

1 Preparation stock solution solid compound(s)

- Before entering the VAS lab prepare the necessary labels at your own department. Denote compound, concentration, date and name (initials) on this label and label your tubes in advance at your own department.
- Switch the bio safety cabinet to full power.
- Clean working area in the bio safety cabinet with 70% ethanol
- Cover the surface area with a under pad and use two tissues on top of this under pad.
- Spray the outside of the tissues with 70% ethanol.
- Try to use a minimal amount of compound and use preferably 1.5 ml eppendorf tubes.

➤ Weighing a compound:

- Put the jar/bottle with your compound on the prepared under pad and tissues
- Place a glass beaker on the scale.
- Place the eppendorf tube in the glass beaker and set the scale to zero (Tare).
- Touch the chemical bottles and eppendorf tubes with your fingers, except your pinky. You can use your pinky or palm for operating the cabinet or balance.
- Take your Eppendorf tube and use it on/above the prepared under pad and tissues.
- Take the required amount of compound and put it in the eppendorf tube using a disposable anti-static (plastic) spatula. It's better to use a plastic spatula.
- Avoid contact with the outer surface of the eppendorf tube when transferring the compound.
- Close the eppendorf tube and weight the amount of your compound.

Change gloves in case you think your gloves are contaminated.

- Clean the outside of the tube with a tissue containing an appropriate solvent.
- Wrap parafilm around the tube and centrifuge the tube that contains the compound.
- Calculate the amount of solvent you need for dissolving the compound.
- Add the solvent to the compound.
- Seal the eppendorf tube with parafilm and vortex until completely dissolved.
- Clean the original compound/carcinogen bottle with an appropriate solvent.
- Clean up the cabinet and dispose tissues and other waste in the small plastic bag (present in the Laminar Flow Cabinet). The plastic bag has to be disposed in the blue biohazard container.
- Take of 1 pair of gloves and leave the weighing facility with your original compound and your work solution in a box.
- Store the solutions in the storage room or use it immediately.

* If you are planning to expose cells with a compound dissolved in DMSO, leave the solution at room temperature 1 hour in advance.

2 Preparation stock solution from liquid compound(s)

- Before entering the VAS lab prepare the necessary labels at your own department. Denote compound, concentration, date and name (initials) on this label and label your tubes in advance at your own department.
- Switch the bio safety cabinet to full power.
- Clean working area in the bio safety cabinet with 70% ethanol
- Cover the surface area with a under pad and use two tissues on top of this under pad.
- Spray the outside of the tissues with 70% ethanol.
- Try to use a minimal amount of compound and use preferably 1,5 ml eppendorf tubes.

- Pipetting
 - Put the jar/bottle with your compound on the prepared under pad and tissues
 - Place a glass beaker on the scale.
 - Place the eppendorf tube in the glass beaker and set the scale to zero (Tare).
 - Touch the chemical bottles and eppendorf tubes with your fingers, except your pinky. You can use your pinky or palm for operating the cabinet or balance.
 - Take your Eppendorf tube and use it on/above the prepared under pad and tissues.
 - Pipet the calculated volume in the tube/ep.
 - ☞ Important: -Take the density of the compound into account!
 - Try to prevent that the dilution volume is bigger as the maximum capacity of your tube.
 - Avoid contact with the outer surface of the eppendorf tube when transferring the compound.
 - Close the eppendorf tube and weight the amount of your compound.

Change gloves in case you think your gloves are contaminated.

- Clean the outside of the tube with a tissue containing an appropriate solvent.
 - Wrap parafilm around the tube and centrifuge the tube that contains the compound.
 - Calculate the amount of solvent you need for dissolving/diluting the compound.
 - Add the solvent to the compound.
 - Seal the eppendorf tube with parafilm and vortex until completely dissolved.
 - Clean the original compound/carcinogen bottle with an appropriate solvent.
 - Clean up the cabinet and dispose tissues and other waste in the small plastic bag (present in the Laminar Flow Cabinet). The plastic bag has to be disposed in the blue biohazard container.
 - Store the solutions in the storage room or use it immediately
- * If you are planning to expose cells with a compound dissolved in DMSO, leave the solution at room temperature 1 hour in advance.

3 Dissolving compound in the industrial bottle, for instance BPDE

Preparing a stock solution can also be done in the original chemical jar/bottle. Information about the amount of compound in the jar/bottle should be provided by the manufacturer.

- Calculate the amount of the solvent you need to add in order to achieve the desired stock concentration.
- Add the calculated solvent volume to bottle.
- Seal with parafilm and vortex.
- Aliquot in desired volumes in eppendorf tubes.
- Denote compound, concentration, date and name on a label which can be stick on the tubes.
- Seal tubes with parafilm and store at correct temperature (preferably in a box).