

100× Phosphatase Inhibitor Cocktail MIX III

Cat: IKM1080

Storage: Store at -20°C, 1 year.

Size: 1 * 1mL / 10 * 1mL

Introduction

Extracts such as cells or tissues contain many endogenous proteases, phosphatases, etc. In the in vitro environment, the proteins in the extracts are prone to degradation or modification, which affects subsequent protein detection. Therefore, adding inhibitors such as proteases and phosphatases to the extract is an effective method to prevent protein degradation and demodification.

100 × phosphatase inhibitor Cocktail MIX III is a mixture of three phosphatase inhibitors, which can effectively inhibit serine / threonine phosphatase and alkaline phosphatase, and is used to maintain the phosphorylation state of proteins. Among them, (-)-p-Bromotetramisole Oxalate is a potent irreversible alkaline phosphatase inhibitor; cantharidin is a reversible serine/threonine phosphatase inhibitor; calyculin A is a reversible protein phosphatase 1 and 2A (PP1 and PP2A) inhibitor. It can be widely used in WB, Co-IP, pull-down, IF, IHC, kinase assay and so on.

This product is presented with an additional 100 mM EDTA, and customers can choose whether to use it according to their own needs.

Kit Components

Composition	Component	Solvent	Size	Storage
100 × Phosphatase inhibitor Cocktail MIX III	2.5 mM (-)-p-Bromotetramisole Oxalate	DMSO	1*1mL/10*1mL	-20°C
	500 μM Cantharidin			
	1 μM Calyculin A			
100mM EDTA	100 mM EDTA	Water	1*1mL/10*1mL	2-8°C

Product advantage

1. The product composition is clear and the concentration is clear.
2. Product compatibility is good, efficient inhibition of a variety of proteases, comprehensive protection of protein from degradation
3. This series of products are given an additional 100mM EDTA, which can be used directly without additional preparation.
4. Flexible customization. Customers can customize the required MIX or custom one-component proprietary Kits based on their own experiments. Specifications and packaging can also be customized.

Protocols (only for reference)

1. Before use, the product should be thawed at room temperature, and centrifuged at a low speed before opening the lid, so as to better shake the liquid adhered to the pipe wall to the bottom of the pipe.
2. The inhibitor mixture was added to the solution sample (such as cell lysis solution or tissue extract) at a ratio of 1 : 100 and used after mixing.
3. The product can be properly subpackaged and stored, and the lysis solution containing the inhibitor mixture should be used now, and should not be used after cryopreservation.
4. If you need to use EDTA, you can add it to the cracking solution in an appropriate proportion and use it after mixing.

Note

1. If the subsequent experiments need to detect the activity of metalloproteinases in the extract, EDTA should not be added.
2. This product is only for scientific research. Do not use in medicine, clinical diagnosis or treatment, food and cosmetics.
3. Please do not store in ordinary residential areas.
4. For your safety and health, please wear experimental clothes and wear disposable gloves and masks.

FAQ

Q : Can Inhibitor Cocktail not be diluted according to the specified proportion?

A: Our Cocktail uses a classic concentration ratio. In order to ensure that it has sufficient inhibitory effect on different types of proteases, it is recommended to dilute it in accordance with the specified ratio. However, the inhibitory effect of protease activity is related to many factors, such as the difficulty of target protein degradation, the concentration of protease, the activity of protease and the concentration of inhibitor, which will affect the final protective effect. The content of phosphatase in samples from different sources is different. Therefore, in actual use, the concentration can be adjusted appropriately according to the experimental results.

Q : Why use the inhibitor Cocktail? What are the advantages compared with the commonly used PMSF?

A : Cocktail is a low-toxic, comprehensive protein protection reagent that maximizes the protection of proteins from degradation by proteases. More reliable than a single inhibitor. PMSF is a classical serine protease inhibitor, which is widely used in the process of cell lysis and purification of proteins. However, PMSF has obvious shortcomings in many aspects, one of which is high toxicity. Therefore, AEBSF is selected as its substitute in protease inhibitors. The second is easy degradation failure, which is easy to fail during cell lysis, and the protease inhibitor Cocktail not only protects more comprehensively, but also has a more lasting effect.

Q: Will DMSO affect the experiment?

A : In general, DMSO does not affect the experimental results. DMSO is also used as a solvent for many bioactive substances, such as protein crystallization and co-immunoprecipitation. DMSO is stable at room temperature, and DMSO is also an excellent cell cryoprotectant at low temperature.

Q : I used Cocktail, but the results are still not ideal. Why?

A : For most proteins, Cocktail has a good effect. If there is still a low protein yield, it is necessary to carefully check the experimental steps and program design to reduce the links leading to protein degradation. For example, before cell treatment, the lysis system should be fully prepared, and Cocktail should be added in advance and mixed well. The cells to be broken should be added to the prepared lysis solution immediately after collection or removal from the refrigerator, so as to ensure the whole low temperature operation. If the target protein is special, there is no inhibitor of a specific protease family in the Cocktail, and a specific protease inhibitor can be added to the lysate.