

Inhibitor Handling Instructions

1. How to deal with the product(s) after receiving?

The packaging of the product(s) may have turned upside down during transportation, resulting in the inhibitor adhering to the neck or cap of the vial. Take the vial out of its packaging and gently shake to let the inhibitor fall to the bottom of the vial. For liquid products, centrifuge at 200-500 RPM to gather the liquid at the bottom of the vial. Try to avoid loss or contamination during handling.

2. What is the range of the weighing errors for our products?

Weighing range	Error range
5 - 25 mg	0.1 mg
50 -500 mg	1 mg
>1 g	2 - 5 mg

3. How do you prepare stock solutions?

According to your experimental design, select a suitable solvent to prepare a concentrated stock solution. The solubility information, tested by our Quality Control Department, can be found on the product page of our website. For in vitro experiments, DMSO is often used as the solvent. It is then diluted to make a working solution (e.g. 1:1000 dilution in cell culture medium). A typical table for solubility is outlined below:

Solubility (25°C)	DMSO X mg/mL*
	Water slightly or not soluble
	Ethanol slightly or not soluble

*The solubility value is obtained from our technical team; the product can be easily dissolved in DMSO at X mg/mL. The solubility value may be higher, but our technical team did not measure for a higher value.

If you are having difficulties dissolving an inhibitor, we suggest you:

(1) Recalculate the concentration of the stock solution, according to the following formula: the actual concentration (mg/mL) = molecular weight (g/mol) × concentration (mM) × 10⁻³

(2) Check whether the solvent has been contaminated, e.g. DMSO absorbs moisture.

(3) Some inhibitors are difficult to dissolve due to their structure or chemical characteristics and require additional mixing methods such as vortexing or ultrasonication. The inhibitor may need to be ultrasonicated for 1 h or overnight. If necessary, heat the inhibitors no higher than 50°C to prevent altering the product.

(4) Send an email to techbio@selleckchem.com and let our tech support staff know if you cannot dissolve the inhibitor completely. We will try to provide additional assistance.

4. How do I store the inhibitor(s)?

You can find the inhibitor storage recommendations on the product webpage. Here are some general guidelines:

Storage	3 years -20°C powder As a powder, the inhibitor can be stored at -20°C for up to 3 years.
	2 weeks 4°C in solvent The inhibitor in solvent can be stored at 4°C for up to 2 weeks.
	2 years -80°C in solvent The inhibitor in solvent can be stored at -80°C for up to 6 months.

Aliquot the stock solution to routine usage volumes and store at -20°C or -80°C. Avoid repeated freezing and thawing. Once thawed, aliquots may be kept at 4°C for 2 weeks.

5. How to prepare the working solution?

(1) Calculate the dilution required by using our dilution calculator.
<http://www.selleckchem.com/dilutioncalculator.jsp>

(2) Slowly add the stock solution into the solvent until obtaining the desired concentration. Mix by vortexing or repeated pipetting.

(3) Add a drop of working solution onto a slide and check whether there is precipitation under the microscope. If present, allow the working solution to stand for 10 min, vortex or pipette and then recheck whether there is precipitation under the microscope. If there is still the precipitation, please send an email to techbio@selleckchem.com for help.

(4) Our compounds are mainly liposoluble, so it is common that there is precipitation when diluted by aqueous solvent such as cell culture medium or PBS. They can be completely dissolved by ultrasonication.

6. How do I sterilize the working solution?

We suggest you to use filters to sterilize the working solution. **DO NOT sterilize the working solution by autoclaving.**

7. What issues need special attention for cell-based assays?

(1) DMSO is used to prepare the stock solution in most cell-based assays. The stock solution is then diluted in the culture medium to prepare a working solution. Make sure the concentration of DMSO is < 0.3% to avoid poisoning the cells. A negative control in the experiment is usually the culture medium with DMSO at the same concentration.

(2) Our compounds are mainly liposoluble, so it is common that there is precipitation when diluted by the cell culture medium. However, they can be completely dissolved by ultrasonication. If there is still the precipitation, please send an email to techbio@selleckchem.com for help.

8. What issues need special attention for animal experiments?

(1) Our compounds are mainly liposoluble. If DMSO is used to prepare the stock solution and then diluted to prepare the working solution for animal experiments, it may not be possible to obtain the required doses. Instead, hydrotropy agents, such as sodium carboxymethyl cellulose (CMC-Na), Tween 80, or glycerol, are needed. Please send an email to techbio@selleckchem.com if you require further assistance.

(2) Administration volumes considered good practice (and possible maximal dose volumes):

Species	Route and volumes (mL/kg)					
	Oral	s.c.	i.p.	i.m.	i.v. (bolus)	i.v. (slow inj.)
Mouse	10 (50)	10 (40)	20 (80)	0.05 (0.1)	5	(25)
Rat	10 (40)	5 (10)	10 (20)	0.1 (0.2)	5	(20)
Rabbit	10 (15)	1 (2)	5 (20)	0.25 (0.5)	2	(10)
Dog	5 (15)	1 (2)	1 (20)	0.25 (0.5)	2.5	(5)

For more details, please read: Diehl KH, et al. J Appl Toxicol, 2001, 21(1), 15-23.

(3) Drugs that are administered orally sometimes cannot be made into a clear solution due to limitations in solubility. It may not influence the efficacy of the drugs by gavage using suspensions or emulsions.

(4) If DMSO is used to prepare the stock solution, make sure the concentration of DMSO is < 5% to avoid toxicity.

(5) Conversion of different model animals based on BSA

Species	Weight (kg)	Body Surface Area (m ²)	K _m factor
Baboon	12	0.6	20
Dog	10	0.5	20
Monkey	3	0.24	12
Rabbit	1.8	0.15	12
Guinea pig	0.4	0.05	8
Rat	0.15	0.025	6
Hamster	0.08	0.02	5
Mouse	0.02	0.007	3

Animal A (mg/kg) = Animal B (mg/kg) multiplied by $\frac{\text{Animal B } K_m}{\text{Animal A } K_m}$

Value based on data from FDA Draft Guidelines: Center for Drug Evaluation and Research, Center for Biologics Evaluation and Research. (2002) Estimating the safe starting dose in clinical trials for therapeutics in adult healthy volunteers. U.S. Food and Drug Administration, Rockville, Maryland, USA

For example, to convert the dose of resveratrol used in a mouse (22.4 mg/kg) to a dose based on surface area for rat, multiply 22.4 mg/kg by the K_m factor for a mouse and then divide by the K_m factor for a rat. This calculation results in a rat equivalent dose for resveratrol of 11.2 mg/kg.

Rat dose (mg/kg) = mouse dose (22.4 mg/kg) $\times \frac{\text{mouse } K_m(3)}{\text{rat } K_m(6)} = 11.2 \text{ mg/kg}$

9. What issues need attention for special products?

(1) For oily compounds, add DMSO to dissolve the compound and prepare a stock solution with a concentration according to the volume available. You do not need to weigh the compound.

(2) For unstable compounds, add nitrogen to the vial when preparing stock solutions.

10. What are some safety precautions I must take when using products from Selleck Chemicals?

You can download the material safety data sheet (MSDS) from the product page for hazard identification, first aid measures, fire fighting measures, etc.