis and sensitivity. May cause cancer or adverse reproductive effects. Prolonged exposure by inhalation may cause increased sensitivity.

**ii. First Aid and Treatment**

1. **Eyes** – In case of contact immediately flush eyes with plenty of water for at least 15 minutes. Get medical aid.
2. **Skin** – In case of contact immediately flush skin with plenty of water for at least 15 minutes while removing contaminated clothing and shoes. Contact a physician. Wash contaminated clothing prior to reuse.
3. **Ingestion** – If swallowed, wash out mouth with water and induce vomiting provided person is conscious - never give anything by mouth to an unconscious person. Get medical aid.
4. **Inhalation** – if inhaled, remove to fresh air. If not breathing, give artificial respiration using one way valve. If breathing is difficult, give oxygen. Get medical aid.
5. **Known Antidote** – None listed.

**5. Trypan blue stain; CAS # 72-57-1**

- a. Synonyms:** C.I. 23850, Benzamine blue, Direct blue 14, Niagra blue 3B
- b. Form:** Dark blue, or purple-brown to black powder (soluble in water)
- c. Procedural use:** Used in the molecular laboratory to stain cultured cells for microscopic examination. The typical expected quantity of use is about 500 microliters of a 0.1% solution per month.
- d. Hazards:** OSHA possible select carcinogen. IARC Group 2B carcinogen – may cause cancer based on animal data and has teratogenic effects on laboratory animals. Trypan blue may cause irritation to the skin, eyes, and respiratory tract. May be harmful if swallowed or inhaled. Avoid dust generation, excess heat, and strong oxidants. Hazardous polymerization does not occur.
- e. NFPA ratings:** Health = 1; Flammability = 0; Reactivity = 0; Special = CA
- f. Risk Control Measures:** Gloves should be used when potential for skin contact exists. Disposable latex rubber, nitrile, neoprene, PVC, or butyl gloves should be worn. Safety splash shield or goggles & mask should be worn, especially when weighing powder or

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transferring solution. Use of large quantities may warrant the use of a NIOSH approved respirator. A laboratory coat should be worn when working with chemicals.

- i. Engineering / Ventilation Controls:** Ensure access to a safety shower and eyewash in areas where Trypan blue is used. Use engineering controls such as a chemical fume hood or weighing hood to reduce airborne Trypan blue concentrations to as low as reasonably achievable.
- g. Storage Requirements:** Powder easily becomes airborne and may result in personal exposure and area contamination. Use care to avoid dispersing dust. Keep container tightly closed to prevent Trypan blue from entering the atmosphere. Store in cool, dry, ventilated area. Keep from contact with oxidizing materials, reducing agents, acids, bases, metal and other contaminants.
- h. Waste Disposal:** RCRA – Not a regulated waste, however the carcinogenic properties of this chemical may present a hazard if it is poured down the drain, or put in the regular trash. Concentrations of < 0.1 % of Trypan blue in solution may be disposed of down the drain or, if mixed with biologic agents, in the regulated medical waste. Containers of this material may be hazardous when empty since they retain product residues – dusts and solids. Contact the EH&S Environmental Protection Division @ 567-2955 for proper waste disposal information, containers, or pick-up.
- i. Emergency Response to Exposure**
  - i. Signs & symptoms of overexposure:** These include general irritation of the eyes, skin, or respiratory tract.
    1. **Eyes** will become irritated.
    2. **Skin** exposure may cause redness and irritation at the area of exposure.
    3. **Ingestion** – harmful if swallowed – may cause gastrointestinal irritation with nausea, vomiting, and diarrhea.
    4. **Inhalation** – Trypan blue may cause respiratory tract irritation. The toxicological properties of this chemical have not been fully investigated.
    5. **Chronic** – Prolonged or repeated exposure may cause cancer according to animal studies.
  - ii. First Aid and Treatment**
    1. **Eyes** – In case of contact immediately flush eyes with plenty of water for at least 15 minutes. Get medical aid.
    2. **Skin** – In case of contact immediately wash skin with plenty of soap and water for at least 15 minutes while removing contaminated clothing and shoes. Wash contaminated clothing prior to reuse.
    3. **Ingestion** – If swallowed, DO NOT INDUCE VOMITING unless directed to do so by medical personnel. Never give anything by mouth to an unconscious person. If conscious and alert, rinse mouth and drink 2-4 cupfuls of milk or water. Get medical aid.
    4. **Inhalation** – If inhaled, remove from exposure to fresh air immediately. If not breathing, give artificial respiration. If breathing is difficult, give oxygen. Get medical aid.
    5. **Known Antidote** – None listed.