

**STANDARD OPERATING PROCEDURE  
STAN MAYFIELD BIOREFINERY PILOT PLANT**

**TITLE: Autoclave Operation**

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**A. Scope**

This procedure describes how to operate the autoclave.

**B. Safety and Training Requirements**

Refer to UF lab safety policies regarding equipment listed in section D below before starting any process work.

Review the location of fire extinguishers, fire blankets, safety showers, spill cleanup equipment and protective gear before beginning any process work.

Refer to UF Biosafety guidelines and the NIH Guidelines whenever handling biological cultures/genetically modified organisms.

During operations in the plant, the following safety gear will be utilized at all times:

- Lab Coat
- Safety Goggles
- Protective Gloves (nitrile, neoprene)
- Autoclave gloves

To prevent serious burns and injuries:

- Always – read and understand operating instructions before starting.
- Always – use caution when opening door and removing contents.
- Never – attempt to open door with any pressure in chamber.
- Never – attempt to run machine with door open.
- Never – attempt service or repairs unless qualified and authorized.

Failure to do so may cause damage and void manufacturer's warranty.

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**C. Related Documents and SOPs**

1. UF Biosafety manual
2. Autoclave manual
3. Steam Supply SOP-9305

**D. Preparation/Materials/Equipment**

1. Autoclave
2. Autoclave gloves
3. Paper towels
4. Aluminum foil
5. Rubber bands
6. Autoclave tape

**E. Detailed Procedure**

1. Put autoclave gloves on.
2. Assure the steam valves are open in the back of the autoclave.
3. Assure that the Steam Supply is operating normally according to SOP-9305
4. Place the materials you wish to sterilize inside the autoclave and close the door.
  - a. When autoclaving flasks or calibrated cylinders, make sure they are covered with a folded paper towel that has been secured with a rubber band.
  - b. When autoclaving bottles or other screw-capped containers, make sure the cap is loose when placing it inside the autoclave (unless it can be pressurized).
  - c. When autoclaving instruments or other equipment/materials, make sure they are covered in aluminum foil in order to maintain sterility after the cycle is completed and the equipment/materials are removed from the autoclave.
  - d. Place a piece of autoclave tape on all the equipment/materials to be autoclaved.
5. Select type of cycle depending on type of material to be sterilized:
  - a. Gravity for dry materials
  - b. Liquid for liquids.

**CAUTION: When autoclaving liquids, make sure the container is loosely capped. If the container does not have a cap, make sure the level is no more than 40% of the total volume of the container.**

6. Set the sterilization time.

**NOTE: Biohazard waste must be sterilized for 60 minutes.**

7. Begin the sterilization by pressing on the "START" button. The sterilization time will begin counting down once the chamber is up to temperature.
8. The pressure in the chamber should be ~15 psi during sterilization.
9. After the sterilization cycle is complete, DO NOT open the autoclave door until the chamber pressure is at "0 psi."
10. Stand behind the autoclave door and open it slowly

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11. Take the equipment/materials out of the autoclave and place on a heat resistant surface to allow it to cool before using.

**F. Data Archival and Analysis**

Record all autoclave runs in the autoclave log sheet and keep next to autoclave at all times.