

STERILE TECHNIQUES

The requirements for sterile technique depend on the experimental situation including the growth media used, the experimental organism, the duration of the experiment and the intended use of the culture. For these experiments the most serious contaminant is mold. Many common molds grow well on yeast media and compete effectively to over-grow the plates and obscuring the yeasts that do manage to grow. Bacteria are less problematic since most do not grow well on these media. It is advised therefore to store sterilized media at room temperature thereby detecting contamination before use.

1. Sterilizing with moist heat.

Moist heat provided by an autoclave or pressure cooker is an efficient way to sterilize most materials. At a pressure of 15 psi above atmospheric pressure, water reaches a temperature of approximately 121°C before it boils. Most materials are effectively sterilized by 15 minutes of exposure to this temperature. Small batches of media may be sterilized in a pressure cooker if needed.

2. Sterilizing with dry heat

Dry materials such as glass and metal may be sterilized in an oven, but this requires temperatures of 160°C and longer time of 2 hours.

3. Sterilization by Flaming

The flame from a gas burner will effectively sterilize items such as metal inoculating loops or glass spreaders. Dip the items in alcohol and then ignite by flaming.

4. Maintaining Sterility

The most common sources of contamination during experimentation are dust in the air and on bench surfaces. Keep sterile items covered be careful to re-sterilize if necessary. Maintain a suitable area for media prep and storage.