EHS
ENVIRONMENTAL
HEALTH AND SAFETY

Autoclave Use Guidelines

Autoclaves provide an effective and economical way of sterilizing and decontaminating items but can pose a safety hazard if operated incorrectly. These guidelines were developed to help prevent injuries and to ensure that biological wastes are effectively decontaminated prior to disposal in accordance with the Ministry of the Environment Guideline C-17: *Non-Incineration Technologies for Treatment of Biomedical Waste (Procedures for Microbiological Testing).* All waste contaminated with biohazardous material (including RG1 organisms), must be decontaminated prior to disposal.

1.0 MATERIAL COMPATIBILITY

Not all materials are compatible with autoclaving. Toxic fumes and/or vapours can be produced if incompatible materials are autoclaved. Users must recognize acceptable material and packaging.

Items that must never be autoclaved include oils, waxes, some plastics, flammables, corrosives, radioactive materials, and chlorinated substances.

2.0 PERSONAL PROTECTIVE EQUIPMENT (PPE) REQUIREMENTS

Appropriate personal protective equipment must be worn while using an autoclave. Closed-toed shoes, lab coat, eye protection (goggles, face shield), heat resistant long-cuff gloves, rubber apron, rubber sleeve protectors may be required based on a risk assessment. That is, depending on the inherent risk, PPE should be tailored to the task. For example, if there is a splash risk, goggles and/or a face shield should be worn as appropriate.

3.0 FACTORS TO CONSIDER FOR EFFECTIVE DECONTAMINATION/STERILIZATION

Steam treatment is mainly a function of temperature, pressure and time.

- Temperature: Effective sterilization occurs when the steam temperature exceeds 121°C.
- Pressure: Autoclave pressurization should be at least 15 pounds per square inch (psi).
- Time: The amount of time needed to sterilize most organisms is dependent upon the temperature and pressure. At 121°C in a vessel pressurized to 15 psi, autoclave bags of solid laboratory waste require at least 30 minutes to sterilize.

The autoclave's superheated steam must contact all areas of the load. Bottle caps should be loose during the autoclave cycle so vapor expanding during heating can penetrate and will also

avoid a possible explosion. Do not overfill containers and bags. Steam and heat cannot penetrate as easily to the interior of a densely packed bag. The innermost part of the bag may not be properly treated. An over packed chamber does not allow efficient steam distribution. Considerably longer cycle times may be required to achieve decontamination if an autoclave is tightly packed.

The autoclave may offer dry heat cycles. These can be utilized for sterilizing laboratory supplies, which can withstand high temperatures but would be affected by a steam cycle. The required exposure times for dry heat will depend on the material composition, load volume, and can require significantly more time than what would be required for steam sterilization at the same temperature. Because the required times for dry heat sterilization will vary on the material treated and the surface and content, it is best to vary initial loads to determine the most effective temperature and length of cycle.

Factors that will affect your selected cycle times include:

- solid or liquid material
- volume of material to be autoclaved
- shape and size of containers used
- thermo conductivity properties of the container
- viscosity of the liquid
- density of the material
- position of the load within the autoclave chamber
- manufacturer's recommendation for media sterilization

It is important to recognize the length of time required to achieve this temperature will be dependent upon the factors stated above. In addition the sensing device which records the cycle temperature is not located within the load of material being autoclaved rather on the periphery. Therefore additional time may be required to ensure the centre of the load has achieved this temperature. For example, with samples that have a very high biological load the contact time might require as much as 60 minutes.

4.0 PACKAGING

a) Use only approved autoclave bags (capable of withstanding temperatures of at least 127°C)
b) Ensure items are labeled with contact information. Biohazardous waste awaiting autoclave treatment must bear the EHS hazardous waste label available from the EHS office

c) Do not overfill containers and bags
d) If outside of bag is contaminated, double bag
e) Never store hazardous waste directly on the floor. Use a secondary container.
f) Indicator tape or temperature sensitive strips must be used on each load
g) Do not put sharp or pointed objects into an autoclave bag. Use an appropriate puncture resistant sharps container.

5.0 LOADING

Before loading the autoclave;

a) Visually confirm the pressure within the chamber is at zero before opening the autoclave door
b) Check inside the chamber for any items left by the previous user that could pose a hazard.
c) Ensure that all materials are compatible with autoclaving. Some bags can impede steam penetration while others may melt.
d) Do not mix contaminated and clean items together during the same autoclave cycle. Clean items generally require shorter decontamination times (15-20 minutes) while a bag of infectious waste (24" x 36") requires a longer cycle to be effectively decontaminated throughout.
e) Autoclave bags should be loosely opened to allow steam penetration and to prevent shattering
f) Loosen the caps of liquid containers to allow steam penetration and prevent shattering

When loading the autoclave;

a) Load the autoclave according to manufacturer recommendations
b) Arrange containers, bags and trays in a manner that allows steam to circulate. Avoid stacking or crowding.
c) Use secondary containers (e.g. autoclavable trays) to contain spills
d) Ensure the door is fully latched and that the correct cycle has been selected.
e) Complete the autoclave log with all required information.

6.0 OPERATION OF AN AUTOCLAVE

Each autoclave will have specific instruction for its own use. It is important to follow the manufacturer's recommendation and each user must receive hands-on training on its proper use.

Determining the Correct Cycle Time

As the cycle time will vary with the composition of the load, it is important to determine the appropriate time requirement. The following steps outline this process.

(a) Select the typical bag size and contents
(b) Place a biological indicator in the centre of the load
(c) Attach a string or tape to it to allow easy retrieval
(d) Run the autoclave using the typical cycle selection
(e) Retrieve the biological indicator and process as per manufacturer's instructions
(f) Upon a "FAILED" test, increase the cycle time by 15 minutes and retest
(g) Retest until the indicator proves a successful autoclave cycle.

Autoclave Operation

Select the appropriate cycle type and length of time and start the autoclave. Waste must be autoclaved using settings that have been proven by biological indicators to be consistently successful for sterilization.

Ensure that the autoclave reaches the desired temperature before leaving it unattended. If the door begins leaking steam, abort the cycle and tighten the door further. If you must abort, press the abort button on the control panel. Wait until pressure is zero before resealing the door and reattempting the cycle.

Cycle Failure

If a problem is encountered and the load does not undergo the full cycle treatment, then the load must be re-treated with a complete cycle. Record any problems in the Autoclave Log or Problem Log.

7.0 UNLOADING

The process of unloading the autoclave poses the greatest risk of injury to autoclave users. Not only is there potential for serious burns from steam and contact with heated materials, but hazardous vapours may be produced by the inadvertent autoclaving of incompatible chemicals. Consequently, extreme caution must occur during this final stage.

When unloading the autoclave;

a) Don appropriate PPE
b) Verify the autoclave cycle log to confirm decontamination parameters were met

c) Prevent steam burns and shattered glassware by visually confirming the pressure within the chamber is at zero before opening the autoclave door
d) Open the door slightly and stand back to allow steam to escape.
e) Ensure items have cooled before removing and place in a safe location.
f) Material removed from the autoclave upon decontamination, should be clearly marked as treated (e.g. defacing biohazardous symbol)
g) Record any problems encountered in the Autoclave Log and report to responsible person

Remember, agar plates will melt and the agar will become liquefied when autoclaved. Avoid contact with molten agar. Use a secondary tray to catch any potential leakage from an autoclave bag rather than allowing it to leak onto the floor of the autoclave chamber. If there is a spill inside the autoclave chamber, allow the unit to cool before attempting to clean up the spill. If glass breaks in the autoclave, use tongs, forceps or other mechanical means to recover fragments. Do not use bare or gloved hands to pick up broken glassware.

8.0 DISPOSING OF TREATED BIOMEDICAL WASTE

After successful autoclave treatment, all bags must have any biohazard symbols defaced. These bags must then be placed inside a black domestic garbage bag which is free of markings or labels, and then tied closed. The waste may then be disposed of through the municipal system (i.e. drain or landfill).

9.0 RECORDKEEPING

Type of Record	Minimum Retention Period
Autoclave Logs	2 years
Training	1 year after the individual has left the facility
Maintenance including repairs, testing and certification	5 years
Performance and efficacy verification	5 years
Incidents potentially causing injury or exposures	10 years

Autoclave use log (each load of material shall be logged as follows):

- Date, time, and operator's name
- Contact information: Laboratory, room number, phone number
- Type of material sterilized
- Temperature, pressure, and length of time the load is sterilized
- The autoclave print-out or the cycle wheel when filled.

10.0 CYCLE VERIFICATION

Tests using biological indicator ampoules such as *Geobacillus stearothermophilus* to prove that autoclaving has rendered the laboratory waste non-hazardous must be conducted at least every 6 days or before any waste is placed into the municipal waste stream as specified in Guideline C – 17. Autoclave tape indicators can only verify that the autoclave has reached normal operating temperatures for decontamination. Most chemical indicators change color after being exposed to 121°C, but cannot measure the length of time spent at 121°C.

Procedures

1. Place ampoules in waste:
   a) Solid waste: place ampoule in the middle of the contents
   b) Liquid waste: (e.g. Flasks containing liquid) ampoule must be suspended in such a way that ampoule is not in contact with the liquid

2. Autoclave waste as per normal operating procedures.

3. On completion of autoclave cycle:
   a) Retrieve the ampoules from waste and allow to cool
   b) Process biological indicator as per manufacturer's instructions
   c) Incubate at 55 - 59 C, for <u>at least</u> 24 hours

Important Note: A control ampoule (an ampoule that has not been autoclaved) MUST be incubated with the test ampoules

4. Checking the Results:
   a) After 24 hours, check for a color change in ampoules (indicating growth) to determine if the results are positive or negative
      ★ <u>Negative growth in test ampoule</u>: Autoclaved waste can be discarded as non-hazardous treated biomedical waste

      ★ <u>Positive growth in test ampoule</u>: Autoclaved waste NOT to be thrown out. The waste must be re-autoclaved for a longer cycle time and must be re-tested with new ampoules, using the procedures above, until a negative test is reached.

      ★ <u>Control ampoule</u>: Must show positive growth. If it does not, check the expiry date of the ampoules. The control ampoule must be autoclaved prior to disposal.

5. Document test results on Autoclave Cycle Verification Testing Table. Save cycle printout (if available)

11.0 INCIDENT REPORTING

Do not to leave an autoclave operating unattended for a long period of time. Always be sure someone is in the vicinity while an autoclave is cycling in case there is a problem.

When an accident arises out of an autoclave's operation resulting in injury or death to a person and/or property damage, report to your supervisor. Obtain medical treatment in the event of a serious injury or illness by calling the Department of University Safety at ext. 4444. Participate in the investigation and completion of the Supervisor's Incident Investigation Report Form by providing information regarding the circumstances that resulted in the injury/incident.

In the event of a critical injury as defined under the Occupational Health and Safety Act, Regulation 834, the Supervisor has the following and immediate responsibilities:

•Call University Safety at ext. 4444 to co-ordinate emergency services response (police, fire and ambulance).

•Call the Assistant Director, Environmental Health and Safety at 520-2600 extension 1108 for Ministry of Labour notification.

•Attend the scene of the incident as soon as possible and secure the area.

12.0 MAINTENANCE

Regular Maintenance Check

• It is recommended that a daily or weekly maintenance check be done as appropriate for the frequency of use. Check door gaskets, interior surfaces, screens, exterior surfaces and any other components. Clean or disinfect exterior surfaces and clean interior surfaces and screens.

• Before every load, inspect primary and secondary containers for cracks, stress fractures, chips, and other damage.

Yearly Maintenance and Inspections

• It is recommended that all autoclaves receive maintenance by trained personnel, annually or based on manufacturer's recommendations.

• Autoclave pressure vessels fall under the Technical Standards and Safety Authority (TSSA) Regulation on Boilers and Pressure Vessels if they have a maximum working pressure greater than 15 psi (103kPa), a capacity greater than 1.5 cubic feet (42.4L), and an internal diameter

greater than 6 inches. Autoclaves which fall under the TSSA Regulation must undergo yearly inspections by TSSA certified individuals.

Repairs or Alterations

Repairs and alterations of autoclaves must be conducted by a certified contractor, the manufacturer or Facilities Management and Planning.

13.0 ADDITIONAL INFORMATION

For additional information on Autoclave Safety, please contact the Manager of Laboratory and Academic Program Safety at ext. 3000 or by email at ehs@carleton.ca.