

CELL CULTURE RULES

1. Operator must notify to Cell Culture Staff the intention to work in cell culture room in order to receive specific instructions and to have assigned spaces (incubator & +4°C fridge).
2. Gloves and a dedicated lab coat must be used at any time. Protective glasses are advisable during particular work phases.
3. HOODS are only intended for sterile work
Access: sign IN and OUT.
Connection: you'll find specifics on each hood.
Once the alarm silenced, leave the cabinet to run under ventilation range for 10-15 minutes.

The basic principle of the flow hood is the airflow through the perforations in working surface. Thus, do not overload it and avoid to obstruct the perforations with paper or unnecessary items. Operation should be possibly carried out in the central part of the work surface and in any case never close to the front air regain grid. The minimum safety limit is about 10 cm from the front window.

Keep the hood as clear as possible and clean it (with ethanol 70%) after each use. Switch off and **close it after 6 p.m.**

Disconnection:

- I. Disconnect vacuum system
- II. Do not leave personal stuff inside the hood (1-2 boxes of tips, 1-2 boxes Pasteur pipettes are allowed).
- III. Close the hood.
- IV. Turn on UV lamp.

4. INCUBATORS: Each incubator is assigned to 3-4 operators (whose names are posted on the front door).

Parameters (CO₂ & T) can be changed only by maintenance staff, after having informed Cell Culture Staff. Each operator is responsible for cleaning of his/her own space.

Incubator water pans should be checked daily and refilled when necessary with sterile dH₂O (RX). In IFOM is also available a non toxic germicide to keep cell incubator trays free of bacterial and fungal growth.

5. **CLEANING:** At the end of each work session, operator must clean carefully with Pharmacidal or Ethanol 70% all the surfaces. No potentially hazardous materials should remain on the bench or under the hood at any time.

Operators must clean incubators and hoods thoroughly every two months; Cell Culture Staff will remind your turn by e-mail. Cleaning should be done with mild soap, followed by Pharmacidal or Ethanol 70%.

NEVER USE BLEACH: IT IS CORROSIVE!

DEDICATED INCUBATORS & HOODS

Some incubators and hoods are not reserved to operators or group, but to specific uses:

Human Primary Cells (*view link "human primary cells"*), Baculovirus Incubator (T:27°C; no CO₂), Mycoplasma untested cells, Endothelial cells, ES cells, Special conditions incubators (CO₂ & T).

1. All incubators/hoods will be used based on previous booking.
2. Specific requests as far as the incubation conditions are concerned should be indicated to the Cell Culture personnel. Incubator parameters can be changed only by maintenance staff.
3. At the end of usage the incubator and hood should be cleaned and the water changed.
4. In order to maintain good working conditions, the material of unknown origin is going to be eliminated.
5. Since this is a common commodity operators are asked to limit the time to strictly necessary.

-80°C CELL FREEZING SPACES & crioboxes

Operators can find a cell freezing dedicated space in -80°C fridges in each “controcarridoio” (instrument corridor), marked as “Cell freezing reserved”.

Cryoboxes use:

1. Crioboxes are not intended for vial storage, therefore leave cells in them for a maximum of 1 week. Remember that cell viability after thawing decreases as long as you leave cells at -80°C instead of in the Nitrogen tank.
2. Nobody can have personal cryoboxes: you can neither keep them in your own space nor write your name directly on them (use a label, please!).
3. Ask for cryoboxes in Cell Culture room D, by signing in the apposite book.

PLASTICWARE

Common cell culture plastic-ware should be opened under the hood, bags must be sealed and, possibly, marked with the user name. Operator must put opened bags into the Cell culture cabinet, in his own space (look at the label with the incubator number).

WASTE DISPOSAL

The YELLOW CONTAINERS under each hood are intended for Pasteur pipettes, needles, scalpels...Do not use for anything else.

Empty medium bottles with Sekusept are suitable for liquid waste.

VACUUM TRAP: when the trap under your hood is maximum 2/3 full you are asked to empty and rinse it. Before connecting it to the vacuum system Sekusept must be added.

ECOBX: must contain contaminated items (plastic pipettes, flasks, Petri dishes...) but no bags or paper wraps. When the box is full it must be closed, dated and brought outside the room.

WHITE BIN: is only intended for “clean” rubbish (paper, bags, wrap...).

RECYCLABLE PLASTIC: plastic bottles which didn't contain toxic substances and weren't in contact with biological samples can be recycled by rinsing and trashing them in the dedicated white container.