

General Autoclave Procedures

Important Notice: Please refer to your specific instrument instructions for the best temperature and/ or pressure and/ or time settings that will meet the desired end results for your specific application.

Sterilization techniques are critical when working with any microorganisms in the laboratory. Proper sterilization of all materials before and after the experimentation is one of the keys to a safe and successful experiment.

Sterilization refers to any process that removes, kills, or deactivates all forms of life (in particular referring to microorganisms such as fungi, bacteria, spores, unicellular eukaryotic organisms such as Plasmodium, etc.) and other biological agents like prions present in a specific surface, object or fluid, for example food or biological culture media. (WHO Glossary). Achieving this task is much more difficult than is commonly realized. It is difficult to guarantee sterility, and even the most severe methods often fail. However, for practical purposes sterilization can generally be achieved using dry heat, steam, incineration, gamma radiation, or severe chemicals. The most commonly used chemicals are the gases of formaldehyde or ethylene oxide and the liquids formalin, glutaraldehyde, hypochlorite and chlorite solutions. In all cases, the key to success is good penetration by the sterilizing agent for a sufficient time. This is particularly important for large objects where the center may be unpenetrated even after a long period of time.

Proper autoclave treatment will inactivate all resistant bacterial spores in addition to fungi, bacteria, and viruses, but is not expected to eliminate all prions, which vary in their resistance. This document is not meant to assist in prion elimination. Please refer to the current research literature and protocols related to prions and sterilization.

Steam Sterilization Defined

Steam sterilization is carried out in an autoclave. The pressurized steam produced has a high latent heat. This intense heat leads to hydrolysis and coagulation of proteins, which kills microbes, spores, and viruses. Steam sterilization typically involves exposing an item to steam at a temperature of 121 °C (250 °F) and a pressure of 100 kPa (15 psi) for 15 to 30 minutes.

Compared with dry heat sterilization, steam sterilization is more efficient because the moisture in steam is a good conductor of heat and is superior at penetrating the load. However, the steam must be able to penetrate. Even with pressure, steam sterilization is often not able to sterilize the inside of a sealed container. Steam and pressure is a widely accepted method for items that can accept heat, moisture and pressure.

Dry Heat Sterilization Defined

In contrast to steam sterilization, dry heat sterilization, does not involve water. Dry heat sterilization typically involves exposing an item to a temperature of 170°C under normal air pressure for one to two hours. The longer duration ensures that even the most resistant spores are inactivated due to protein denaturation.

Certain situations call specifically for dry heat sterilization. For example, dry heat is required for hydrophobic items, such as fats and oils; items that will be damaged by moisture, such as powders; and instruments that may become corroded.

To sterilize toothpicks using the Steam Sterilization Method

Moist heat provided by an autoclave is an efficient way to sterilize most materials. To sterilize toothpicks, place the desired quantity in a beaker or Pyrex dish, cover with foil, and autoclave. At a pressure of 15 psi above atmospheric pressure, water reaches a temperature of approximately 121 °C before it boils. Most unsealed materials are sterilized after 15 to 20 minutes exposure to this temperature and pressure. However, your application may need a higher temperature and/or pressure and/or time.

Important Notice: Please ensure that your autoclave is operating within specification and refer to your specific autoclave instructions for the best item preparation, pressure, temperature, and time settings that will meet the desired end results for your specific application.

To sterilize toothpicks using the Dry Heat Method

Dry heat requires the use of a temperature-regulated oven. It is limited, however, to materials that will not degrade at the required high temperatures. Plastic and wood items generally are not sterilized using dry heat. Items sterilized using dry heat are often placed in a pre-heated oven at 160 °C (320 °F) and baked for at least 2 hours. However, your application may need a higher temperature and/or time.

Important Notice: Please ensure that your heat sterilizer is operating within specification and refer to its specific instructions for the best item preparation, and temperature and time settings that will meet the desired end results for your specific application.

To sterilize objects made of polypropylene using the Steam Sterilization Method

To sterilize polypropylene objects, such as the smartSpatula, place the desired quantity in a beaker or Pyrex dish, cover with foil, and autoclave. 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 to 20 minutes exposure to this temperature and pressure. However, your application may need a higher temperature and/or pressure and/or time.

Important Notice: Please ensure that your autoclave is operating within specification and refer to your specific autoclave instructions for the best item preparation, pressure, temperature, and time settings that will meet the desired end results for your specific application.

Disclaimer:

The above directions are based upon commonly used techniques for sterilization of laboratory consumables. Every application requiring sterility is different and care should be taken to understand the level of the desired sterility and the techniques that would apply.

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