

Sterilizing Liquids and Labware Using the Autoclaves

Caution: Before autoclaving anything, make sure the material is autoclave-safe!

If you are sterilizing **EMPTY, dry glass labware, autoclave-safe plastic labware, foil-wrapped items, or any other NON-LIQUID item**, follow the steps here:

1. Prepare the items for sterilization (Fig #1):

glass bottles with caps - washed, air-dried completely, capped with cap half-loosened, autoclave tape on top of lid.

plastic large centrifuge bottles with caps - washed, air-dried completely, capped with cap very loosely covering the opening and NOT tightened, autoclave tape on top of lid.

glass test tubes with caps - washed, air-dried completely, capped firmly, put in non-labeled rack, autoclave tape on front of rack.

plastic pipet reservoirs/basins - washed, air-dried completely, wrapped in heavy-duty aluminum foil with no exposed corners or edges, 1 piece of autoclave tape on the whole batch only.

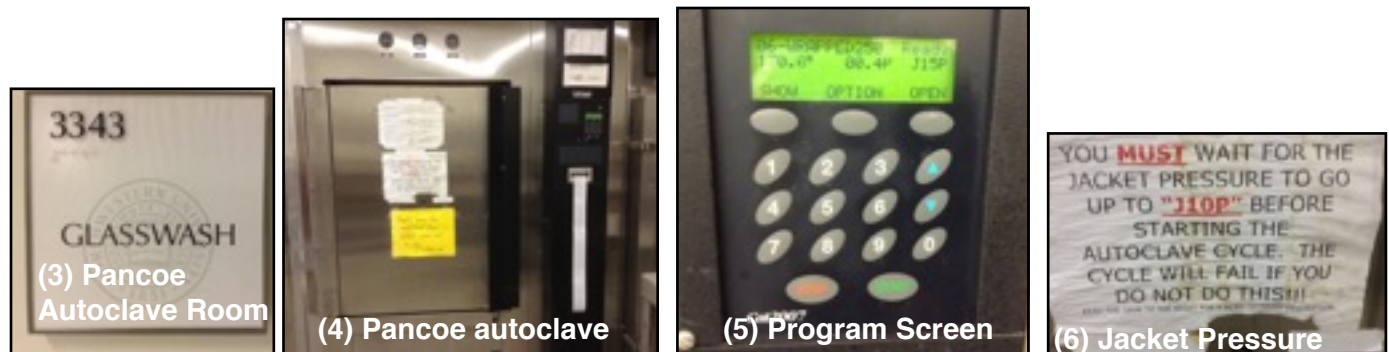
chunking spatulas or other glass/metal utensils - washed, air-dried completely, wrapped in aluminum foil with no exposed corners or edges, 1 small piece of autoclave tape on each item.

glass or plastic containers with no lids (Flasks, beakers, etc.) - washed, air-dried completely, covered in heavy-duty aluminum foil with no holes and aluminum hanging at least 1 inch over the edge of the opening, autoclave tape on top of the foil cover.



- Place all items in autoclave bins - make sure there is enough space between each item inside a bin (Fig #2). Locate and bring with you the pair of orange autoclave gloves.
- Use the large grey cart to bring the bins to the autoclave room in Pancoe 3343 (same as the plate pouring room, Fig #3).

NOTE: if this autoclave is busy, you can also use the Pancoe 4th floor autoclave. It is exactly the same type - USE THE AUTOCLAVE ON THE RIGHT SIDE in Pancoe 4343 for both wet and dry cycles and ONLY use the LEFT-SIDE autoclave for dry cycles (the liquid cycle is too long).



The autoclaves should always be left ON - BUT - sometimes they will be OFF when you are ready to use them. If the autoclave is OFF:

- Check on the panel that the Tab indicates 'FIXED' and not 'BROKEN'. If OK, proceed - otherwise try a different autoclave (Fig #4).
 - TURN ON the autoclave simply by pushing the 'ON' button above the keypad and Program screen (Fig #5). Make sure the door is closed. You will hear the steam start to run inside the jacket.
 - BEFORE STARTING A CYCLE, make sure the **Jacket pressure reads higher than J10P** on the program screen (or also look at the pressure gauge above the door) (Fig #6). *This may take a little longer to start up, but you can proceed to load the autoclave chamber during this time.*
- If the autoclave is free - proceed to loading:
 - Press 'OPEN' on the screen. STEP BACK and wait a minute for the autoclave to automatically open - you will hear the door unlock. PUT ON THE AUTOCLAVE GLOVES!!

CAUTION!! Step back away from the edges of the door to avoid getting burned by the steam, which may immediately vent.

- From behind the door, open the door fully and then position the autoclave cart in line with the rails inside the autoclave to unload the shelf (Fig #7).

NEVER!! load the autoclave without using the cart to support the shelf.
Be very careful to not touch anything without gloves on, since all surfaces are HOT!!

- Wearing gloves, pull out the shelf completely to access it from the side (Fig #8).



NOTE: If another lab's items are in the autoclave you can take the bins out and put them on any open peripheral bench tops in the room. **If the contents of the containers are AGAR Media, make sure to notify the owner lab that you removed these from the autoclave.** Any other items (liquid without agar and dry items) are OK to leave on the bench top.

- Load your bins onto the shelf. Generally, it is better to put the heavier things on the bottom (Fig #9).
- Push the shelf back into the autoclave completely (Fig #10).
- To disengage the cart from attaching to the autoclave, **push forward** on the black knob of the cart release handle (Fig #11); this will lift the gripping mechanism (Fig #12). Move the cart back away and close the door. Close the door gently but quickly enough that it is completely closed before the auto-lock engages. You should hear the door lock.

5. Check that the jacket pressure is greater than 10 psi. Select your cycle according to the posted program key (Fig #13 for 3rd floor and Fig #16 for 4th floor autoclaves). The numbers we use are **#06 - Wrapped, for Dry Labware** (30 min. sterilization with 15 minute drying - 1.0 hour for the Total Cycle, 3rd floor; and 20 min. sterilization with 20 minute drying - 45 min for the Total Cycle, 4th floor). PRESS START on the keypad to activate the cycle (Fig #14). The cycle parameters and progress log will print out on the paper below the screen (Fig #15). Reference this for start and stop times, if needed.

NOTE: ALWAYS monitor the screen for a minute until the cycle starts completely before leaving to ensure that the autoclave is operating properly. If the pressure starts increasing normally and no steam is leaking from the door edges, then everything should be fine. IF THIS IS NOT THE CASE, then press the 'STOP' button to abort the cycle and come get Sarah or Robyn.

Cycle	Temperature (°F)	Sterilization time (min.)	Exhaust time (min.)
01 - Unwrapped	250	30	5
02 - Unwrapped	250	45	15
03 - Unwrapped	250	20	10
04 - Unwrapped	250	30	15
05 - Wrapped	253	15	15
06 - Wrapped	253	30	15
07 - Wrapped	253	30	15
08 - Wrapped	252	5	5
09 - Liquid	260	15	0
10 - Liquid	255	45	0
11 - Liquid	250	30	0
12 - Liquid	252	15	0
13 - B&D Test	274	3.5	2
14 - Leak Test	???	???	???

(13) Program Key - 3rd Floor



(14) Keypad



(15) Cycle Progress Log Printout

4th floor ENB-WANCOE
Autoclave Programs

Program	Program Name	Temp (°F)	Sterilization time (min)	Drying time (min)
1	Unwrapped 250	250	5	5
2	Unwrapped 250	250	10	10
3	Unwrapped 250	250	15	15
4	Unwrapped 250	250	20	20
5	Unwrapped 250	250	15	10
6	Wrapped 250	250	20	20
7	Wrapped 270	270	7	15
8	Wrapped 270	270	10	25
9	Liquids 250	250	15	0
10	Liquids 250	250	15	0
11	Liquids 250	250	15	0
12	Liquids 250	250	15	0
13	B&D Test	274	3.5	2
14	Leak Test			

(16) Program Key - 4th Floor

Autoclave Estimated Times

Cycle	Total Time
01 - Unwrapped	10 min
02 - Unwrapped	30 min
06 - Wrapped	45 min
09 - Liquids	1h 20 min
10 - Liquids	1h 10 min

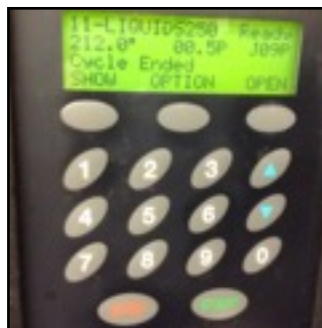
(16) Program Key - 4th Floor

6. Start a timer or note the start time so that you can be sure to retrieve the items shortly after autoclaving finishes.

NOTE: DO NOT leave anything in the autoclave OVERNIGHT!!!

7. When the cycle ends, this will be indicated on the screen by the message “Cycle Ended” (Fig #17). Follow the procedure in Step 4 to open the door, retrieve the items, and bring them back to the lab.

NOTE: Be sure to close the autoclave door completely before leaving (Fig #18).



(17) Cycle Ended Screen



(18) Close Door Sign

Caution: Before autoclaving anything, make sure the material is autoclave-safe!

If you are sterilizing **LIQUIDS in a container with a lid (bottles & tubes) or covered with aluminum foil (glass flasks & beakers)**, follow the steps here:

1. Prepare the items for sterilization:

glass bottles with caps containing liquid media, water or buffers - liquid filled no more than container fill-line, capped with cap slightly-loosened one turn to prevent loss, autoclave tape on top of lid.

glass bottles with caps containing liquid media with agar - liquid filled no more than HALF-VOLUME of the container, capped with cap slightly-loosened one turn to prevent loss, autoclave tape on top of lid.

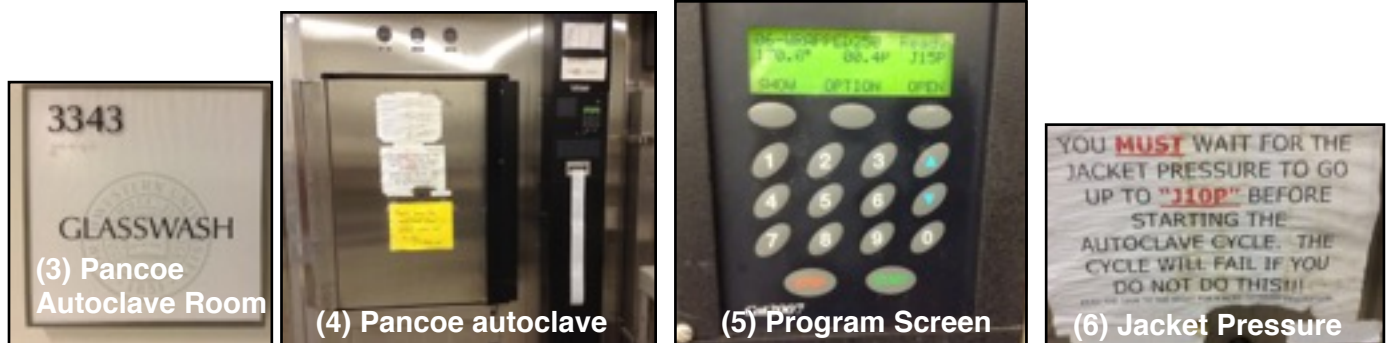
glass test tubes with caps containing liquid media, liquid media with agar, water or buffers - liquid filled no more than 2/3 full, capped firmly, put in labeled rack, autoclave tape on front of rack.

glass or plastic containers with NO LIDS containing liquid media, liquid media with agar, water or buffers (flasks, beakers, pitchers, etc.) - liquid filled no more than HALF-VOLUME of the container, covered in heavy-duty aluminum foil with no holes and aluminum hanging at least 1 inch over the edge of the opening, autoclave tape on the side of the foil cover overlapping on the glass to secure the foil.

2. Place all items in autoclave bins - make sure there is enough space between each item inside a bin (Fig #2). Locate and bring with you the pair of orange autoclave gloves.

3. Use the large grey cart to bring the bins to the autoclave room in Pancoe 3343 (same as the plate pouring room, Fig #3).

NOTE: if this autoclave is busy, you can also use the Pancoe 4th floor autoclave. It is exactly the same type - USE THE AUTOCLAVE ON THE RIGHT SIDE in Pancoe 4343 for both wet and dry cycles and ONLY use the LEFT-SIDE autoclave for dry cycles (the liquid cycle is too long)



The autoclaves should always be left ON - BUT - sometimes they will be OFF when you are ready to use them. If the autoclave is OFF:

- Check on the panel that the Tab indicates 'FIXED' and not 'BROKEN'. If OK, proceed - otherwise try a different autoclave (Fig #4).
- TURN ON the autoclave simply by pushing the 'ON' button above the keypad and Program screen (Fig #5). Make sure the door is closed. You will hear the steam start to run inside the jacket.
- BEFORE STARTING A CYCLE, make sure the **Jacket pressure reads higher than J10P** on the program screen (or also look at the pressure gauge above the door) (Fig #6). *This may take a little longer to start up, but you can proceed to load the autoclave chamber during this time.*

4. If the autoclave is free - proceed to loading:

- Press 'OPEN' on the screen. STEP BACK and wait a minute for the autoclave to automatically open - you will hear the door unlock. PUT ON THE AUTOCLAVE GLOVES!!

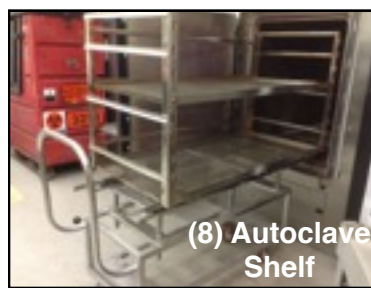
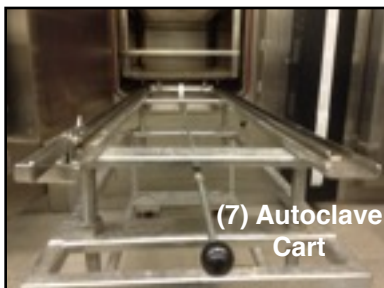
CAUTION!! *Step back away from the edges of the door to avoid getting burned by the steam, which may immediately vent.*

- From behind the door, open the door fully and then position the autoclave cart in line with the rails inside the autoclave to unload the shelf (Fig #7).

NEVER!! *load the autoclave without using the cart to support the shelf.*

Be very careful to not touch anything without gloves on, since all surfaces are HOT!!

- Wearing gloves, pull out the shelf completely to access it from the side (Fig #8).



NOTE: If another lab's items are in the autoclave you can take the bins out and put them on any open peripheral bench tops in the room. **If the contents of the containers are AGAR Media, make sure to notify the owner lab that you removed these from the autoclave.** Any other items (liquid without agar and dry items) are OK to leave on the bench top.

- Load your bins onto the shelf. Generally, it is better to put the heavier things on the bottom (Fig #9).
- **ADD 1 inch deep of tap water to the bottom of the bins when autoclaving liquids. Use the designated plastic beakers in the autoclave rooms to get water.**
- Push the shelf back into the autoclave completely (Fig #10).
- To disengage the cart from attaching to the autoclave, **push forward** on the black knob of the cart release handle (Fig #11); this will lift the gripping mechanism (Fig #12).
- Move the cart back away and close the door. Close the door gently but quickly enough that it is completely closed before the auto-lock engages. You should hear the door lock.

5. Check that the jacket pressure is greater than 10 psi. Select your cycle according to the posted key. The numbers we use are **#10 for Liquid Media with agar** (45 min. sterilization - 1.5 hours Total Cycle) and **#11 for all other Liquids** (30 min. sterilization - 1 hour Total Cycle). PRESS START to activate the cycle. The cycle parameters will print out on the paper below the screen. Reference this for start and stop times, if needed.

NOTE: ALWAYS monitor the screen for a minute until the cycle starts completely before leaving to ensure that the autoclave is operating properly. If the pressure starts increasing normally and no steam is leaking from the door edges, then everything should be fine. **IF THIS IS NOT THE CASE, then press the 'STOP' button to abort the cycle and come get Sarah or Robyn.**

Cycle	Temperature (°F)	Sterilization time (min.)	Exhaust time (min.)
01 - Unwrapped	250	30	5
02 - Unwrapped	250	45	15
03 - Unwrapped	250	20	10
04 - Unwrapped	250	30	15
05 - Wrapped	251	15	15
06 - Wrapped	251	30	15
07 - Wrapped	251	30	15
08 - Wrapped	252	5	5
09 - Liquid	260	35	0
10 - Liquid	255	45	0
11 - Liquid	250	30	0
12 - Liquid	252	35	0
13 - B&D Test	274	3.5	2
14 - Leak Test	???	???	???

(13) Program Key - 3rd Floor



(14) Keypad



(15) Cycle Progress Log Printout

4th floor ENB - VANCE
Autoclave Programs

Program	Program Name	Temp (°F)	Sterilization time (min.)	Drying time (min.)
1	Unwrapped 250	250	5	5
2	Unwrapped 250	250	10	10
3	Unwrapped 250	250	15	15
4	Unwrapped 250	250	20	20
5	Unwrapped 250	250	25	25
6	Wrapped 250	250	30	30
7	Wrapped 270	270	7	15
8	Wrapped 270	270	10	25
9	Liquids 250	250	35	0
10	Liquids 250	250	45	0
11	iso T	185	30	0
12	Liquids 250	250	15	0
13	B&D Test	274	3.5	2
14	Leak Test			

(16) Program Key - 4th Floor

Autoclave Estimated Times

Cycle	Total Time
01 - Unwrapped	10min
02 - Unwrapped	30min
06 - wrapped	45min
09 - Liquids	1h 40min
10 - Liquids	1h 10min

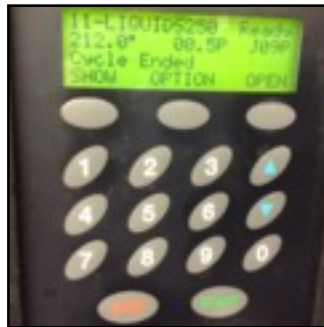
(16) Program Key - 4th Floor

6. Start a timer or note the start time so that you can be sure to retrieve the items shortly after autoclaving finishes.

NOTE: DO NOT leave anything in the autoclave OVERNIGHT!!!

7. When the cycle ends, this will be indicated on the screen by the message “Cycle Ended” (Fig #17). Follow the procedure in Step 4 to open the door, retrieve the items, and bring them back to the lab.

NOTE: Be sure to close the autoclave door completely before leaving (Fig #18).



(17) Cycle Ended Screen



(18) Close Door Sign