

## ab204708 - Deproteinizing Sample Preparation Kit – TCA

For the removal of macromolecules which might interfere with target metabolites allowing further analysis.

View kit datasheet: [www.abcam.com/ab204708](http://www.abcam.com/ab204708)

(use [www.abcam.cn/ab204708](http://www.abcam.cn/ab204708) for China or [www.abcam.co.jp/ab204708](http://www.abcam.co.jp/ab204708) for Japan)

This product is for research use only and is not intended for diagnostic use

PLEASE NOTE: With the acquisition of BioVision by Abcam, we have made some changes to component names and packaging to better align with our global standards as we work towards environmental-friendly and efficient growth. You are receiving the same high-quality products as always, with no changes to specifications or protocols.

### **Storage and Stability:**

Store kit at room temperature upon receipt. Kit has a storage time of 1 year from receipt.

### **Limitations:**

Do not use kit or components if it has exceeded the expiration date on the kit labels

Do not mix or substitute reagents or materials from other kit lots or vendors. Kits are QC tested as a set of components and performance cannot be guaranteed if utilized separately or substituted.

### **Materials Supplied**

Item	Amount	Storage Condition (Before Preparation)	Storage Condition (After Preparation)
Trichloroacetic Acid Solution/TCA	3 mL	RT	RT
Neutralization Buffer I/Neutralization Solution*	4 mL	RT	RT

### **Materials Required, Not Supplied**

**These materials are not included in the kit, but will be required to successfully utilize this assay:**

- Microcentrifuge
- Microcentrifuge tubes
- Pipettes and pipette tips

### **Reagent preparation**

Briefly centrifuge small vials at low speed prior to opening.

**Trichloroacetic Acid Solution/TCA:** Ready to use as supplied. Store at room temperature, chill on ice before use.

**Neutralization Buffer I/Neutralization Solution:** Ready to use as supplied. If precipitation is observed, shake bottle a few times to resuspend precipitate. Store at room temperature, chill on ice before use.

### **Sample preparation**

Samples do not require additional preparation. Simply follow the sample preparation procedure described in the protocol you are using and perform the deproteinization step when indicated.

### **Assay procedure**

**Ensure Trichloroacetic Acid Solution/TCA and Neutralization Buffer I/Neutralization Solution are cold prior starting the protein precipitation procedure.**

#### **Protein Precipitation:**

1. Samples with high protein concentration (e.g. serum, urine):
2. Mix 100  $\mu$ L sample with 15  $\mu$ L of cold Trichloroacetic Acid Solution/TCA in a microcentrifuge tube.
3. Keep the sample on ice for 15 minutes.
4. Centrifuge at 12,000 x g for 5 minutes. Carefully transfer supernatant to another tube.
5. Samples with protein concentration < 25 mg/mL (e.g. tissue lysate, cell lysate, and yeast lysate),:
6. Mix 150  $\mu$ L sample with 15  $\mu$ L of cold Trichloroacetic Acid Solution/TCA in a microcentrifuge tube.
7. Keep the sample on ice for 15 minutes.
8. Centrifuge at 12,000 x g for 5 minutes. Carefully transfer supernatant to another tube.

**NOTE:** Procedure can be stopped at this point and deproteinized samples can be stored at -70°C for up to one month if necessary. We recommend nevertheless to complete the procedure and analyze the samples as soon as possible.

#### **Sample Neutralization:**

1. To neutralize excess TCA, add 10  $\mu$ L of cold Neutralization Buffer I/Neutralization Solution to the collected supernatant. Mix well.
2. Vent sample tube as there may be formation of CO<sub>2</sub>. Place sample on ice for 5 minutes.  
Samples are now deproteinized, neutralized and can be directly used in a variety of assays.

**NOTE:** For further sample analysis, if reaction buffer capacity is 100 mM or stronger, sample volume up to 50  $\mu$ L may be used if total reaction volume is 100  $\mu$ L. For systems with weak buffer capacities, lower sample volume must be used to maintain optimum pH in the reactions.

### **Calculations**

Addition of Trichloroacetic Acid Solution/TCA and Neutralization Buffer I/Neutralization Solution dilutes protein sample concentration down to 80% compared to the original concentration.

Correct values from your final calculations using 0.8 as dilution factor.

**Technical Support**

Copyright © 2023 Abcam. All Rights Reserved. The Abcam logo is a registered trademark. All information / detail is correct at time of going to print.

**For all technical or commercial enquiries please go to:**

[www.abcam.com/contactus](http://www.abcam.com/contactus)

[www.abcam.cn/contactus](http://www.abcam.cn/contactus) (China)

[www.abcam.co.jp/contactus](http://www.abcam.co.jp/contactus) (Japan)