

Version 2a Last updated 26 August 2020

# ab270547

## Inorganic Phosphate Binding Resin

A product of Expedeon, an  
Abcam company

Applicable to Expedeon product codes:

View ab270547

Inorganic Phosphate Binding Resin datasheet:

<https://www.abcam.com/ab270547>

(use [www.abcam.cn/ab270547](http://www.abcam.cn/ab270547) for China, or [www.abcam.co.jp/ab270547](http://www.abcam.co.jp/ab270547) for Japan)

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

## Table of Contents

1. Overview	3
2. Materials Supplied and Storage	3
3. Technical considerations	4
4. Preparation of Inorganic Phosphate Binding Resin	5
5. Pipetting of Inorganic Phosphate Binding Resin	5
6. Removal of inorganic phosphate from buffers	6
7. Removal of inorganic phosphate from protein samples	7

## 1. Overview

Inorganic phosphate (Pi) is a product of many enzymatic reactions, for example, those catalyzed by phosphatases and ATPases. Measurement of released Pi allows enzyme activity to be determined, but contamination of enzymes or buffers by Pi can lead to unacceptably high assay backgrounds.

Inorganic Phosphate Binding Resin (ab270547) provides a quick and easy way to remove contaminating inorganic phosphate from buffers and protein samples (e.g. tissue extracts). The resin works over a broad range of pH values and is unaffected by many commonly used buffer additives. This method is quicker than dialysis and unlike desalting does not necessarily lead to a significant dilution of the sample.

## 2. Materials Supplied and Storage

Store at +4°C immediately on receipt.

Item	Size
Inorganic Phosphate Binding Resin	5 g

### 3. Technical considerations

#### 3.1 Buffer compatibility:

Conditions	Interference
100 mM Sodium Acetate, pH 5.0	None
100 mM MES, pH 6.0	None
100 mM MOPS pH 7.0	None
100 mM Hepes, pH 7.5	None
100 mM Tris pH 8.0	None
50 mM Glycine pH 2.3	None
100 mM Sodium Carbonate, pH 9.2	Significant
1-3 M NaCl	None
3 M KCl	None
100 mM Tris pH 8.0 / 0.1% Tween 20	None
100 mM Tris pH 8.0 / 0.1% bovine serum albumin	None
100 mM Tris pH 8.0 / 2mM dithiothreitol (DTT)	None
20% saturated ammonium sulphate/10 mM Tris, pH 8	Slight
50 mM Tris/150 mM KCl 500 mM imidazole, pH 7.5	None
100 mM Tris base	Significant
0.1 M HCl	Slight
100 mM MES pH 4.8	None
1M Hepes, pH 7.5	None
0.1 M HCl	None
5 mM EDTA, pH 8.0	Slight
50 mM Tris/ 150 mM NaCl/ 10 mM EDTA, pH 8.0	Slight

**Δ Note:** Interference refers to an apparent reduction in capacity for inorganic phosphate when the resin is incubated in the stated buffers. The results may also be explained by a reduced rate of clearance of inorganic phosphate and we were not able to distinguish between these two possibilities under the experimental conditions employed.

### 3.2 Related products:

Inorganic Phosphate Binding Resin (ab270547) is suitable for use with the ATPase Assay Kit (ab270551) and GTPase Assay Kit (ab270553) to help prevent contamination.

## 4. Preparation of Inorganic Phosphate Binding Resin

- The resin is supplied as slurry in water. Upon prolonged storage or after rough handling (e.g. magnetic stirring) the solution above the resin may appear cloudy. In this situation, carefully decant the liquid and replace with distilled water.
- Since it is a dense matrix it settles rapidly, thus each wash step only takes a few minutes.
- This approach may also be used to exchange the resin into a buffer if required.

## 5. Pipetting of Inorganic Phosphate Binding Resin

- Many pipette tips have a fine bore at the point and pipetting of the resin may be difficult, particularly in the 50 to 200  $\mu$ l range. This problem is easily overcome by using scissors to remove 3-4 mm of plastic from the end of the tip to increase the bore.
- Inorganic phosphate binding resin settles quickly and it is necessary to agitate the solution while dispensing multiple aliquots. Magnetic stirring may be used with caution but gentler methods, such as shaking or overhead stirring, are less damaging to the beads. If any 'fines' are generated upon stirring, the resin should be washed as described in section 4.

## 6. Removal of inorganic phosphate from buffers

- To clear buffers completely of contaminating inorganic phosphate the total capacity of the resin that is added to the buffer must exceed the amount of inorganic phosphate in the solution. The capacity of this resin is >30  $\mu\text{mol/g}$ .
- The rate of clearance will be influenced by a number of factors (e.g. amount of resin, concentration of inorganic phosphate, total amount of inorganic phosphate, rate of mixing, and so on) and the best conditions in any given situation will have to be determined experimentally.
- Certain buffer additives may reduce the rate of adsorption of inorganic phosphate or the capacity of the resin. Further information concerning interfering substances may be found in the Technical Considerations section.

## **7. Removal of inorganic phosphate from protein samples**

### **7.1 Direct addition of resin:**

- 7.1.1 The resin is added directly to the sample and mixed by gentle inversion or by shaking on a rotary device.
- 7.1.2 It may be separated from the clarified sample by filtration through filter paper or by centrifugation.
- 7.1.3 With samples up to 2 mL volume the resin may be pelleted by centrifugation in a microfuge for about 30 seconds, and the supernatant carefully removed with a pipette.
- 7.1.4 To minimize dilution of protein samples most of the liquid in a portion of the resin may be removed using a sintered glass funnel attached to a vacuum pump. It is necessary to apply suction for only a few seconds. Add the resin to the sample using a clean spatula.

### **7.2 Addition of resin to dialysis buffers:**

- 7.2.1 If protein samples are to be dialyzed to remove phosphate, the resin can be added to the dialysis buffer to scavenge inorganic phosphate. In this way it may be possible to reduce the number of buffer changes.

# Technical Support

Copyright © 2020 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)