SOP – Autoclave Operation

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

Risks/Hazards

- Broken glassware if the autoclave door is opened quickly, and sufficient time wasn't provided for glassware to approach room temperature.
- Super-heated liquids if shaken or moved during the cooling process.
- Vapors and gases from 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 if the seal of the autoclave door malfunctions or when autoclave is improperly loaded. Sealed containers may explode during autoclaving.

Safety

- Documentation of training includes signature of both the supervisor and individual trained (use Biosafety MOU).
- Personal protective clothing and gear must be worn when using an autoclave.
- Procedural and instructional documents from the autoclave manufacturer must be kept, read, and adhered.
- Autoclaves must be inspected at least annually.
- Inspection services can be performed by the manufacturer (contract). The inspection, service and repair records must be available for request for Health and Safety Officers.

Personal Protective Equipment

- Eye/face protection – safety goggles/face shield; if possibility of ocular splash
- Heat resistant gloves.
- Lab coats – long sleeved and knee length.

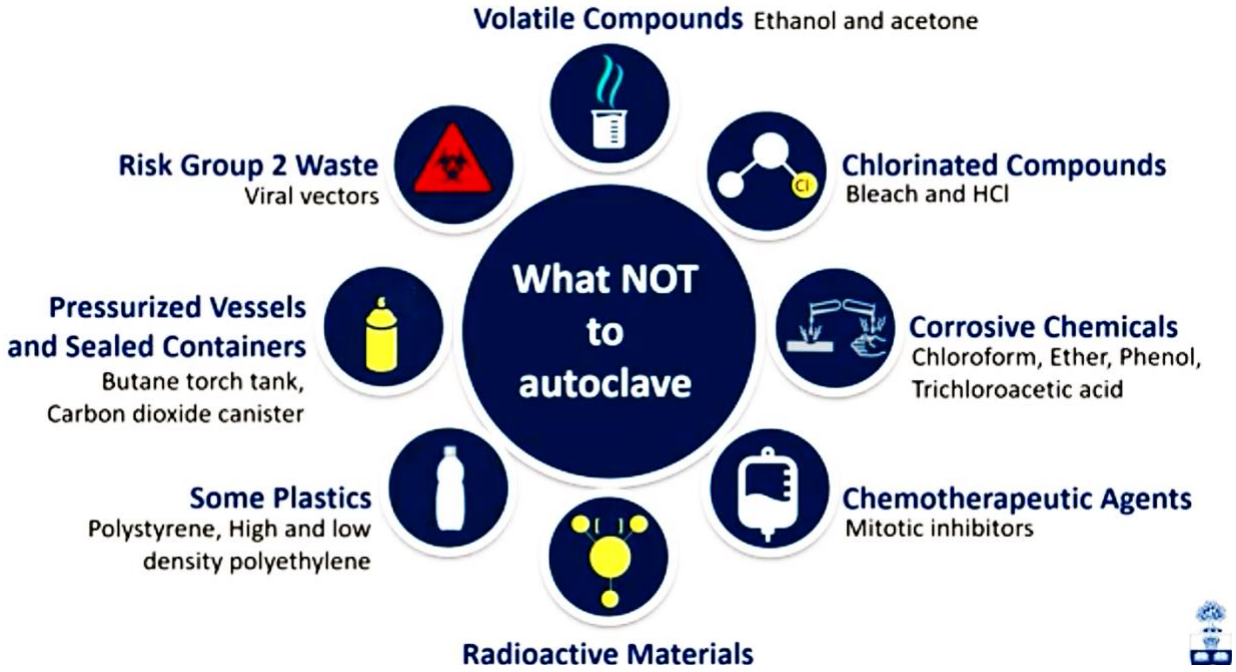
Operator Instructions

Training

The fundamentals are covered in **EHS601**. Supervisors must ensure personnel, whether taken **EHS601** or not, have completed a training session on safe operating procedures prior to using the autoclave. All in-house training **must** be documented, dated and signed by the trainee and trainer.

Limitations (Do Not Autoclave):

- Chemicals (includes most disinfectants, e.g. bleach).
- Radioactive materials.
- Some plastics – ensure materials are autoclavable.



Permitted For Autoclaves:

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

Material Preparation

- Confirm materials can be autoclaved.
- Inspect glassware for cracks prior to autoclaving.

Primary Container: is in direct contact with the sample to be autoclaved. The container **must** allow steam penetration, and not sealed to prevent an explosion. Examples include flasks or vials containing liquids, autoclave bags etc.

Loose seals by:

- Loosening caps.
- Capping containers with aluminum foil.
- Opening polypropylene (PP) bags before loading into the autoclave.

Secondary Container: holds the primary container for preventing spills.

- These containers must be able to withstand repeated autoclaving. Metal or plastic autoclave trays are often used.

Transporting Materials to the Autoclave

- Use a secondary container to collect any spillage in case of an accident during transport.

Autoclave Tape (chemical indicator)

- Apply to each item that will be autoclaved.
- Black lines appear when the tape is exposed to high temperatures.

Autoclave Procedure

Loading an Autoclave

- Verify previous cycle logs (time, temperature, and pressure)
- Wear appropriate PPE.
- Place samples in the autoclave. **Do not** mix solid and liquid materials.
- Avoid overloading the chamber, or compressing contents that can impede steam penetration.
- Containers holding liquids **should not** be more than **75%** full. This allows liquid expansion and prevents overflow.
- Position items on the side to enhance flow of steam, and ensure containers allow steam penetration (slightly open autoclave bags).
- Close autoclave door firmly.

Operating an Autoclave

- Ensure autoclave door is locked.
- Choose the appropriate cycle. Factors include:
  - Whether goal is decontamination or sterilization.
  - Composition of the load (solid or liquid).
  - Density of the material.
  - Volume and viscosity of liquids (i.e. larger volumes will require more time).

Cycle Selection:

- **slow exhaust** = for liquids to prevent contents from boiling over.
- **fast exhaust** = for glassware.
- **fast exhaust and dry** = for wrapped items.
- **Do not** open the door while the autoclave cycle is occurring.
- If there's a problem with the autoclave, abort the cycle and contact CRAFT staff.

Unloading an Autoclave

- Wear appropriate PPE: heat-insulating gloves, eye protection if necessary, lab coat, etc.
- Ensure the cycle is complete and both the temperature and pressure returned to a safe range. Check chamber pressure gauge is zero.
- Open the autoclave door slightly to allow steam to escape while simultaneously allowing pressure in liquids and containers to stabilize.

- Do not disturb containers of super-heated liquids or remove caps prior to unloading these materials.
- Check autoclave tape for colour change and cycle log for time and temperature attained.
- If disposing biological liquid waste, allow to cool before pouring down the drain.

Maintenance and Repair

- Only qualified professionals are permitted to make repairs to the autoclave.
- Report malfunctions to CRAFT staff so repairs can be scheduled with the supplier.

Incident Response

- All incidents and spills, must be reported to the supervisor and department.
- For injuries, seek first aid and/or medical assistance, and fill out an Accident report within 24 hours.
- If clothing absorbs hot water/steam, remove clothing and apply cool water/ice to the affected body part.

Spill Clean-Up

- Once spills in the secondary containers is cooled, it can be poured down the drain.
- Wait until the autoclave and materials have cooled to room temperature before cleaning up the spill.
- For biological spills before autoclaving (during loading), follow the spill procedures in the biosafety manual/emergency response plan. If the spill occurred after autoclaving, then the biological material should no longer be hazardous.
- Dispose of cracked glassware properly.
- All spills to be reported to principal investigator/supervisor, CRAFT staff.