

Please report all accidents and near misses. Things happen and it is important to learn from others. Environmental Health and Safety and your colleagues appreciate hearing about incidents, reviewing their causes, and learning from them.

BUNSEN BURNER SAFETY

Bunsen burners are used in the laboratory to create a sterile environment around the vicinity of the open flame. The flame is used for heating and sterilization of materials such as an inoculation loop for streaking bacteria on an agar plate. Bunsen burners are potential fire hazards due to the open flame and should be used with caution.

What Happened?

Recent incidents involving the spreading process of bacteria/yeast on agar plates have led to fires inside the laboratory. Two separate incidents occurred within days while utilizing the “Spread-plating with a turntable and glass rod” method (“Method A”). In the spread-plate process, a spreader is dipped in ethanol from a jar, flamed over a Bunsen burner, and then used to spread the culture on the agar plate.

In the first incident, flaming ethanol was allowed to drip into a biohazard box located nearby on the floor thus igniting the box on fire. The lab used a fire extinguisher and pulled the building fire alarm. The New Haven Fire Department, Yale Fire Code Compliance, Yale Police, and Yale EHS responded to the incident. Yale custodial staff performed cleanup of the floor. Research staff cleaned lab benches. The lab manager highlighted safe work practices at the next lab meeting.

In the second incident, a Bunsen burner on a bench top was placed too far under the center bench shelf. The light fixture under the shelf caught fire and melted the plastic, which dripped into the beaker of ethanol, which then caught fire. Lab personnel used a fire extinguisher to put the fire out. Fire Code Compliance was on site when EHS arrived. Lab personnel had cleaned up most of fire extinguisher dust on bench top. An electrician was contacted to confirm that the electrical outlets under the bench were not damaged. Yale custodial staff used a HEPA vacuum for any remaining dust.



What Went Right?

- Lab personnel knew how to use a fire extinguisher to put the fire out.
- The incident was reported, and an investigation followed.

What Should Have Been Done Differently?

- All flammable materials such as the biohazard box and excess chemicals in the vicinity should have been removed.
- The Bunsen burner should have been placed away from any overhead shelving and light fixtures.
- It is recommended that 70% ethanol (not 100%) be used for spread-plating.

What Corrective Actions Have Been Taken?

Lab personnel were retrained on how to work safely when using a Bunsen burner.

How Can Incidents Like This Be Prevented?

To avoid any future fires in laboratories due to the incorrect performance of this technique, EHS recommends the following modifications:

- Discuss safe performance of this technique during your next lab meeting.
- Ensure researchers are trained and aware of potential risks when working with a Bunsen burner.
- Consider the use of a touch-activated Bunsen burner.
- Consider fire extinguisher training for all lab members and familiarize yourself with the nearest location of a fire extinguisher.
- Consider substituting the glass bead method (“Method B” listed on pg. 2) for the spread-plate method to avoid flaming items that can drip and cause fires (glass beads can be purchased through common lab supply vendors such as Fisher Scientific, Zymo Research, etc.).

Suggestions for Working in Lab Areas with a Bunsen Burner

- Never leave a Bunsen burner, or any open flame, unattended. This generally means the burner is within sight and within easy reach.
- Stay near the lighted burner. If you must leave the area, turn off the flame.
- Before igniting the flame, check to ensure there are no flammable items above or near the burner or items which will be damaged by the heat.
- For streak plating techniques, do not use 100% ethanol because it is more volatile. Instead, use 70% ethanol, which has a lower flash point.
- Shut off gas valve when work with a Bunsen burner is completed.

More Information

- **Reference Article: ‘Aseptic Laboratory Techniques: Plating Methods’ by Erin R. Sanders-**<https://bit.ly/3P0jW3B>
Page 3, Section #4 includes:
Method A: Spread-plating with a turntable and glass or metal rod
Method B: Spread-plating with glass beads: the "Copacabana Method"
- **Corresponding Video by Erin R. Sanders-** <https://bit.ly/3F18Lho>
Section #5: Spread-plating with a turntable and glass or metal rod
Section #6: Spread-plating with glass beads: the "Copacabana Method"
- **LSU EHS on Bunsen Burner Safety-** <https://bit.ly/3LU42pg>
- **Cornell EHS on Bunsen Burner Safety-** <https://bit.ly/38Y68G4>
- **Yale University Fire Extinguisher Training (Office of the Fire Marshal)-** <https://bit.ly/3ypoPxi>

“Method A”

<https://bit.ly/3FrvVTw>

Spread Plate Technique
Principle, Procedure and Uses

The diagram illustrates the spread plate technique. On the left, a serial dilution series is shown with a flask of concentration 'C' and six test tubes with concentrations $10^1 C$, $10^2 C$, $10^3 C$, $10^4 C$, $10^5 C$, and $10^6 C$. A 10 mL transfer is indicated between tubes. Below this, a petri dish is shown with the formula: $\text{count colonies} = N \times \text{each colony given from a single cell}$. On the right, a vertical sequence of three petri dishes shows the plating process: 1. Inoculate plate containing solid medium with 0.1 mL. 2. Spread inoculum over surface evenly. 3. Colonies grow only on surface of medium. The final step is labeled 'Spread Plate Technique'.

“Method B” (from Zymo Research)

The diagram illustrates Method B, the Copacabana Method, for spread plating using beads. It consists of four numbered steps: 1. Shake Beads to Spread Cells. 2. Invert Plate. 3. Pour off Rattler™ Beads, Autoclave, and Reuse. 4. The final step shows the beads being collected in a beaker. The diagram is enclosed in a green rounded rectangle.