



Sterilization Methods

Sterilization is the killing or removal of all microorganisms, including bacterial spores, which are highly resistant. Sterilization refers to the antimicrobial process during which all microorganisms are killed or eliminated in or on a substance or substrate by applying different processes.

Sterilization procedures involve the use of heat, radiation, chemicals, or "physical removal" of microbes (Table. 1). The type of sterilization should always be chosen as required, by taking into consideration the quality of materials and tools used and the possible adverse effects of sterilization on them.

Heat	Flaming Incineration Boiling Moist heat Dry heat
Radiation	UV Gamma rays Electron beam X-rays
Chemicals	Ethylene oxide Ozone
Plasma	Ionizing gas
Filtration	Membrane

Disinfection methods are different from sterilization as they kill many but not all microorganisms.





Sterilization by Heat

Heat is the most effective and rapid method of sterilization and disinfection. Excessive heat acts by coagulating cell proteins, whereas less heat interferes with metabolic reactions. Sterilization occurs by heating above 100°C, which ensures lolling of bacterial spores. Sterilization by hot air in a hot air oven (i.e., dry heat) and sterilization by autoclaving (i.e., moist heat) are the two most common methods used and will be described in the following. In particular, when sterilization occurs by heat, two types of techniques can be identified and will be briefly illustrated here:

- 1. Sterilization by dry heat;
- 2. Sterilization by moist heat.

1. Dry Heat

The use of dry heat is based on the removal of the water content of microbes and subsequent oxidation, thus killing or removing all microorganisms, including bacterial spores. Dry heat sterilization technique requires longer exposure time (1.5-3 h) and higher temperatures than moist heat sterilization

Dry heat does most of the damage by oxidizing molecules. The essential cell constituents are destroyed and the organism dies. The temperature is maintained for almost an hour to kill the most resistant spores. Advantages and disadvantages of dry heat oven sterilization can be shown in (Table 2).





Advantages	Disadvantages
A dry heat cabinet is easy to install and has relatively low operating costs	Time-consuming method because of slow rate of heat penetration and microbial killing
It penetrates materials	High temperatures are not suitable for most materials
It is nontoxic and does not harm the environment	
It is noncorrosive for metal and sharp instruments	

The most common time-temperature relationships for sterilization with hot air sterilizers are the following:

- 170°C for 30min;
- 160°C for 60min;
- -150° C for 150min or longer depending on the volume

2. Moist Heat (Autoclave)

Due to the fact that the heat conductivity of water is several times higher than that of the air, heat sterilizes more quickly and effectively in the presence of hot water or steam than dry heat. Moist heat acts by denaturation and coagulation of protein, breakage of DNA strands, and loss of functional integrity of cell membrane; in addition, the structural components for microorganism replication are destroyed.





Sterilization by moist heat is one of the most used sterilization techniques because it is simple, low cost, and devoid of any risk for the operators. (Table 3) represent the advantages and disadvantages of autoclave sterilization

Advantages	Disadvantages
Nontoxic to patient, staff, environment	Deleterious for heat- sensitive or moist-sensitive instruments/materials
Cycle easy to control and monitor	Microsurgical instruments damaged by repeated exposure
Rapidly and efficacy microbicidal	May leave instruments wet, causing them to rust (in case of metallic devices)
Least affected by organic/ inorganic soils among sterilization processes listed	Potential for burns
Rapid cycle time	
Penetrates medical packing, device lumens	

However, the time used for sterilization depends on the size and content of the load. Autoclave is a metallic cylindrical vessel (Fig. 1); on the lid, there is a:

- Gauge for indicating the pressure,
- Safety valve, which can be set to blow off at any desired pressure,
- Stopcock to release the pressure. It is provided with a perforated diaphragm.







Water is placed below the diaphragm and heated by electricity, gas, or stove. Typical sterilization process is composed by three phases (Fig .2): heating phase, stationary phase (t=15-30min), and cooling phase.









Sterilization by Ethylene Oxide

The materials used in chemical sterilization are liquids or gases. Liquid agents are used primarily for surface sterilization. Among sterilizing gases, those working at low temperature function by exposing the materials to be sterilized to high concentrations of very reactive gases (e.g., ethylene oxide (EtO), beta propiolactone, or formaldehyde).

Due to their alkylating effect, these compounds cause the death of microbes by damaging their proteins and nucleic acids. The chemical agents used for sterilization must be chemically compatible with the substances to be sterilized. The advantages and disadvantages of ethylene oxide sterilization represented in (Table 5).

Advantages	Disadvantages
Low temperature	Excessively long cycle
Short aeration time	Safety concerns— carcinogenic to humans
High efficiency: it destroys microorganisms including resistant spores	Toxicity issues: toxic residues on surgical instruments and tubing
Large sterilizing volume/ chamber capacity	EtO is flammable
Noncorrosive to: plastic, metal, and rubber materials	Requires special room conditions, safety equipment, and separate ventilation system

There are at least three stages in a typical EtO sterilization cycle (Fig. 3):





- 1. **Preconditioning:** This step prepares the chamber environment to meet the ideal conditions for temperature, pressure, and humidity. First, air is removed from the chamber to allow for gas penetration; then, steam is injected into the chamber, which humidifies the load as EtO is only effective in a humid environment. The chamber is heated by either steam or hot water, which is present in the jacket of the sterilizer.
- 2. **Sterilization:** This step is the actual sterilization process. The EtO enters the chamber via evaporation with a certain amount of steam to keep the humidity level up as well as to make sure the EtO is reaching all parts of the load.
- 3. Aeration (degassing): It is the most important and longest part of the EtO sterilization cycle. In fact, materials such as plastics and rubbers absorb gas and, if applied to patients, the toxic gas could damage human body tissue. For this reason, it is very important to have an excessive aeration stage to remove any remaining EtO gas and to allow absorbed gas to evaporate again from the sterilized items.

