

AUTOCLAVE SAFETY GUIDELINES

An autoclave is a commonly used piece of equipment in biomedical laboratories. Autoclaves pose many hazards including physical hazards (e.g. heat, steam and pressure) and biological hazards. These guidelines are governed by **ADEM: Alabama Department of Environmental Management “Solid Waste Disposal Act” 335-13-7-08, infectious waste treatment regulations, CDC: Centers for Disease Control-Biosafety in Microbiological and Biomedical Laboratories (BMBL) and UA’s Biological Safety Manual**. These guidelines are intended to provide practical information that can be utilized by all researchers to safely operate the autoclaves at UA. Individual labs are encouraged to use this document as a guide for training new personnel on the safe use of autoclaves.

Controls for different brands of autoclaves may have unique characteristics for loading, load sizes, and cycle types and settings. The type of materials you sterilize will determine the type of sterilization cycle you use. For this reason, it is important to review and understand the owner’s manual before using any autoclave for the first time. Always ensure the owner’s manual is readily available in case questions or concerns arise during operation.

1. DEFINITIONS:

AUTOCLAVE: a device designed to sterilize equipment or biological waste by means of heat, and pressure within a chamber.

BIOHAZARDOUS/MEDICAL AGENT: a pathogen that is capable of replication and is a disease causing microorganism capable of causing diseases in humans, animals, or plants. Examples included viruses, microbes and sub-viral particles. The term agent refers to the agent itself, products of infectious agents and the components of infectious agents that present a risk of illness or injury.

BIOHAZARDOUS/MEDICAL MATERIAL: any material that harbors biohazardous agents including human or animal blood, body fluids, or tissues that may be contaminated with biohazardous agents.

BLOOD: human blood, human blood components, and products made from human blood or animal blood infected with human pathogens.

PERSONAL PROTECTIVE EQUIPMENT: specialized clothing or equipment worn by an employee for protection against a hazard. General work clothes (e.g., uniforms, pants, shirts, or blouses) not intended to function as protection against a hazard are not considered to be personal protective equipment.

REGULATED WASTE: liquid or semi-liquid blood or *other potentially infectious materials* (OPIM) see definition; contaminated items that would release blood or other potentially infectious materials in a liquid or semi-liquid state if compressed; items that are caked with dried blood or other potentially infectious materials and are capable of releasing these materials during handling; contaminated sharps; and pathological and microbiological wastes containing blood or other potentially infectious materials.

STERILIZE: the use of a physical or chemical procedure to destroy all microbial life including highly resistant bacterial endospores.

Other Potentially Infectious Materials (OPIM)

1. The following human body fluids:

- Semen
- Vaginal secretions
- Cerebrospinal fluid (fluid surrounding the brain and spinal cord)
- Synovial fluid (fluid surrounding bone joints)
- Pleural fluid
- Pericardial fluid
- Peritoneal fluid
- Amniotic fluid
- Saliva in dental procedures
- Any body fluid that is visibly contaminated with blood
- All body fluids in situations where it is difficult or impossible to differentiate between body fluids

2. Any unfixed tissue or organ (other than intact skin) from a human, or non-human primate (living or dead).

3. HIV-containing cell or tissue cultures, organ cultures, and HIV or HBV-containing culture medium or other solutions, and blood, organs or other tissues from experimental animals infected with HIV or HBV.

4. Any pathogenic microorganism

5. Human cell lines

2. RESPONSIBILITIES:

Institutional Biosafety Committee (IBC)

The IBC is responsible for advising the University on all matters related to biosafety. With regard to medical waste disposal, the IBC describes University waste disposal procedures on the EH&S website.

<http://ehs.ua.edu/compliance/biological-safety/>

EH&S is responsible for the following:

- Preparing and updating, in a timely manner, biosafety policies and guidelines with regard to medical waste.
- Providing biosafety and medical waste disposal information as needed.
- Inspecting Departmental autoclave facilities on a routine basis to ensure compliance with the waste disposal regulations and campus waste disposal policies.

Principal Investigator

The Principal Investigator is responsible for the following:

- Segregating all medical waste at the point of generation from other laboratory waste
- Training and supervising laboratory staff on proper autoclave operations.
- Providing personal protective equipment, such as gloves and protective aprons to laboratory staff members who must operate the autoclave
- Ensuring that all laboratory staff members who must operate the autoclave are familiar with emergency procedures prior to operating the autoclave.

General Autoclave Safety Practices:

1. Before using the autoclave, check inside the autoclave for any items left by the previous user that could pose a hazard (e.g. sharps).
2. Clean the drain strainer before loading the autoclave.
3. Load the autoclave properly as per the manufacturer's recommendations.
4. Individual glassware pieces should be within a heat resistant plastic tray on a shelf or rack and never placed directly on the autoclave bottom or floor.
5. Make sure the door of the autoclave is fully closed (latched) and the correct cycle has been selected before starting the cycle.
6. When the cycle is complete, open the door slowly. Keep your head, face, and hands away from the opening.
7. Wear heat-resistant gloves when opening the autoclave door after a cycle. If there is a sharps hazard (e.g. biological waste), wear heat AND cut resistant gloves.
8. At a minimum, when removing items from an autoclave, a rubber apron, rubber sleeve protectors and heat-resistant gloves should be worn.
9. **Do not autoclave items containing corrosives, solvents or volatiles or radioactive materials.**

Additional Practices for Autoclaving Liquids:

1. When running an autoclave cycle with liquids, the cycle time is longer, but uses lower temperatures to minimize evaporation of the liquids being sterilized.
2. **To prevent bottles from shattering during pressurization, the caps of containers with liquids must be loosened before loading.**
3. Use only borosilicate glass (Pyrex™ or Kimax™) which can withstand the high autoclave temperature.
4. Use a tray with a solid bottom and walls to contain the contents and catch spills.
5. Before removing autoclaved items, wait 10 minutes for autoclaved liquid loads.
6. Let liquids stand for a full hour before touching with ungloved hands. Be sure others in the area know a heat hazard is present.

Additional Practices for Autoclaving Dry Loads:

1. Add 1/4 to 1/2 inch of water to the tray so the bottles will heat evenly.
2. Check plastic materials to ensure they are compatible with the autoclave.
3. Before removing autoclaved items, wait 5 minutes for loads containing only dry glassware.
4. For dry loads, let the glassware cool for 15 minutes before touching it with ungloved hands.

Autoclave Monitoring & Maintenance:

Autoclave monitoring and maintenance is an important aspect of a properly functioning autoclave. Follow the manufacturer's recommendations for preventative maintenance and ensure all contractors are approved by the manufacturer. Autoclave operators should ensure that each autoclave is monitored as follows:

Heat Sensitive Tape Monitoring – Operators should use heat-sensitive sterilization indicator tape for **each load** to indicate the load has undergone an effective steam sterilization process. This tape only indicates that the proper temperature has been reached, but does not indicate it was heated for the proper length of time. Please see the flow chart below for the segregation of waste.

Biohazardous Waste Segregation

All biohazardous waste must be treated (ie. decontaminated/sterilized) at some point before disposal. Not all biohazardous waste can be treated at UA, and must be done by a company specializing in transporting, treating, and disposing of this waste stream. It is therefore critical to know which biomedical waste streams can be treated on campus, and which biohazardous waste streams must be segregated and treated elsewhere. Below is a flowchart indicating the types of waste generated at UA, and the relevant procedures to follow.

Storage and labeling

Biohazardous waste must not be stored for long durations of time and it must be labeled as a biohazard.

This is important so that others, custodial staff in particular, are able to clearly distinguish it from other types of waste in the lab.

TREATABLE BIOMEDICAL WASTE (on-site)	NON-TREATABLE BIOMEDICAL WASTE (on-site)
human/animal cultures	Human blood waste
stocks/specimens of microorganisms (BSL1, BSL2)	Animal anatomical waste
human diagnostic specimens (other than urine and feces)	Sharps waste
disposable laboratory material in contact with any of the above	Cytotoxic waste
	Stocks of Organisms BSL2 & Higher



On-site treatment: Microbiological Waste

Microbiological waste is the only biomedical waste stream that can be treated at the university before disposal. Liquid waste is typically treated with bleach in the lab before disposal. Solid waste is steam sterilized by any autoclave undergoing biological monitoring.

NEVER put bleach-treated waste into an autoclave. It is also unnecessary to bleach waste that has been autoclaved.

If disposable laboratory material does not come into contact with any other microbiological waste described previously or any other hazardous materials, it is not hazardous waste. This can be disposed directly into a regular garbage bin within the lab. If uncertain, check with your supervisor before disposal.



On-site Non-Treatable: Biomedical Waste

Biomedical waste that cannot be treated will be segregated and held in a designated biomedical waste storage area for removal by campus EHS. Each lab is responsible for the waste they generate until it has been collected.

All biomedical waste must therefore be labeled with name of the principal investigator (PI), lab location (building and room number), and lab extension.

Recordkeeping:

Operators should maintain documentation of any autoclave preventative maintenance or repairs. These records should indicate who performed the work, the type of maintenance or repairs conducted, and the date the autoclave was serviced. The records should be maintained either in the room with the autoclave, or signage should be posted indicating the location of any records that document autoclave maintenance or repairs.

- 1) Record all data from any run in the Daily Autoclave log : date, run-time, temperature, time-in of treatment; the type of load (clean material or waste); quantity of waste treated; printed name and signature of the person responsible for treatment and any relevant information when applicable;
- 2) The person in charge of the autoclave will be responsible to maintain all records and logs;
- 3) Results of Biological testing results (growth/no growth), should be kept in the log. when appropriated;
- 4) **All data collected in the log must be maintained for three (3) years.**
- 5) The autoclave operator (or person in charge) should notify their supervisor and record any incident or problems when working or monitoring the autoclave.

Training:

Each laboratory must develop and implement an autoclave safety training program. All users must be trained before operating an autoclave and the laboratory PI/supervisor is responsible for ensuring each person in the lab is appropriately trained. All training must be documented and records should be maintained in the lab with your other safety training certificates. The laboratory PI/supervisor is encouraged to use this policy as a guide for training new personnel.

Autoclave Failure:

Discontinue use immediately if an autoclave is not working properly. Post a sign alerting others not to use the autoclave. Mechanical failure needs to be attended by a trained technician. Contact the service company responsible for maintenance of your autoclave or your department's safety representative for further guidance.

References:

- 1) Environmental Health and Safety web site
<http://ehs.ua.edu/compliance/biological-safety/>
- 2) CDC Biological and Infectious Waste
<http://www.cdc.gov/nceh/ehs/etp/biological.htm>
- 3) Biosafety in Microbiological and Biomedical Laboratories (BMBL) 5th Edition , Dec 2009
<http://www.cdc.gov/biosafety/publications/bmb15/BMBL.pdf>