

Tissue culture shared facilities in the NDMRB

Please follow these advisory guidelines:

1. Training
 - The lab manager is responsible for training in the tissue culture lab. New users may not use the facility until they have had their induction.
2. Lab coats - Clean blue only.
 - No white coats allowed in TC vice-versa no blue coats in the main lab.
 - Lab coats should be changed regularly. Return any that are no longer needed back to the collection bin.
3. Hoods
 - Hoods must be booked in advance using the laminated calendars. If you are later than 15 minutes you may lose your slot.
 - Ensure that you clean up any spills (including under the staging) and spray and wipe down the hoods after use. Please do not use virkon to clean the hoods as it is corrosive and will cause damage over time. 70% ethanol is sufficient
4. Aspirators
 - These must be emptied either when they are full or on Friday lab clean. Decontaminate with 1% Virkon (final concentration) for at least 15 minutes before pouring down the sink with plenty of water.
5. Bins
 - Autoclave bins are for all contaminated waste, including gloves whether contaminated or not. Please do not over fill autoclave bags, when full tie with a cable tie and put outside in big autoclave bin (don't leave them for the next users).
 - No liquids or tips in the bins
 - Tips go in sweet jars, also known as dispo jars (when these are full, label as waste and leave them outside, beside the autoclave bins).
 - Black bags are for general waste such as paper and non-contaminated waste.
 - No gloves in black bins.

6. Recycling

- Tip boxes and cardboard go to recycling
- Empty plastic or glass containers including ethanol/media bottles should be rinsed out, label defaced by scribbling or crossing out information.

If they have the recycling triangle they go to the recycling box in the main lab or into general waste if they can't be recycled. See the waste streams on the NDMRB poster if in doubt

7. Fridges

- Label everything in the fridge with your name, or group name (or initials), date opened and expiry date. There is limited space, if bottles of media have expired please throw them out.

8. Incubators

- Each group is responsible for their own incubators – It is recommend that the incubators are cleaned out weekly (please use only aquasan to sanitize water in the incubators) and that you decontaminate your incubators monthly.
- Training can be provided by the lab manager on the high temperature decontamination cycle.

9. Water baths

- Please remember to turn these off in the afternoon *Especially on Fridays*

10. Microscope

- The Zeiss microscope belongs to the Huber group please don't use it unless you have seen Sarah for training.
- Don't change the settings and remember to turn off when you have finished.

11. Contamination

- Occasional bacterial contamination can occur please review your aseptic technique.
- Use the 10% Virkon diluted to 1-2% for decontamination.
Use tablets rather than the powder wherever possible.

If the contamination is widespread you must inform the lab manager.
They will advise on the appropriate decontamination procedure.

12. Mycoplasma testing

- Mycoplasma testing should be done on a regular basis. (monthly is advised for cell lines which are regularly used)
- Be aware that from time to time there will be untested and possibly contaminated cell lines in use.
- If you find you have mycoplasma you must let the lab manager know. They will advise on the appropriate decontaminated or containment procedures.
- Adenovirus, unscreened cells or those with mycoplasma should only be handled in the hood at the far end

13. Spill procedure

- Spills must be cleared away as soon as possible
- Small spills can normally be cleared away using blue roll, dispose of all blue roll used in the autoclave bins

Large Biological spill (e.g. bacterial or tissue culture suspension; blood) at CL1 or 2:

- Sprinkle Virkon powder liberally over spill. Do not add Virkon solution as this will increase the size of the spill. Avoid dust formation in confined areas.
- Cordon off area and leave for at least thirty minutes for all fluid to be absorbed by Virkon and for disinfection to occur.
- If spill involves Class 2 genetically modified micro-organisms or other hazardous microorganisms place in autoclave bag and send to be autoclaved.
- If spill involves bloods place in bag and put in clinical waste (yellow bag) system at appropriate point.
- For all other biological spills place in sink and run to drains.

14. Communal areas

- Racks are specifically for TC. Do not remove.
- Beakers with Virkon waste must be labeled with name date and time. These should be emptied and washed out in a timely manner.
- Make sure the shared work spaces are tidy and if required cleaned down with 70% EtOH.
- Please use the outside cupboards for storage, don't store excessive amounts in the TC lab.

Each group is responsible for their own consumables, but some shared items;

Gloves, paper towels etc. can be found in the TC cupboard.

Ethanol must be stored in a fire cabinet, bottles are labelled tissue culture.

It is advised that a weekly check rota is displayed in the lab and that there is space for sign off when people have completed the weekly checks.

Before *you leave the lab* make sure you have:

- Cleaned the hood and work surfaces.
- Put everything you have used back where it lives.
- Emptied the autoclave bin.
- Empty the aspirator if full.
- Turned off the pump.
- Turned off the microscope.
- Last one out in the afternoon i.e. any time after 4:00pm should also turn off the water bath.
(It only takes a few minutes to warm up if someone needs it later).

Weekly Check:

- Clean all the hoods.
- Wipe the calendar with 70% ethanol.
- The water bath must be emptied on a weekly basis, sprayed down with 70% EtOH and ideally left dry over the weekend.
- Refilled with aquasan (1 tablet in 5-10L of 50% deionised water: 50% tap water) when required.
- Wipe down all surfaces including lintels with 70% EtOH
- Ensure that aspirators are empty and clean.
- Prepare 70% ethanol.
- Throw out the old 10% Virkon and replace with fresh (this will last a week - please date)

This replaces TDI-SOP-020

Associated SOP:

NDMRB-POL-006	Handling, Storage and Disposal of Lab Waste.
NDMRB-SOP-008	Storage of flammable liquids in containers
NDMRB-SOP-007	Handling of biological material
NDMRB-SOP-009	Use of laboratory centrifuges
NDMRB-SOP-040	Good pipetting techniques

Associated COSHH:

Virkon

Trypan blue

<http://www.ndmrbs.ac.uk/coshh-risk-assessment>