

CO2 incubator

Experiments that imitate processes in living organisms are called in vitro. In these, cells and tissue cultures are grown in the laboratory, often over several weeks, in an environment that is as natural as possible.

Cultivation takes place in a **CO2 incubator**, in which not only the temperature, but also the humidity and carbon dioxide content, must be controllable. In addition to this, the oxygen and nitrogen content can also be adjusted in some appliances.

The **CO2 incubator** usually has a chamber volume of between 50 and 300 litres and a temperature range from +4 °C and +8 °C above room temperature to +60 °C (details on the basic operation and features of a **temperature control chamber**). The interiors of **CO2 incubators** are normally made of corrosion-free **stainless steel 1.4301** (acc. to standard ASTM 304) and they are additionally smoothed with electrolytic polishing by some manufacturers to prevent germs from settling and to make cleaning easier. In order to observe the chamber load without affecting the atmosphere inside the chamber, **CO2 incubators** are usually equipped with an inner glass door or additional gas baffle.

Regulating air humidity

Normal temperatures in a **CO2 incubator** lie at around the same temperature as the human body (37 °C). The essential matter is maintaining humidity in a controlled manner, while at the same time avoiding condensation in the interior. This is done in practice either through humidification via water trays in the interior of the **CO2 incubator**, via direct contact of water with the floor or through an activity controllable system for humidification and dehumidification in which the water supply lies outside the working chamber. The water from the external water tank is heated up in a vaporator and fed into the chamber as steam, which means that only sterile water (pyrolytic germ barrier) humidifies the chamber and the samples and the humidity can be applied in specific doses.



CO2 incubator with inner glass door

The active control of humidity guarantees that the required humidity is reached quickly after the door is opened and closed (see Chapter [DIN 12880:2007-05](#)).

Heating the chamber

Precisely maintaining the atmosphere is absolutely essential in a **CO₂ incubator** to protect samples. This is why there is a whole range of different heating systems in practice that are supported in part by motorised fans, and in part by air circulation systems free of turbulence.

Many **CO₂ incubators** are supported by a heated air jacket, and in some cases by a water jacket, ensuring even **temperature distribution** and temperature stability, rapid **recovery times** after the door has been opened and maintenance of the temperature after a power failure. At the same time, the air jacket prevents **condensation** from forming. Some manufacturers, who do without a jacket of air or water, instead heat the chamber directly from all six sides, surrounding it in addition with an insulation coat. In this form of direct heating, a high degree of flexibility and a high density in the arrangement of heating elements form the basis of temperature homogeneity.

Sterilisation and decontamination

Prior to each new load, a **CO₂ incubator** must be cleaned to avoid cross-contamination of the chamber load. In addition to cleaning with disinfection agents, appliance manufacturers have developed various technologies, some of which can be combined.

Some appliances allow sterilisation of the interior with UV light, others provide permanent air filtering. Air filters such as HEPA filters require additional resources of time and money, since they need to be replaced.

Some manufacturers provide a decontamination routine using damp heat at 90 to 95 °C, and which takes many hours, others decontaminate for several hours at 120 °C. Hot-air sterilisation between 160 °C and 180 °C is quite common and recognised as a safe procedure. The German pharmacopeia 10 (DAB 10), a collection of recognized



HEPA filter in the working chamber of a CO₂ incubator

pharmaceutical rules concerning quality, testing, storage, dispensing and labelling of drugs and the substances of legally binding character that are used in their production, stipulates the following reference values for the duration of sterilisation:

Hot air at 180 °C: At least 30 minutes

Hot air at 170 °C: At least 1 hour

Hot air at 160 °C: At least 2 hours

A further quality criterion for modern **CO2 incubators** is the option of decontaminating or sterilizing the interior, including fittings and sensors.

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