**Standard Operating Procedure**

**Safe Autoclave Use**

***This is an SOP template and is not complete until:*** *1) lab specific information is entered into the box below 2) lab specific protocol/procedure is added to the protocol/procedure section and
3) SOP has been signed and dated by the Equipment Manager and relevant lab personnel.*

 Print a copy and keep one by the autoclave, and store a copy with your
*Chemical Hygiene Plan* and/or *Lab Safety Resources Binder*

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| --- | --- |
| **Department:** | Click here to enter text. |
| **Date SOP was approved by PI/lab supervisor:** | Click here to enter a date. |
| **Equipment Manager:** | Click here to enter text. |
| **Lab Safety Coordinator/Lab Manager:** | Click here to enter text. |
| **Lab Phone:** | Click here to enter text. |
| **Office Phone:** | Click here to enter text. |
| **Emergency Contact:** | Click here to enter text. |
| *(Name and Phone Number)* |
| **Location(s) covered by this SOP:** | Click here to enter text. |
| *(Building/Room Number)* |

**Type of SOP:** ☐ Process ☐Hazardous Chemical ☐Equipment

1. **Introduction**

The Autoclave is a specialized piece of equipment designed to deliver heat under pressure to a chamber, with the goal of decontaminating or sterilizing the contents of the chamber. The autoclaving process is typically used to destroy microorganisms and disinfect labware or equipment.

1. **Risks/Hazards**

Since autoclaves use heat, steam and high pressure for sterilization, the potential hazards and safety risks (the risk is highest when unloading the autoclave) for operators include:

* Broken glassware if the autoclave door is opened too quickly, and sufficient time is not provided for glassware to approach room temperature.
* Super-heated liquids also pose a risk if shaken or moved during the cooling process.
* Vapors and gases from the accidental autoclaving of volatile chemicals.
* Heat burns from autoclave chamber walls and doors and/or hot materials.
* Steam burns from the steam coming out of the autoclave and materials following completion of cycle.
* Scalds from hot fluids due to boiling liquids and/or spillage in autoclave.
* Autoclaving certain chemicals may cause an explosion.
* Explosions can also occur when the seal of the autoclave door malfunctions or when autoclave is improperly loaded. If sealed containers are used they may explode during the autoclave process.
* In accordance with local and state regulations, all biohazardous waste must be biologically-inactivated before it is disposed of as regular trash. It is against OSHA regulation to dispose of non-decontaminated waste in the common waste stream.
1. **Safety Expectations**

In order to ensure the health and safety of personnel using the autoclave, it is imperative that each department maintain the autoclave equipment as per manufacturer and safety guidelines, and ensure personnel are trained in the proper use of an autoclave.

* Supervisors are responsible for ensuring that employees are properly trained before operating any autoclave units.
* Name of the individual responsible for the autoclave is to be posted near the autoclave. This Standard Operating Procedure should be present near the autoclave unit for reference.
* Documentation of training, which includes the signature of both the supervisor and the individual trained, must be kept by the supervisor.
* Personal protective clothing and equipment must be worn and made available by the department for when operating an autoclave.
* Procedural and instructional documents provided by the autoclave manufacturer must be kept, read, and adhered to when working with autoclave unit.
* Autoclaves must be inspected at least annually by a professional. Inspection services may be managed by the manufacturer’s preventative maintenance contract. (Contact EH&S for a list of autoclave maintenance companies)
* The individual responsible for the autoclave should perform a basic visual inspection of the autoclave on a monthly basis (see Maintenance/Repair section for further information).
1. **Personal Protective Equipment (PPE)**

Autoclaves utilize steam, heat, and pressure to complete the sterilization cycle. Therefore, the appropriate Personal Protective Equipment (PPE) must be worn.

* **Eye/face protection – safety goggles /face shield**; safety goggles worn always; face shield worn if there is a possibility of splash back
* **Heat-insulating gloves** that provide complete coverage of hands and forearm
* **Lab coats** – long sleeved and knee length
* **Close toes shoes** – ensure the foot is entirely covered
* **Long pants or long skirt** – legs must be covered
1. **Training**

The fundamentals of autoclave safety are discussed in [Autoclave Safety Training](https://oregonstate.bridgeapp.com/learner/programs/b647156e/enroll) as offered on Bioraft found on the EHS training website (<https://ehs.oregonstate.edu/training/training-materials>). Supervisors must ensure that all personnel, whether they have taken the online autoclave training or not, have successfully completed an in-person training session on the safe operating procedures prior to using the autoclave. This requirement is applicable to both new and experienced personnel. All in-house training must be documented, dated and signed by both the trainee and trainer, and available to view upon request. Documentation of this and any other in-house training should be kept by the supervisor for a minimum of 3 years after the person has left the lab.

1. **Uses and Limitations of the autoclave**

Although autoclaving is an economical and environmentally friendly way of sterilizing and decontaminating items, not all materials can be autoclaved. In fact, some materials present specific hazards if autoclaved.

Items that **CAN** be autoclaved include, but are not limited to:

* Cultures and stocks of infectious material
* Culture dishes and related devices
* Discarded live and attenuated vaccines
* Contaminated solid items such as: petri dishes, eppendorf tips, pipettes, gloves, paper towels, lab coats, solid and liquid waste
* Items for sterilization such as: glassware, media, liquid solutions
* Some equipment -ask supervisor for confirmation

Items that should **NOT** be autoclaved include:

* Chemicals (includes most disinfectants, e.g. bleach)
* Samples containing solvents or substances that may emit toxic fumes
* Radioactive material
* Certain plastics that may melt upon high heat/pressure
1. **Material Preparation**

Ensure that all material is prepared properly prior to autoclaving:

* Check to confirm the materials can be autoclaved
* Inspect glassware for cracks prior to autoclaving

*Primary Container*: the container that comes into direct contact with the contaminated material or fluid to be autoclaved. The primary container must allow steam penetration, while avoiding pressure build-up. Examples of primary containers include flasks or vials containing liquids, autoclave bags etc. **Therefore, the container *CANNOT* be sealed – sealed containers may explode in an autoclave!** Loose seals can be achieved by:

* Loosening screw caps or using self-venting caps.
* Capping open containers for sterilization with aluminum foil.
* Opening plastic (autoclave) bags slightly prior to loading them into the autoclave. Autoclave bags:
	+ Are typically made of polypropylene (PP), which is strong and puncture resistant.
	+ Come in a variety of sizes and may or may not have the international biohazard symbol printed on it.
	+ Keep in mind polypropylene does not have good steam permeability. To ensure that steam reaches the contents – autoclave bags must be opened prior to autoclaving.

*Secondary Container*: the container used to hold the primary container, which will prevent any spills from occurring.

* Primary containers must be placed into trays or buckets big enough to contain any materials that may melt or spill during the autoclaving.
* These secondary containers must be made of a material that can withstand repeated autoclaving. (Typically, metal or plastic autoclave trays are used as a secondary containment)
* Materials being stored and/or transported to the autoclave must be kept in secondary containment at all times!
1. **Loading Procedures**

Prior to using an autoclave verify that the autoclave has been functioning correctly by reviewing the previous cycle log recordings (time, temperature, pressure, and notes).

* Wear the appropriate PPE required to safely handle the material being loaded into the autoclave.
* Place material in autoclave. Do not mix solid and liquid materials.
* Avoid overloading the chamber, or compressing the contents: this will impede steam penetration.
* Containers holding liquids should not be more than 75% full. This allows for liquid expansion, thereby preventing overflow.
* Place packages on their edges to enhance flow of steam.
* Place empty flasks or tubes horizontally to prevent trapping air pockets.
* Ensure containers do not touch each other so all surfaces are sterilized.
* Ensure all containers allow steam penetration (slightly open autoclave bags).
* Close and latch autoclave door firmly.
1. **Operating Procedures**

Before using the autoclave, all users must have taken Autoclave Safety Training (as found on the Bridge website) and have documented in-person training.

* Close and lock door.
* Choose the appropriate cycle (e.g., gravity, liquid, or dry cycle) for the material. Consult the autoclave manual for assistance in choosing a cycle. The manuals for operation of the autoclave should be located near the autoclave.
* Set appropriate time and temperature if you are using a customized cycle.
* Start your cycle and fill out the autoclave user log with your contact information. A completed cycle usually takes between 1-1.5 hours, depending on type of cycle.
* Do not attempt to open the door while autoclave is operating.
* If a problem with the autoclave is perceived, abort the cycle and contact the person in charge of the autoclave immediately.
1. **Unloading Procedure**

The greatest risk of personal injury occurs during the process of unloading the autoclave. Refer to the Risks/Hazards section for a list of potential hazards and/or risks operator may encounter; exercise caution to eliminate the possibility of such occurrences when operating the autoclave.

* Wear necessary PPE: heat-insulating gloves, eye protection if deemed necessary, lab coat, closed-toe shoes and wear clothing that covers your legs.
* Ensure that the cycle is complete and both the temperature and pressure have returned to a safe range. Check chamber pressure gauge before opening door: the gauge should read **ZERO**.
* Carefully open the autoclave door a little bit, taking care to avoid the steam. This will allow the steam to escape while simultaneously allowing the pressure within liquids and containers to stabilize.
* Do not disturb containers of super-heated liquids or remove caps prior to unloading these materials. Gently transfer containers to trolley.
* Check autoclave tape for color change and cycle log recorder for time and temperature attained.
* If disposing of biological liquid waste after autoclaving, first allow to cool before pouring it down the drain.
1. **Autoclave User Log**

Entries must be placed in the User Log each time the autoclave is used. These records are used for maintenance/service schedules and reporting of incidents, accidents and/or faults.

* Entries should include: operator's name, lab name, date, time/duration, chamber pressure, and condition of the autoclave.
* The log book must be kept adjacent to the autoclave.
* An Autoclave Use Log example is provided in this document.
1. **Maintenance and Repair**
* Autoclave must not be used by any individual until the autoclave has been deemed safe for operation.
* Do not attempt to make repairs- only qualified professionals are permitted to make repairs to the autoclave.
* Report possible malfunctions to the individual responsible for the autoclave, as soon as possible, so that repairs can be scheduled with the autoclave company/supplier.
* Monthly maintenance should be performed on each autoclave using the following guidelines:

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| **Inspect door gasket for damage and cleanliness**  | With the autoclave switched OFF and the door OPEN, visually inspect the gasket and its mating surfaces for signs of debris, damage and corrosion. Clean by wiping with a damp cloth. |
| **Check the autoclave temperature probes for signs of damage** | With the autoclave switched OFF and the door OPEN, visually inspect the probe at the rear of the inside of the vessel for signs of debris or damage. |
| **Door operation** | Close the door to check all functions are correct in conjunction with the operating manual. |
| **Water condition**  | Check the condition of water in the water tank or trough.  |
| **Make sure that the chamber drain strainer (which can be found in the front drain hole of the sterilizer) is free of debris.**  | This should ideally be done once per day because a clogged chamber drain strainer will prevent your autoclave from sensing temperature. |
| **Wipe up any spills in the autoclave chamber with a chamber cleaning solution or water.** | Do not use hypochlorites that contain bleach, corrosive chemicals, acids or seawater, as these can damage the stainless steel chamber. |
| **Visually check for water and/or steam leaks** |  If you see a leak, identify the root cause and call in a professional for repairs as soon as possible. |

1. **Equipment Malfunction**

If the autoclave does not operate exactly as expected, do not attempt to fix the problem. A notice shall be placed on the autoclave indicating that it is not to be used until the problem is diagnosed and corrected.

* Record the problem in the autoclave log book.
* Contact or your supervisor to report the problem.
* Keep in mind only qualified professionals are permitted to make repairs.
1. **Incident Response**

If an accident happens the following documents must be completed:

* Online OSU HR Advocate Public Incident Reporting Form within 24 hours of the incident
* If the student or employee’s incident resulted in the need for medical treatment, have the employee complete the worker section of the SAIF 801 Form and fax to risk management at 541-737-4855 within 24 hours.
* If any injury occurs, seek first aid and/or medical assistance as deemed necessary by the degree of the injury.
* If clothing absorbs hot water/steam, remove clothing, and apply cool water to the affected body part.
* A notice must be placed on the autoclave to indicate that the unit is out of service until the cause of the incident is identified, pro-active measures are taken to prevent such incidents in the future, and the autoclave is deemed safe for operation.
* A near-miss report must be filled out if an injury/accident nearly occurs
1. **Spill Clean-Up**

Spills may occur due to a boil-over or breakage of containers during the autoclave procedure.

* Use of secondary containers will make spill cleanup much easier – once cooled can be poured down the drain.
* No operation of the autoclave should be allowed until the spill is cleaned up. (Spills not cleaned up will become harder to remove)
* The operator is responsible for the clean-up of the spill. Wait until the autoclave and materials have cooled down to room temperature before attempting to clean-up the spill.
* If spill of biological material occurred before autoclaving (during loading), follow your lab’s spill procedures found in your lab’s biosafety manual/emergency response plan. If spill is found after autoclaving, then any biological material should no longer be hazardous (autoclaving procedure if done properly will make the biologicals non-viable).
* Dispose of cracked glassware properly.
* All spills to be reported to principal investigator/supervisor – record of spills to be kept.
* For further information on how to respond to spills please go to <https://ehs.oregonstate.edu/spill-response>
1. **Lab Specific Protocol/Procedure**

[*Include any lab-specific protocols/procedures here. This should include the types of waste or cultures that will be autoclaved, and the run time required to sterilize the specific items*]

1. **References**
2. CDC Biological and Infectious Waste <http://www.cdc.gov/nceh/ehs/etp/biological.htm>
3. Biosafety in Microbiological and Biomedical Laboratories (BMBL) 6th Edition, 2020. <https://www.cdc.gov/labs/BMBL.html>
4. Arizona State University, Standard Operating Procedures; Safe Autoclave Operations. <https://nielsenlab.engineering.asu.edu/wp-content/uploads/2016/08/SOPs-Nielsen_Lab_ASU.pdf>
5. University of Toronto, Standard Operating Procedures; Safe Autoclave Operations. <https://ehs.utoronto.ca/wp-content/uploads/2019/04/Autoclave-SOP.pdf>

**Documentation of Training** (signature of all users is required)

* Prior to conducting any work with the autoclave designated personnel must provide training to his/her laboratory personnel specific to the hazards involved in working with this substance, work area decontamination, and emergency procedures.
* The Principal Investigator must provide this SOP to all laboratory personnel.
* The Principal Investigator must ensure that his/her laboratory personnel have attended appropriate laboratory safety training or refresher training.

**Principal Investigator SOP Approval**

By signing and dating here the designee certifies that the Standard Operating Procedure (SOP) for *Insert SOP Name* is accurate and effectively provides standard operating procedures for laboratory personnel.

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Signature Printed Name/Title Date

By signing this document, I acknowledge that I have read and understand the content of this SOP and have had in-person autoclave specific training:

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| **Name** | **Signature** | **Date** |
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|  |  |  | **Autoclave Log** |  |  |
| **Building & Room:**  |  |  |  |  |  |
| **Primary Contact for the Autoclave:**  |  |  |  |
|  |  |  |  |  |  |  |
| **DATE OF TREATMENT**  | **START TIME** | **RUN TIME** | **CHAMBER PRESSURE** | **CONDITION OF AUTOCLAVE** | **LAB NAME** | **USER NAME** |
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