

Principal Investigator: \_\_\_\_\_

Date Approved: \_\_\_\_\_

## Autoclaves

Autoclaves use high pressure and high temperature steam to effectively kill microorganisms or inactivate biohazardous material. Steam must be in contact with the material being sterilized in order for the process to be effective. The potential risks of using an autoclave are heat and steam burns, hot fluid scalds, injuries to hands and arms from the door, or bodily injury in the event of an explosion. If a load of biohazardous waste is inadequately autoclaved, there is potential for human and environmental exposure to biohazardous materials.



Personal Protective Equipment			
 <b>Lab Coat</b>	 <b>Gloves</b>	 <b>Eye Protection</b>	 <b>Closed-Toe Shoes</b>
Traditional lab coat.	Heat-resistant gloves with arm sleeves.	ANSI Z87.1-compliant safety glasses or safety goggles.	Shoes that full cover the foot and ankle.

## Compatible and Incompatible Materials

When autoclaving compatible materials, do not exceed the temperature for which they are rated. Melted plastic releases fumes, ruins autoclave surfaces, and can clog chamber drains.

Compatible	Incompatible
<ul style="list-style-type: none"> <li>● Tissue culture flasks</li> <li>● Surgical instruments</li> <li>● Glassware</li> <li>● Pipette tips</li> <li>● Media solutions</li> <li>● Animal food and bedding</li> <li>● Polypropylene</li> <li>● Stainless steel</li> </ul>	<ul style="list-style-type: none"> <li>● Flammable liquids and solvents</li> <li>● Corrosives, including bleach</li> <li>● Radioactive materials</li> <li>● Sharps</li> <li>● Anatomical pathological waste or animal carcasses/parts</li> <li>● Heat labile materials</li> <li>● Acids, bases, organic solvents</li> <li>● Polystyrene, polyethylene</li> <li>● Liquids in containers not designed for sterilization</li> </ul>

## Procedures for Transporting Materials to Autoclave Rooms

### *Contaminated Materials*

All contaminated materials must be transported utilizing secondary containment, which must have a biohazard label affixed. Autoclaved bags shall be loosely secured and placed in a lipped bin large enough to contain the bag(s). Containers with liquids (such as liquid media) shall be placed in secondary containment to prevent leakage. The container or bin must be carefully transported in a cart or rolling bin which has easily cleanable surfaces.

Biohazardous materials shall be transported by direct routes that avoid high-traffic and clean/sterile storage areas. Extra caution shall be exercised in areas which disrupt smooth transport (such as elevator entrances, ramps, etc.) to prevent accidental release of contaminated materials.

Bins or carts used to transport soiled materials shall be cleaned after each use, unless said bins or carts are dedicated and labeled for biohazardous material transport. If labeled and exclusively used for biohazardous materials, they shall be cleaned when visibly soiled.

### *Freshly Autoclaved Materials*

Materials being removed from the autoclave should be allowed to cool to room temperature before transporting. Never transport superheated materials.

## Appropriate Containers

Individual items should be placed into secondary containment in the form of heat-resistant plastic trays, not directly on the chamber bottom or floor. The bins must be polypropylene (PP) or polypropylene copolymer (PPCO). DO NOT use polyethylene (HDPE, LDPE, PET, PETG) plastic bins. The bottom of the bin will typically have the abbreviation listed. If secondary containers must be removed from the autoclave rooms, return them promptly.

Use only borosilicate glass (Pyrex or Kimax) for liquids, as these can withstand high autoclave temperatures.

## Packaging Autoclave Loads

- Use heat-sensitive indicator tape to help identify processed items. Note that the tape does not guarantee that an autoclave cycle was successful, just that an attempt was made.
- Always use a secondary container to hold loads.
- Add 100 ml of water to waste bags and to secondary containers to ensure adequate steam generation.
- Wear dry, heat-resistant gloves, a fully buttoned lab coat to cover exposed skin, and closed-toe shoes to load and unload the autoclave.
- Check the sediment screen before loading an autoclave run; contact your department representative or EH&S if you notice any debris or clogging.
- Load/stack items in the autoclave to allow efficient steam circulation; do not overcrowd or allow items to touch the sides or the top of the chamber.
- Autoclave clean items and waste separately.

## Autoclaving Liquids

When running an autoclave cycle with liquids, the cycle time is longer but uses lower temperatures to minimize evaporation of the liquids. Liquid cycles also have a longer depressurization time to avoid “boil-over” of liquids.

- To prevent bottles from shattering during pressurization and heating, the caps of containers with liquids must be loosened before loading.
- Use a heat-resistant autoclave tray with a solid bottom and walls to contain the contents and catch spills.
- Bottles of liquid should not be more than  $\frac{2}{3}$  (two-thirds) full. In some autoclaves, there is a temperature sensor device that can be inserted into a full beaker and capped with a rubber stopper.
- Wear gloves when unloading anything from the autoclave. Let the items completely cool before touching with ungloved hands. Be sure to let others in the area know that heat hazard is present.

## Autoclave Cycles

Cycle	Parameters	
	Temperature	Time
Hard Goods	121.1 °C	30 minutes
Wrapped Goods	121.1 °C	30 minutes
Waste	121.1 °C	30 - 45 minutes*
Gravity	121.1 °C	30 minutes

\*30 minutes for ASSC/OMB; 45 minutes for SRC

## Autoclave Logging and Failure

- Log waste autoclave runs on the log sheet next to the autoclave.
- Enter the date of the run, the PI name, operator name, type of waste, and duration of the cycle on the log.
- Discontinue use immediately if an autoclave is not working properly. Post a sign alerting others not to use the autoclave until the issue is resolved.
- Mechanical failures need to be attended to by a trained technician. Contact your PI or departmental equipment contact for further assistance. Additionally, contact EH&S to inform them of the autoclave failure.

## Unloading the Autoclave

- Open the door slowly. Stand to the side and keep your head, face, and hands away from the opening.

- Allow steam to dissipate and materials to cool with the door open prior to unloading the autoclave. Removing contents too soon may cause heat stress and fracturing of materials, especially glass.
- Use a cart and secondary container to transport items.

### **Disposing of Autoclaved Waste**

- After the waste has been autoclaved and cooled, place a completed Autoclave Waste Label on the bag.
- Place the autoclaved bag into an opaque (black) trash bag.
- Placed the bagged waste into an approved location/dumpster.

### **Training**

The PI/Supervisor for each laboratory shall ensure personnel are provided hands-on operation and safety training before operating an autoclave. Training should cover information in this SOP and components on autoclave safety, including (but not limited to) appropriate heat-resistant personal protective equipment, proper body placement to prevent steam burns, and items that can and cannot be autoclaved.

### **First Aid & Emergencies**

Report any incidents to the PI/Supervisor and EH&S.

#### ***Skin or Eye Contact***

If any tissues appear to have been burned due to steam exposure, seek medical attention immediately. Run the affected area under cool water for at least 15 minutes. Apply a dry, sterile bandage. Do not rub the affected area.

