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CO_2 Incubators – Best Practices for Set-up and Care

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Executive Summary

CO_2 incubators are required to maintain an optimal environment for cell growth, by providing carbon dioxide control in a humidified atmosphere with constant temperature. In this guide we give you some best practices and tips, ranging from installation and daily operation to the maintenance required to keep a contamination-free environment for reliable cell growth.

Setting up a new incubator

When the decision for a certain incubator model has been made, the next step is installing the device in the cell culture lab. For gassed incubators a risk assesment should be performed. A recommended measure are gas detectors that issue an alert when critical gas concentration is reached in the laboratory. In addition, a ventilation system ensures air exchange both during normal operation and in case a critical gas concentration is reached. When gas cylinders are used they should be clearly labeled and securley anchored in suitable safety cabinets. The tubing connecting the incubator to the gas cylinder or the central gas supply should be appropiate for the pressure of the gas used to avoid any leakage of CO_2 which can cause suffocation if the concentration in the air is too high. This guide will help you with general tips on installation, set-up, and care of your new CO_2 incubator.

Tips on installation and initial set-up*

> Avoid placing your incubator in direct sunlight, or close to vents, air-conditioning ducts or the exhaust of heat- or cold-generating equipment, as these can interfere with chamber conditions. Follow manufacturer's specifications on allowable room temperature to facilitate stable incubation at 37 °C.

> Do not place your incubator directly on the floor. Use abase with casters, which offers not only the possibilities of flexible movement and improved access to the back side for cleaning and service, but also keeps the unit away from dust and dirt on the floor that can enter when opening the door.

> Position the incubator to allow clearance for opening the door, access to the CO_2 sampling port (if an external gas analyzer is used to measure gas concentration), and access to any other port.

> Gas connections: Gas connection set-up depends on the manufacturer, so follow instructions in the operating manual. We recommend gas quality of 'high grade' (>99.5 %) for gas supplies. In some regulated fields, medical grade gas is required.

> Initially clean and disinfect the incubator interior and shelves, and other chamber equipment. Install all internal components and make sure your incubator is level, with the help of a small water level placed on the second shelf of the incubator. Level the incubator by adjusting the feet or the base of stacking stand, according to the manufacturer's instructions. Don't forget to lock the leveling feet in place by tightening the locking nuts on each foot!

> Run the automatic self-sterilization program, if your incubator is equipped with one.

> Fill the water tray with warm sterile distilled water, adjust the program set-points if required, and leave the incubator running for at least two hours (preferably overnight) to allow conditions to stabilize. Your incubator is now ready for use!

*Please note that following tips are general tips and do not replace reading the user manual [8] when installing a new unit in the lab.

Proper handling and cleaning and maintenance schedules

> To keep the risk of introducing contamination into the incubator as low as possible, only touch the incubator with fresh or disinfected gloves.

> Keep your incubator contents organized to easily relocate cells and to avoid long and frequent door openings. This reduces the risk of air-borne microorganisms to enter the incubator chamber. Depending on the routine in your lab there are different ways to organize the incubator contents (Fig. 1).

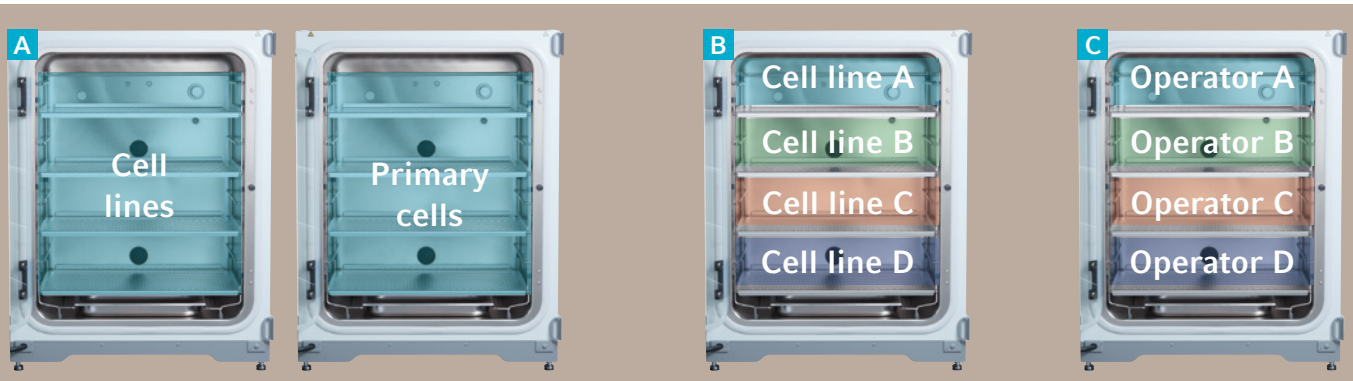

Figure 1: Organize your incubator. Depending on the routine in the laboratory there are different ways to organize incubator contents to reduce the risk of contamination (**A**) Dedicated incubators for keeping cell lines and primary cells separately, (**B**) Dedicated shelves for different cell lines, (**C**) Dedicated shelves for different operators

> A regular cleaning schedule for your incubator prevents contaminations within the environment for your cells. We offer suggestions below, but please decide on frequencies according to your own risk management policies, as they depend on multiple factors, including the number of users, their aseptic skills, and the probability that the cells are contaminated.

Daily: Inspect incubator contents! Remove and disinfect any spills immediately with 70 % Ethanol or Isopropanol. Prefer wipe to spray disinfection. This prevents the formation of aerosols which may be harmful to you and your cells, and enables complete wetting of the surface for proper disinfection.

Weekly: Replace water in water tray, and clean and wipe disinfect the tray using alcohol 70 %. Most suppliers recommend sterile distilled water.

Monthly: Once a month, up to every 6–8 weeks, empty the incubator fully. Using a lint free cloth, clean the chamber interior with soapy water and rinse with water, followed by wiping the surfaces with alcohol 70 % or an equivalent non-corrosive disinfectant. If you have an incubator with many hidden corners, fissures, ducts, or seams, you should pay special attention to these areas, as germs can hide there.

Clean and disinfect the removed shelves and racking similarly. Clean, as well, the exterior of the incubator, especially the surfaces you touch, like the doors. Take care to keep the solutions from coming into contact with any mains electrical outlets or assemblies. If your incubator is equipped with an automatic disinfection program, first reinstall all parts that can withstand the disinfection program. Check if all sensors can stay inside. Keep the HEPA filter out if the unit is equipped with one. Then run the disinfection program overnight, following the manufacturer's instructions.

Every 6 months: Replace the HEPA filter if your unit is equipped with one.

Annually: Service should be done at least once a year by an authorized service engineer. Suppliers offer flexible service performance plans and contracts according to your needs, from basic checks of sensors and functional parts up to replacement of worn parts.
(For Eppendorf Service packages please visit www.eppendorf.com/incubator-service)

CellXpert® C170i Ordering Information

Device Options			Order no.				
Door Handle	Humidity Monitoring/ Water Level Monitoring	Copper	230 V, 50/60 Hz European	230 V, 50/60 Hz UK/HKG	230 V, 50/60 Hz Australia	230 V, 50/60 Hz China	100–120 V, 50/60 Hz USA/Japan
Right			6731 000.011*	6731 000.012*	6731 000.013*	6731 000.014*	6731 010.015*
Right		Yes	6731 000.511	6731 000.512	6731 000.513	6731 000.514	6731 010.515
Right	Humidity monitoring		6731 000.111*	6731 000.112*	6731 000.113*	6731 000.114*	6731 010.115*
Right	Water level monitoring		6731 000.211*	6731 000.212*	6731 000.213*	6731 000.214*	6731 010.215*
Right	Both		6731 000.311*	6731 000.312*	6731 000.313*	6731 000.314*	6731 010.315*
Left			6731 000.021*	6731 000.022*	6731 000.023*	6731 000.024*	6731 010.025*
Left		Yes	6731 000.521	6731 000.522	6731 000.523	6731 000.524	6731 010.525
Left	Humidity monitoring		6731 000.121	6731 000.122	6731 000.123	6731 000.124	6731 010.125
Left	Water level monitoring		6731 000.221	6731 000.222	6731 000.223	6731 000.224	6731 010.225
Left	Both		6731 000.321	6731 000.322	6731 000.323	6731 000.324	6731 010.325

*Stock article; all others are built-to-order

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Device Options	Order no.				
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Right	6734 000.011	6734 000.012	6734 000.013	6734 000.014	6734 010.015

References

[1] Operating Manual Eppendorf CO_2 incubators. www.eppendorf.com

About Eppendorf

Eppendorf is a leading life science company that develops and sells instruments, consumables, and services for liquid-, sample-, and cell handling in laboratories worldwide. Its product range includes pipettes and automated pipetting systems, dispensers, centrifuges, mixers, spectrometers, and DNA amplification equipment as well as ultra-low temperature freezers, fermentors, bioreactors, CO_2 incubators, shakers, and cell manipulation systems. Associated consumables like pipette tips, test tubes, microtiter plates, and disposable bioreactors complement the instruments for highest quality workflow solutions.

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