CM SOP #104 – Autoclave Procedures

Original release date: 02/08/2016 Version: 7 Date last revised: 03/01/2026

I. Purpose & Scope

This SOP describes procedures for the preparation, sterilization, and verification of materials processed in Comparative Medicine autoclaves. Autoclaves use saturated steam under pressure to achieve sterilization by destroying microorganisms, including bacterial spores. Effective sterilization depends on the proper combination of temperature, pressure, steam saturation, and exposure time. This SOP applies to all Comparative Medicine (CM) personnel responsible for preparing materials for autoclaving or operating CM autoclaves. Autoclave use supports routine vivarium operations including sterilization of caging, water bottles, bedding, surgical instruments, and decontamination of biohazardous waste.

II. Roles & Responsibilities

Comparative Medicine Staff

- Prepare materials appropriately for autoclaving.
- Ensure items are compatible with steam sterilization.
- Use appropriate sterilization indicators for each load.
- Follow approved loading and unloading procedures.
- Document sterilization activities and report irregularities.
- Keep effectiveness records and log
- Follow required health and safety protocols

Research Staff, Students, Visitors

Not applicable

CM Management

- Ensure personnel are trained in autoclave operation and sterilization procedures.
- Monitor compliance with sterilization verification requirements.
- Ensure appropriate sterilization indicators and supplies are available.

CM Director and AV

- Provide oversight of sterilization procedures affecting animal housing and research equipment.
- Ensure sterilization procedures align with institutional biosafety and animal welfare standards.

III. General Notes & Definitions

- Autoclaves sterilize materials using steam under pressure, typically at 121°C (250°F).
- Effective sterilization requires adequate steam penetration and proper load configuration.
- Overloading the autoclave or tightly sealing containers may prevent proper sterilization.

- Materials must be compatible with high temperature and pressure conditions.
- **Items That Must NOT Be Autoclaved**
  - Flammable liquids (e.g., alcohols, solvents)
  - Oils or waxes
  - Formalin or volatile chemicals
  - Radioactive materials
  - Sealed containers or tightly capped bottles
  - Dried bleach or bleach-containing materials
  - Pathological waste such as carcasses or tissues (must be incinerated)
  - Sharps prior to placement in approved sharps containers
- Autoclave or sterilization indicators are test tools used to verify the sterility of autoclaved materials or water. They come as Biological Indicator (BI) or chemical indicators. Chemical Indicators are available as temperature strips or temperature tape
- Serious injuries can occur from hot surfaces and from the release of live steam. The use of long heat-resistant autoclave gloves is necessary.
- **Sterilization**: process of completely removing, destroying, or inactivating all forms of microbial life
- **Pathogens**: A subset of microorganisms that cause infectious diseases
- **Biological Indicator (BI):** a sterilization monitoring device containing highly resistant bacterial spores

IV. Materials & Equipment

- Surgical drape or Autoclavable bags
- Autoclave-safe trays
- Biological Indicator
- Chemical indicator: steam sterilization strips and temperature tape
- Permanent marker
- Personal protective equipment (PPE): gloves, lab coat, heat resistance gloves.

V. Procedure

A. **Preparation for Autoclaving Biohazardous Waste**
   1. Collect required supplies:
      a. Autoclavable biohazard bags
      b. Chemical indicator strips
      c. Biological indicator (when required)
      d. Autoclave tape
   2. Fill biohazard bags **no more than ½–¾** full to allow steam penetration.
   3. Place the following inside each bag or pack:
      a. One chemical indicator strip
      b. Biological indicator if required by procedure or schedule.
   4. Label bags with:
      a. Date
      b. Autoclave location
      c. Operator initials if required.
   5. Prepare the bag closure using the gooseneck method:
      a. Twist the top of the bag.
      b. Fold the twisted portion over.

   c. Secure loosely with autoclave tape.
6. Do **not tightly seal the bag**, as steam must penetrate the load.
7. Each separate bag/pouch/unit being autoclaved should have a strip of temperature tape on it for easy visualization as to whether the items have gone through a cycle
8. Place prepared bags in an autoclave-safe tray.

B. **Preparation of Materials Requiring Sterilization Prior to Use**
1. Water Bottles
   a. Fill bottles approximately ¾ full.
   b. Loosen caps to prevent pressure buildup.
   c. Place bottles in racks or cages for stability.
2. Rodent Caging
   a. Assemble cages with bedding, enrichment, food hopper, and lid.
   b. Place autoclave tape on cage lids to confirm exposure to heat.
   c. Avoid excessive tape that can leave residue.
3. Surgical Instruments
   a. Open hinged instruments fully.
   b. Place instruments inside an autoclave pouch or wrapped surgical pack.
   c. Include one chemical indicator strip inside each pack.
   d. Seal the pack using autoclave tape.
4. Label packs with:
   a. Date of sterilization
   b. Operator initials

C. **Autoclave Cycle Parameters**
1. Follow operational instructions in SOP 904: *Autoclave Operation and Care*.

Load Type	Temperature	Exposure Time
Liquids	121°C	30 minutes
Dry goods / equipment	121°C	30 minutes
Toxin-containing materials	Follow EH&S recommendations	

D. **Loading the Autoclave**
1. Ensure loads are not overcrowded.
2. Allow adequate spacing between items.
3. Place liquids in secondary containment trays.
4. Position packs to allow steam circulation.

E. **Assessing Effectiveness**
1. Confirm autoclave tape has changed color.
2. Verify internal chemical indicators show appropriate change.
3. Remove and process biological indicators according to *SOP 111: Biological Indicator Processing*.

F. **Failed Sterilization Cycles**
1. If the autoclave cycle terminates early or alarms, reprocess the load.
2. If chemical indicators fail, remove and repackage the load with new indicators and repeat sterilization.
3. If biological indicators fail, notify the supervisor immediately.
4. **Do not use** materials from loads that failed sterilization.

G. **Recordkeeping**

1. Record each autoclave run in the *CM Form 045: Autoclave Log* located beside each autoclave.
2. Record all results from the indicator in the *3M Attest Log* located in each tech office.
3. Log any failed temperature strip or failed BI tests from the autoclave on the *Health and Environment Check Sheet* and notify supervisor.
4. Retain *Autoclave Logs* in the dirty cage washroom for at least six months. After this timeframe they can then be filed.
5. Retain *3M Attest Log* (provided by 3M) for at least six months in the tech office. After this timeframe they can then be filed.

VI. Health & Safety

- The best practice to avoid burns is to allow autoclaved items to return to room temperature before handling them.
- Keep a safe arms-length distance from the opening of the autoclave chamber to avoid steam exposure
- Personal protective equipment (PPE) must be worn when unloading autoclaved items to avoid burns. Damaged/excessively worn PPE must be replaced.
- Never autoclave Oils, waxes, and flammable materials, radioactive materials, bleach, pathological waste
- Place a WARNING HOT MATERIALS sign where hot material is kept after autoclaving.
- If steam escapes the machine while the autoclave is running, turn the autoclave off at the panel and contact your supervisor immediately. DO NOT OPEN THE AUTOCLAVE DOOR.
- Contact supervisor in the event of an emergency.

VII. References & Attachments

- Product guide from temperature strip (or card) product and BI
- *Guide for the Care and Use of Laboratory Animals*, 8th Edition
- CDC Biosafety in Microbiological and Biomedical Laboratories (BMBL)
- OSHA Autoclave Safety Guidelines
- FAU Environmental Health & Safety policies
- Related SOPs & Forms:
  - *SOP 904: Autoclave Operation and Care*
  - *SOP 110: Autoclave Effectiveness* (combined here)
  - *SOP 111: Biological Indicator Processing*
  - *Form 045: Autoclave Log*

VIII. Revision History

Revision Date	Revision Number	Summary of Changes
04/20/17	2	Including CVT responsibilities, adding to safety and recordkeeping section, new section “general information”, removing detailed information regarding autoclave indicators (new SOP)
02/01/2018	3	Added References, update Preparation for Autoclaving & update Record Keeping

02/26/2018	4	Add procedures for destroying relevant toxins used in animals
05/07/2021	5	Updated procedures to reflect changes to SOP 110 and 111 and current processes.
02/28/2024	6	Update name of "Biological Indicator Sheet and Autoclave Log". Change owner to Director, Veterinary Care
03/01/2026	7	Updated format, made ADA compliant, refined language; combined 104 & 110 (Autoclave Procedures and Effectiveness, respectively)

Approved by: Nicole Compo, DVM, DVSc, DACLAM, Attending Veterinarian