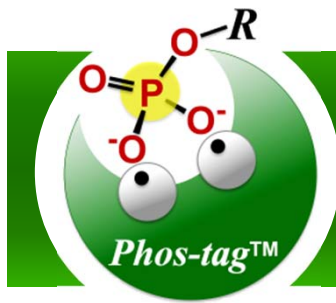


# Wako



# Phos-tag™ Series

★ *New Tools for Research on Phosphorylated Proteins*

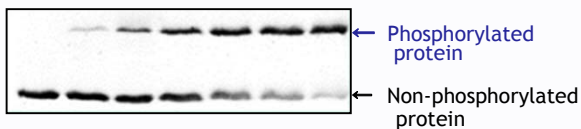
## What's Phos-tag ?

Phos-tag™ is a functional molecule that specifically binds all phosphorylated forms of Ser/Thr/Tyr. It is applicable for the specific separation of phosphorylated proteins as well as for the western blot detection, purification by agarose gel chromatography and MALDI-TOF/MS analysis. Following five kinds of lineup are available:

### Separation

*Specific Separation of phosphorylated proteins*

#### 1 Phos-tag™ Acrylamide



#### 2 SuperSep Phos-tag™ - Ready-to-Use Precast Gels

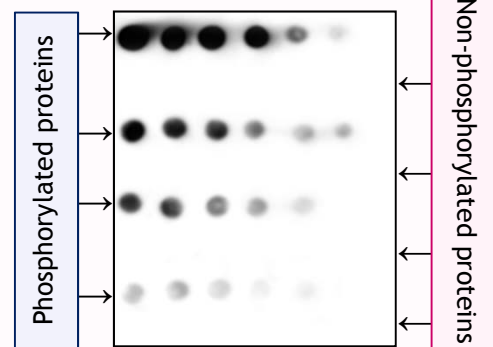


### Detection

#### 3 Phos-tag™ Biotin

*Achieves Specific Detection without anti-phosphorylated antibodies*

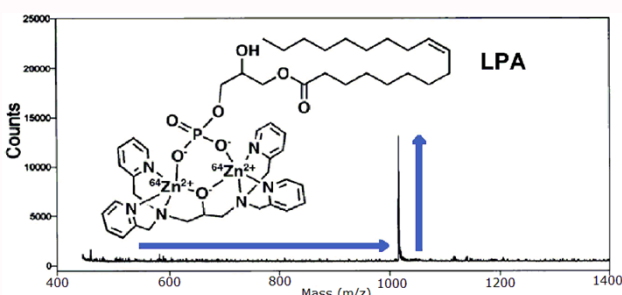
Applicable to Chemiluminescence detection of phosphorylated proteins



### Analysis

#### 4 Phos-tag™ Mass Analytical Kit For MALDI-TOF MS

*Sensitive for detection of phosphorylated molecules.*



### Purification

#### 5 Purification by gel chromatography Phos-tag™ Agarose



*Phosphorylated proteins can be trapped in physiological condition (pH 7.5).*

**[Application]**  
Purification of phosphorylated protein in A431 lysate

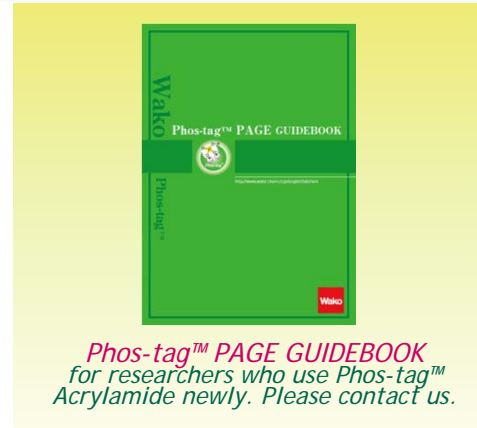
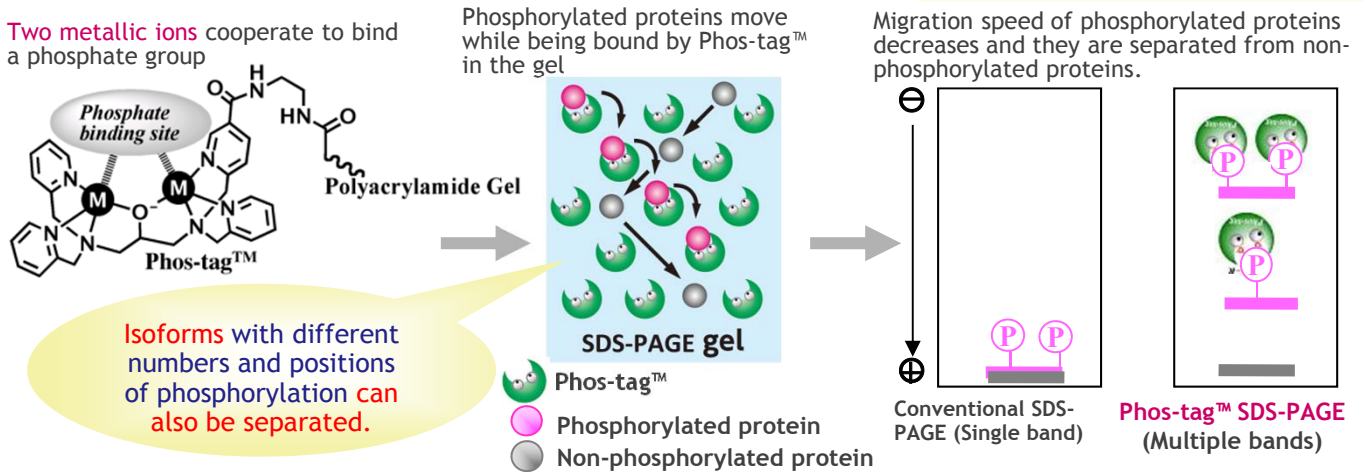
M: Molecular Marker  
Lane 1: Non absorbed fraction  
Lane 2: Absorbed fraction  
Lane 3: Column rinsing fraction

## 1 for Preparation of Self-made Acrylamide Gel Phos-tag™ Acrylamide

### Features

1. Recognizes and separates all phosphorylated forms of Ser / Thr / Tyr
2. Corresponds to the degrees and the positions of phosphorylation
3. Almost the same procedure as that of a conventional SDS-PAGE
4. Operates with your electrophoresis tank

### Principle



## 2 Ready-to-Use Precast Gels SuperSep Phos-tag™ (50 μmol/L Phos-tag™ Acrylamide contained)



### Features

1. Ready-to-use
2. Safety due to precast gel
3. Long-term stability (Stable for 6 months)
4. Almost the same basic mechanism as that of SDS-PAGE
5. Excellent storage stability by its neutral gel buffer.

Note:  
Before using SuperSep precast gel, check a sample for migration pattern problems with "SuperSep Ace" or conventional SDS-PAGE.



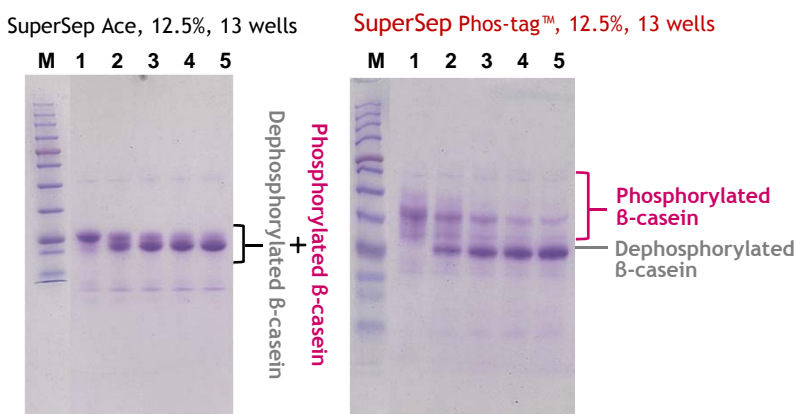
SuperSep Phos-tag™



EasySeparator – an electrophoresis tank optimized with SuperSep precast gel

Plate Size	100 x 100 x 3 (mm)		Acrylamide Conc.	10%; 12.5%; 15% or 17.5 %	
Gel Size	90 x 85 x 1 (mm)		ZnCl <sub>2</sub> Conc.	100 μmol/L	
Well number	13	17	Well volume	30 μL	25 μL
Phos-tag™ Conc.	50 μmol/L				

### Application ~ Dephosphorylation over time of β-casein ~



[Buffer] Tris-Glycine-SDS electrophoresis buffer  
[Sample]  
M: WIDE-VIEW™ Prestained Protein Size Marker III  
1: 0 min. β-casein (with AP treatment)  
2: 15 min. β-casein (with AP treatment)  
3: 30 min. β-casein (with AP treatment)  
4: 45 min. β-casein (with AP treatment)  
5: 60 min. β-casein (with AP treatment)  
AP: alkaline phosphatase  
[Condition] Constant current 35 mA for 60 min.  
[Staining] Quick-CBB Staining (Wako Cat. No. 299-50101)  
[Destaining] Deionized water with microwave treatment

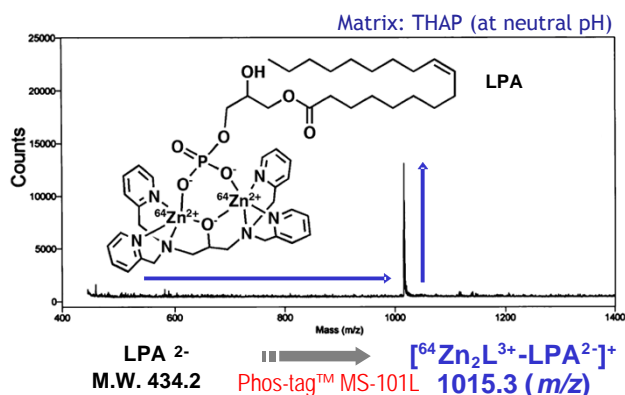
β-casein was dephosphorylated over time.  
Dephosphorylated β-casein can be separated from β-casein with SuperSep Phos-tag™.



## 4 MALDI-TOF/MS with High Sensitivity Phos-tag™ Mass Analytical Kit

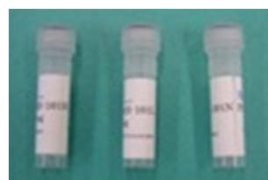
Before use, Phos-tag™ Mass Analytical Kit is mixed with samples for MALDI-TOF/Mass analysis. Phosphorylated molecule-Phos-tag™ complex is detected in a positive mode, and phosphorylated molecule usually difficult to detect can be detected with improved sensitivity.

### Application: Detection of Phosphorylated LPA complex with Phos-tag™



### 【Kit Contents】

- Phos-tag™ MS-101L (C<sub>27</sub>H<sub>29</sub>N<sub>6</sub>O<sup>64</sup>Zn<sub>2</sub><sup>3+</sup> MW: 581.4) ..... 5 mg
- Phos-tag™ MS-101H (C<sub>27</sub>H<sub>29</sub>N<sub>6</sub>O<sup>68</sup>Zn<sub>2</sub><sup>3+</sup> MW: 589.4) ..... 5 mg
- Phos-tag™ MS-101N (C<sub>27</sub>H<sub>29</sub>N<sub>6</sub>OZn<sub>2</sub><sup>3+</sup> MW: 584.3) ... 10 mg



Phos-tag™ Mass Analytical Kit

By the increased positive charge, detection sensitivity of phosphorylated LPA was improved

# Purification



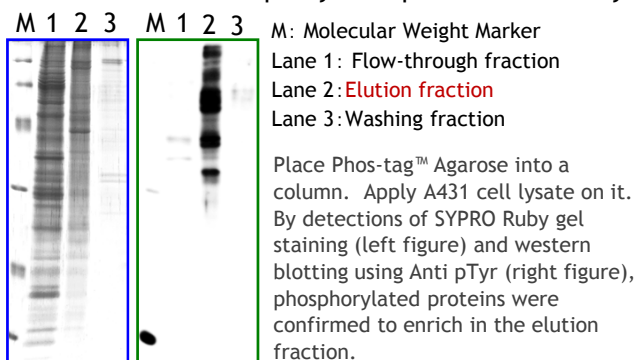
## 5 Purification of Phosphorylated proteins by Affinity Chromatography Phos-tag™ Agarose

### Features

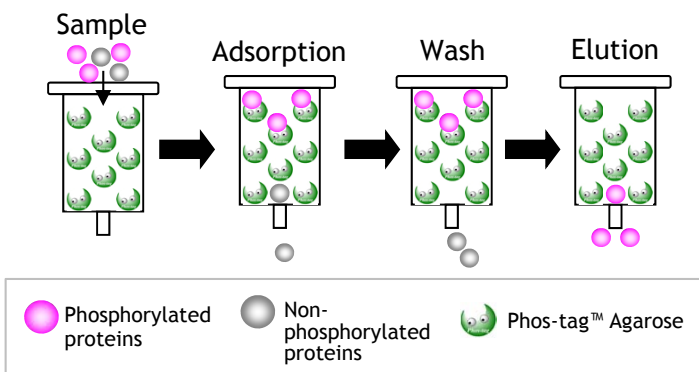
1. Phosphorylated proteins can be purified in 1 hour.
2. Proteins can be trapped in physiological condition (pH 7.5).
3. Purified with no reducing agent or surfactant.

### Application:

-Purification of Phosphorylated proteins in A341 lysate



### Principle of Phosphate Affinity Chromatography



	Description	Pkg. Size	Wako Catalog No.	Product No.	Note
4	Phos-tag™ Mass Analytical Kit	1 Set	305-93551	MS-101KIT	For MALDI-TOF MS
5	Phos-tag™ Agarose	0.5 mL	302-93561	AG-501	For enrichment, separation and purification
		3 mL	308-93563	AG-503	

Listed products are intended for laboratory research use only, and not to be used for drug, food or human use. Please visit our [online catalog](http://www.e-reagent.com) to search for other products from Wako: <http://www.e-reagent.com>

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Fax: 81-6-6203-1999  
Online Catalog: [www.e-reagent.com](http://www.e-reagent.com)

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Boston Sales Office (Cambridge, MA):  
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