



# Bio General Catalog

DNA Purification Kits

Proteins & Peptides

Phosphopeptide Purification & Enrichment

Low Molecular Weight Compounds  
Extraction & Purification

## Worldwide Ordering Information

To find your local distributor, please visit our website at  
<http://www.glsiences.com/products/contact.html>

Simply select your country from the list and your local distributor information will be displayed.

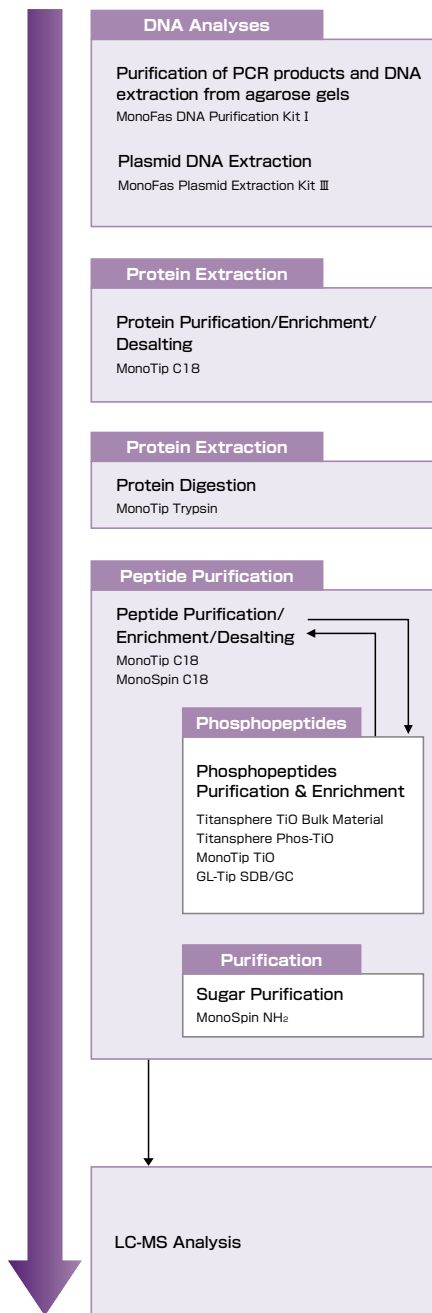
\* All trademarks are the property of their respective owners.

• We reserve the right to change specifications to make improvements without notice.

## CP-ANALYTICA GmbH

Am Pulverturm 17, A-2130 Mistelbach  
tel +43 (0)2572/4381 | fax +43 (0)2572/20791  
[info@cp-analytica.at](mailto:info@cp-analytica.at) | [www.cp-analytica.at](http://www.cp-analytica.at)

**GL Sciences Inc.**



## What is "Silica Monolith"? — 1 - 3

### DNA Purification Kits — 4 - 7

- MonoFas™ DNA Purification Kit I ----- 4
- MonoFas™ Plasmid Extraction Kit III ----- 6

### Proteins & Peptides — 8 - 11

- MonoTip™ Trypsin ----- 8
- MonoTip™ C18 ----- 10

### Phosphopeptide Purification & Enrichment — 12 - 19

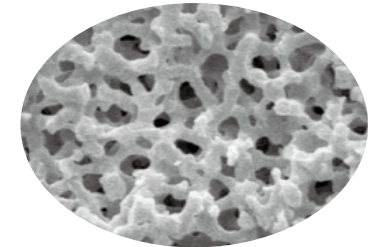
- Phosphorylated Protein Research ----- 12
- Titansphere™ TiO Bulk Material ----- 13
- Titansphere™ Phos-TiO ----- 14
- References ----- 18
- MonoTip™ TiO ----- 19
- GL-Tip SDB/GC ----- 20

### Low Molecular Weight Compounds — 22 - 25 (Drugs, Sugars, Catecholamines, Phosphate Pesticides etc) Extraction & Purification

- MonoSpin™ Series ----- 22

## Three-Dimensional Structure ⇒ Large Surface Area

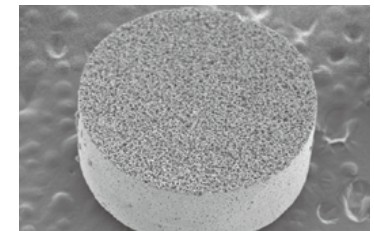
GL Sciences' silica monolith, created synthetically using ethyl silicate, has a very uniform three dimensional structure that shows excellent reproducibility from batch-to-batch.



Silica Monolith

## Solid Silica Gel Structure ⇒ Effective Enrichment for Small Volume Elution

The solid structure of GL Sciences' silica monolith eliminates the need for frits or filters at the ends of the column, thereby reducing dead volume that might otherwise lead to band broadening or sample loss. For example, when used in the form of a spin column, samples loaded in 10 μL volume, rinsed, and eluted with 10 μL elution buffer show excellent recoveries.

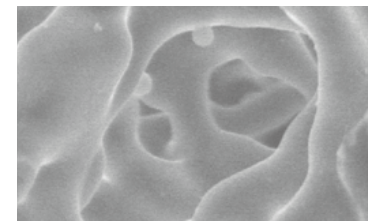


Silica Monolith

## High Porosity ⇒ Fast Processing

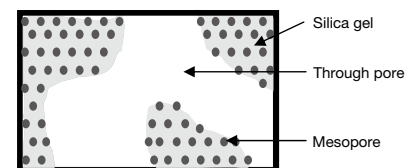
The high porosity of our silica monolith allows high flow rates to be used without loss of resolution or creation of high operating pressure. Even large, delicate analytes, such as long strands of DNA, can be analyzed rapidly without fear of sample degradation.

An optimized balance of through-pores and meso-pores provides the critically important combination of efficiency, separation speed, large-volume sample-loading, and small volume sample recovery.



Enlarged Silica Monolith

## Silica Monolith Structure



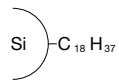
## Available Phases of GL Sciences Silica Monolith

### Silica gel (Si)



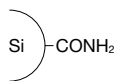
Details	Products	Applications
Unmodified silica surface provides a polar stationary support used for normal phase chromatography or DNA purification in combination with chaotropic salts.	MonoFas I (P.4) MonoFas III (P.6)	Genome DNA purification from legionella bacteria

### Octadecyl (C18)



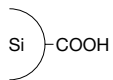
Details	Products	Applications
Silica monolith bonded with Octadecyl silane groups (ODS), produces a hydrophobic stationary support useful for reversed-phase chromatography.	MonoTip C18 (P.10) MonoSpin C18 (P.22)	Drug purification in Biological Sample/Peptides desalting

### Amide



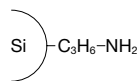
Details	Products	Applications
Optimal for the extraction of sugar chains and various acidic and basic hydrophilic compounds by HILIC mode.	MonoSpin Amide (P.22)	Purification of PA sugar chains

### CBA



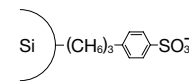
Details	Products	Applications
Modified with carboxy acid groups combining weak cation exchange. Optimal for the extraction of basic drugs.	MonoSpin CBA (P.22)	Basic drug purification in Biological Sample

### Aminopropyl (NH<sub>2</sub>)



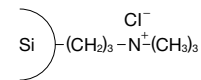
Details	Products	Applications
Silica monolith bonded with aminopropyl (NH <sub>2</sub> ) groups creates a polar stationary phase useful for sugar analysis or purification. Aminopropyl phase is also good for separating compounds using HILIC mode.	MonoSpin NH <sub>2</sub> (P.22)	Purification of PA sugar chains

### Propyl Benzene Sulfonic Acid (SCX)



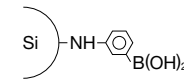
Details	Products	Applications
Silica monolith bonded with propyl benzene sulfonic acid groups creates a stationary phase that combines strong cation exchange and moderate hydrophobicity. SCX phase is particularly well suited for extraction of basic drugs.	MonoSpin SCX (P.22)	Extraction of basic drugs

### Trimethyl aminopropyl (SAX)



Details	Products	Applications
Silica monolith bonded with trimethyl aminopropyl groups creates a stationary phase that combines strong anion exchange and moderate hydrophobicity, particularly well suited for work with acidic drugs.	MonoSpin SAX (P.22)	Extraction of acidic drugs

### Phenylboronic acid (PBA)



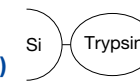
Details	Products	Applications
Silica monolith bonded with phenylboronic acid groups creates a stationary phase particularly well suited for work with compounds containing a catechol structure.	MonoSpin PBA (P.22)	Extraction of catecholamines

### Titanium dioxide Coating (TiO<sub>2</sub>)



Details	Products	Applications
Silica monolith bonded with titanium dioxide creates a stationary phase particularly well suited for work with phosphopeptides and other phospho-group containing compounds.	MonoTip TiO(P.19) MonoSpin TiO (P.22)	Purification of phosphopeptides Purification of glycosate (organophosphate pesticides)

### Trypsin Immobilization (Trypsin)



Details	Products	Applications
Silica monolith bonded with Trypsin is useful for performing rapid and efficient tryptic digests of protein samples.	MonoTip Trypsin (P.8)	Trypsin digestion of proteins

DNA Extraction & Purification

# MonoFas™ DNA Purification Kit I



MonoFas DNA Purification Kit I purifies DNA from PCR products and extracts from agarose gels.

Purified DNA can be used for multiple purposes such as sequence, ligation and digestion with a restriction enzyme etc.

## Features

### Multiple Roles – Purification from the PCR products and extraction from agarose gels

MonoFas DNA Purification Kit I, not only purifies DNA from the PCR reaction liquid, but also purifies DNA from the standard or low melting agarose gels employing TAE or TBE buffer.

MonoFas DNA Purification Kit I can be applied for both centrifuge and aspiration methods.

### Fast to purify

DNA purification from PCR samples in 4 minutes and in 9 minutes from agarose gels. As the spin column uses monolithic silica, it has a high porosity and enables an excellent liquid flow. This eliminates the step of vacuum centrifuge after the washing steps. Large surface areas of monolithic silica enable the binding of fragment DNA in a short time and wash only unnecessary compounds.

### High recovery rate even from small sample volume (10 µL)

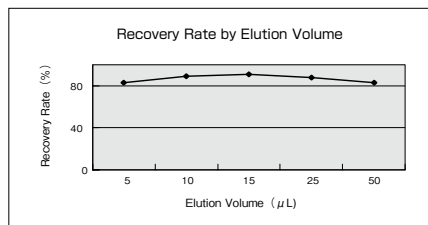
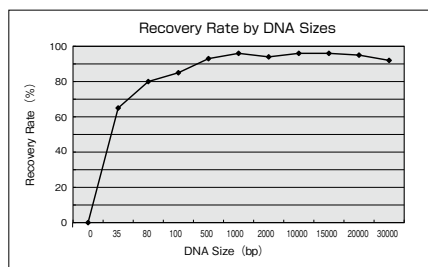
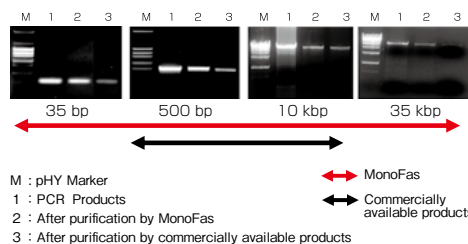
As there is no filter used for MonoFas DNA Purification Kit I, there will be no liquid accumulation in the column and samples can be eluted with a small amount of buffer.

MonoFas DNA Purification Kit I can elute as small as 10 µL.

### Purifies from short to long DNA, 35 bp – 35 kbp

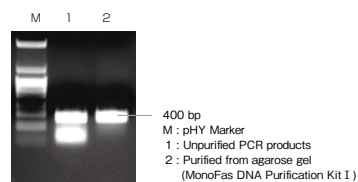
As the retentivity of MonoFas DNA Purification Kit I for nucleic acid is high, the kit can purify short DNA from 35 bp. Meantime the large through pore prevents the damage of DNA and purifies long DNA such as 35 kbp.

Single strand DNA under 80 mer, such as primers, can be removed from the remaining PCR products.

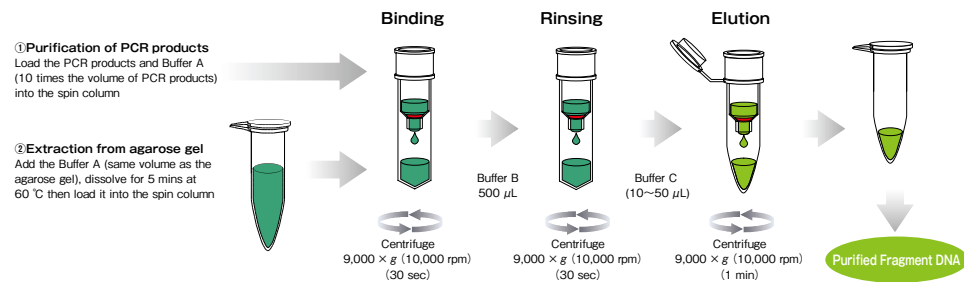


### Up to 1 g of agarose gel can be handled at once

1 g of agarose gel can be handled at once to extract more DNA.

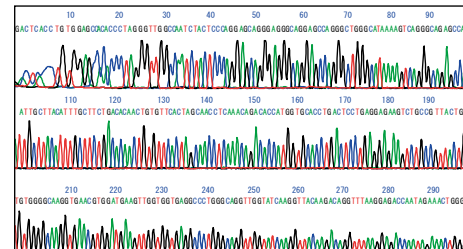


## How to Operate



### Accurate Sequence Analysis

Greater than 98 % precision by fluorescent sequence method and more than 500 bases can be analysed.



### Reference

Condition: Cycle sequencing method with Big Dye Terminator v3.1 manufactured by ABI  
Model: ABI 3730 Genetic Analyzer

### Easy centrifuge on the desk



Steady rotation > 6,200 rpm (+/- 20 %)  
Constant centrifuge acceleration > 2,000 × g (19,600 m/s<sup>2</sup>)

2 mins : DNA purification from PCR products  
7 mins : DNA purification from agarose gel

## Specifications

Description	Purification from PCR products	Extraction from agarose gel
Time	4 mins	9 mins
Maximum DNA Binding Amount	<10 µg	<10 µg
Maximum Agarose Gel Throughput	—	<1 g
Minimum Elution Amount	10 µL	10 µL
Column Volume	1 mL	1 mL
Processable DNA Range	35 bp - 35 kbp	35 bp - 35 kbp
Recovery Rate	>85 % (100 bp - 5 kbp)	>80 % (100 bp - 5 kbp)
	>60 % (5 kbp - 35 kbp)	>50 % (5 kbp - 35 kbp)
Primer Removal Percentage	95 %	—

## MonoFas™ DNA Purification Kit I Part Numbers

Description	Quantity	Cat.No.
MonoFas DNA Purification kit I	50 times	5010-21530
	100 times	5010-21531
	250 times	5010-21532
Buffer A	50 mL	5010-21506
Buffer B	21 mL	5010-21509
Buffer C	10 mL	5010-21508
Spin Column	100 pcs	5010-21541

Due to the exporting regulations (IATA), ethanol cannot be shipped. Please prepare ethanol before use to complete Buffer D-3.

\*Based on monolithic technology, Merck KGaA, Darmstadt, Germany

Plasmid DNA Extraction from *E.coli*

# MonoFas™ Plasmid Extraction Kit III



## Fast to extract!

MonoFas Plasmid Extraction Kit III easily extracts highly pure plasmid DNA from the *E.Coli* cultivated solution by using the centrifuge method and the DNA adsorption on the Silica Monolith. The extracted plasmid DNA can be directly used for the restriction enzyme treatment, ligation, sequence and PCR amplification etc.

## Features

### Fast to extract

Only 8 minutes to extract the plasmid DNA per sample with the MonoFas Plasmid Extraction Kit III.

### Excellent purification performance

By using the silica monolith technology, MonoFas Plasmid Extraction Kit III extracts highly pure plasmid DNA. There is no need for ethanol precipitation and chloroform extraction after the DNA extraction.

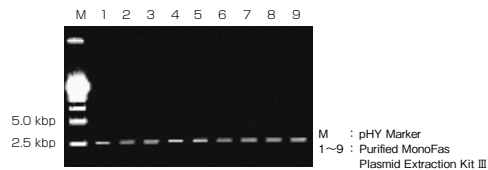
### BAC Clone purification

With the large surface of the silica monolith, BAC clone can also be purified.

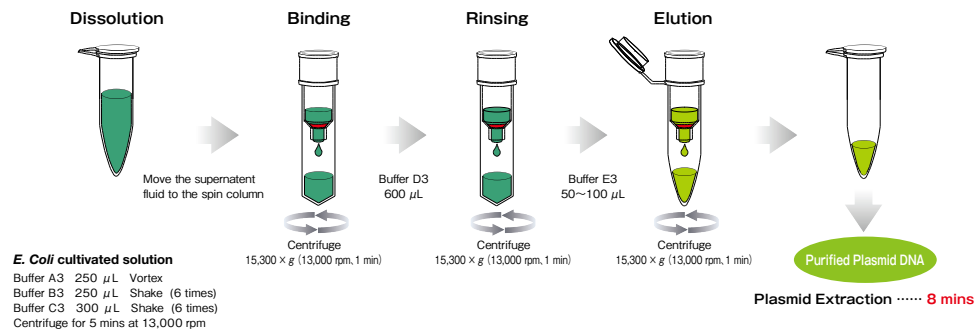
## Stable Recovery Rate

Due to the strict lot control of silica monolith, the same amount of Plasmid can be extracted from the same *E. Coli* culture solution.

The picture below shows the extraction of 2.6 kb low copy Plasmid in JM 109 bacteria coli and the recovery rate.

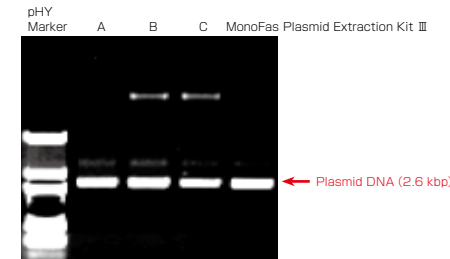


## How to Operate



## Comparison against commercially available plasmid kit

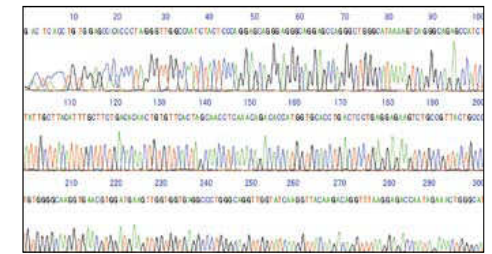
MonoFas Plasmid Extraction Kit III extracts purer plasmid DNA compared to other commercially available plasmid kits. This is due to the silica monolith structure that does not need extra pressure and the large surface areas for better nucleic acid adsorption.



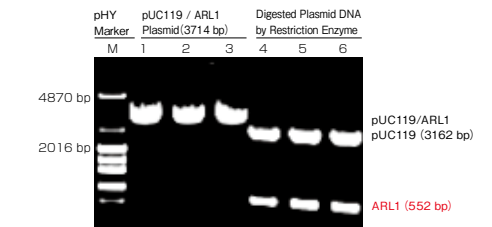
## Application

Extract the Plasmid DNA introducing the ARL1 gene from *E. coli* (DH 5 α) using the MonoFas Plasmid Extraction Kit III (Lane 1 - 3) Digest the extracted Plasmid with the restriction enzyme, EcoRI - Hind III, and confirm the target gene (ARL1 : 581 bp) (Lane 4 - 6)

## Accurate sequence analyses



Sample : DNA to make it amplify from human genome by using Takara PCR Kit .  
Sequence data analyzed with ABI Prism 3730xl Genetic Analyzer



## Specifications

Description	Specification
Cultivated Solution Throughput	1 - 3 mL
High - Copy-Plasmid	15 μg/mL culture
Low - Copy-Plasmid	5 μg/mL culture
Recommended Elution Volume	50 - 100 μL
DNA Purity (O. D. 260 / 280 mm)	1.7 - 1.9
Column Volume	1 mL

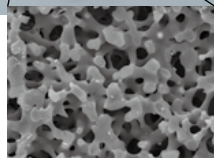
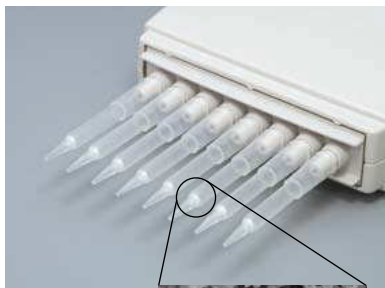
## MonoFas™ Plasmid Extraction Kit III Part Numbers

Description	Quantity	Cat.No.
MonoFas Plasmid Extraction Kit III	50 times	5010-21533
	100 times	5010-21534
	250 times	5010-21535
Buffer A3	50 mL	5010-21515
Buffer B3	50 mL	5010-21516
Buffer C3	50 mL	5010-21517
Buffer D3	21 mL	5010-21521
Buffer E3	10 mL	5010-21519
RNase A Buffer	500 μL	5010-21520

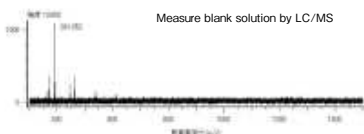
Due to the exporting regulations (IATA), ethanol cannot be shipped.  
Please prepare ethanol before use to complete Buffer D3.

\*Based on monolith technology, Merck KGaA, Darmstadt, Germany

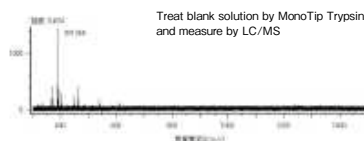
# Easy & Fast Trypsin Digestion MonoTip™ Trypsin



Monolith Structure



Measure blank solution by LC/MS



Treat blank solution by MonoTip Trypsin and measure by LC/MS

## Only 20 minutes to digest trypsin

MonoTip Trypsin is a sample preparation tip packed with silica monolith consisting of continuous through-pores bonded with the protein digestive enzyme Trypsin.

## Features

### Easy pipetting for trypsin digestion

Existing trypsin digestion methods are slow to digest trypsin at 37 °C with a low concentrated trypsin solution. However, MonoTip Trypsin digests trypsin only by a simple pipetting operation without a thermostatic chamber.

### Only 20 minutes to digest

At 37 °C, it usually takes more than 10 hours to digest trypsin. With MonoTip Trypsin, it only takes a few minutes pipetting operation to digest at room temperature for 20 minutes.

### High enzyme activity

With a large silica monolith surface area, MonoTip Trypsin has 100 µg Trypsin fixed per tip. This enables quick digestion of a highly concentrated protein (See application in P. 13). MonoTip Trypsin has trypsin derived from cow pancreas.

### No self-digestion

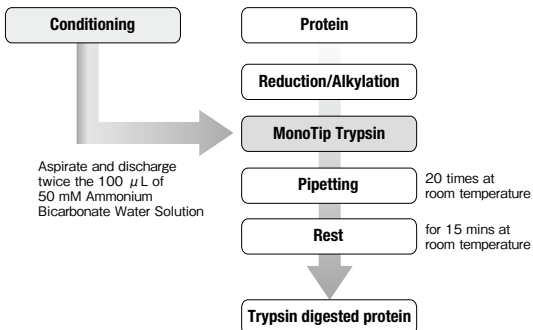
As Trypsin is fixed to the silica monolith, trypsin molecules do not physically contact each other. As there is no self-digestion of trypsin, dramatic high protein purification can be achieved.

Also the fixed trypsin is TPCK treated to inactivate the chymotrypsin activation.

### Long life time enzyme activity

MonoTip Trypsin can be stored at room temperature for 2 weeks and 1 year in a refrigerator.

## How to Operate



**Existing bath method**  
10 hours at 37 °C

**MonoTip Trypsin**  
20 minutes at room temperature (20 °C)

\*It might be difficult to digest samples such as histogenous cells in a short time.

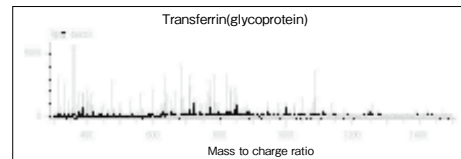
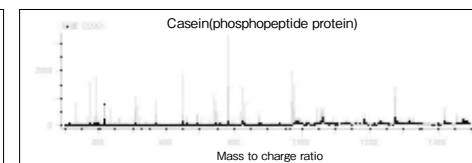
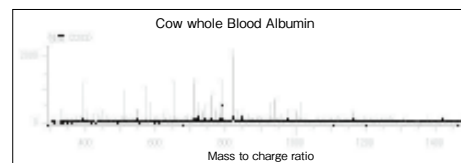
### Reference

High-throughput protein digestion by trypsin-immobilized monolithic silica with pipette-tip formula.

Ota S et.al J Biochem Biophys Methods. 2007 Feb 23;70(1):57-6

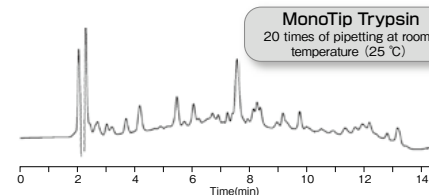
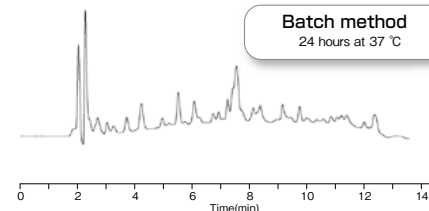
## Applications

β-Casein, cow whole blood albumin and transferrin were digested by MonoTip Trypsin and analyzed by LC/MS(/MS).



### Comparison between batch method (existing method) and MonoTip Trypsin

Sample : Ovalbumin (1 mg/mL)



### Conditions

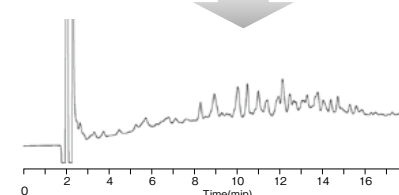
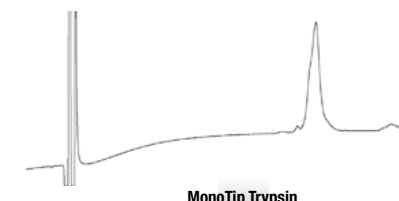
Column : Inertsil WP300 C8 (150 × 4.6 mm I.D.)  
Eluent : A: H<sub>2</sub>O (0.1 % TFA) B: CH<sub>3</sub>CN (0.1 % TFA)  
A/B = 90/10-(10 min)-40 / 60  
Flow Rate : 1 mL/min  
Detection : UV 210 nm

## Specifications

Description	Specification
Time	20 minutes
Sample Volume	20 - 200 µL
Tip Volume	200 µL
Derivation	Cow Pancreas
Organic Solvent Resistance	Acetonitrile less than 20 %
Packing Material	Silica Monolith
Through Pore Diameter	10 - 20 µm
Meso Pore Diameter	300 Å (30 nm)
Surface Area	100 m <sup>2</sup> /g
Chemical Bonding	TPCK Treated Trypsin

### Highly concentrated protein digestion

Deducted/Alkylated transferrin(100 µg)



### Conditions

Column : Inertsil WP300 C8 (150 × 4.6 mm I.D.)  
Eluent : A: H<sub>2</sub>O (0.1 %TFA) B: CH<sub>3</sub>CN (0.1 %TFA)  
A/B = 90/10-(20 min)-40 / 60  
Flow Rate : 1 mL/min  
Detection : UV 210 nm

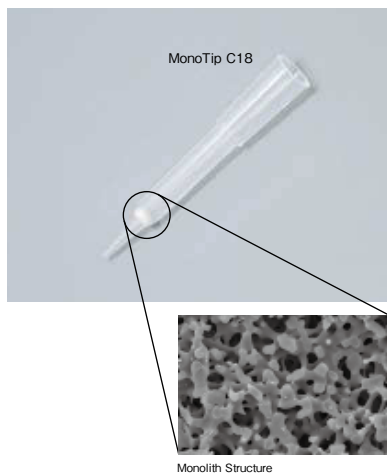
## MonoTip™ Trypsin Part Numbers

Description	Quantity	Cat.No.
MonoTip Trypsin	24 pcs	5010-21012
	96 pcs	5010-21010

\*Based on monolithic technology, Merck KGaA, Darmstadt, Germany

Desalting & Enrichment of Proteins/Peptides

# MonoTip™ C18



## Optimal for Desalting & Enrichment of Proteins/Peptides

MonoTip C18 sample preparation tips are packed with Silica Monolith that consists of continuous through-pores and octadecyl bonding.

Octadecyl group, C18, bonding enables the enrichment and demineralization of proteins and peptides with reverse phase mode by an easy pipetting operation.

## Features

### Easy to operate

An easy pipetting operation enables the enrichment and demineralization of proteins and peptides. See applications below.

### High purification efficiency, large sample loading volume and excellent recovery rate

It was difficult to regulate the interspace of each particle of the existing tips that use particle packing materials. This led to difficulties in achieving high reproducibility and recovery rates. However, with the technical advantage of silica monolith which contains uniform continuous through-pores, MonoTip C18 enabled an effective purification, a high recovery rate and a large sample loading volume.

MonoTip C18 is for large volume (200 µL).

### MonoTip C18

Perfect for peptide and protein samples with concentration of pmol-nmol and molecular weight up to 40 kDa.

## How to Operate

### MonoTip C18

#### Conditioning

- ① 100 % Acetonitrile Aspirate/Discharge Twice
- ② 20 % Acetonitrile (0.1 % TFA)

#### Binding

Aspirate and Discharge sample 6 times  
Add 0.1 % TFA to the sample

#### Rinsing

0 - 20 % Acetonitrile (0.1 % TFA)  
Aspirate - Discharge 3 times

#### Elution

60 % Acetonitrile (0.1 % TFA)  
Aspirate - Discharge 5 times

## Reference

"Simultaneous determination of ten antihistamine drugs in human plasma using pipette tip solid-phase extraction and gas chromatography/mass spectrometry"

Hasegawa C et al., *Rapid Commun. Mass Spectrom* (2006);20: 537-543

"Rapid demonstration of diversity of sulfatide molecular species from biological materials by MALDI-TOF/MS

Kyogashima M et al., *Glycobiology* (2006); 16(8),719-728

Simultaneous determination of methamphetamine and amphetamine in human urine using pipette tip solid-phase extraction and gas chromatography-mass spectrometry.

Kumazawa T, et al. *J Pharm Biomed Anal.* 2007 Jan 8;

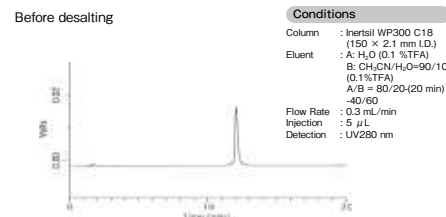
Establishment of a quantitative, qualitative, and high-throughput analysis of sulfatides from small amounts of sera by matrix-assisted laser desorption ionization-time of flight mass spectrometry.

Li G et.al *Anal Biochem.* 2007 Mar 1;362(1):1-7

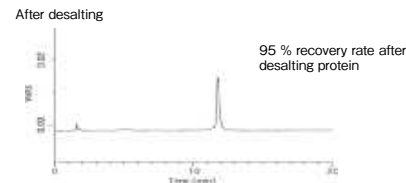
## Applications

### Reduce loss of proteins by desalting

Sample : Dissolve Cytochrome C in Tris-HCl (pH 7.4)  
100 nM NaCl (0.6 mg/mL)

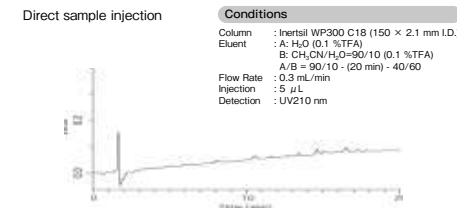


### MonoTip C18

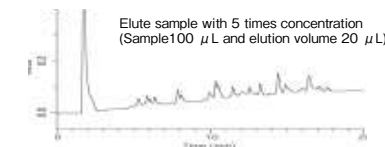


### Effective sample enrichment

Sample : β-Casein Tryptic Digest (0.1 mg/L)



### MonoTip C18



## Specifications

Description	Specification
Sample volume	20 - 200 µL
Sample treatment concentration	pmol - nmol order
Sample loading concentration	100 µg (Angiotensin II)
Tip volume	200 µL
Functional group	Octadecyl
Organic solvent resistance	Acetonitrile 100%
Packing material	Highly pure silica monolith
Through Pore Diameter	10 - 20 µm
Meso Pore Diameter	200 Å (20 nm)
Surface area	200 m <sup>2</sup> /g

## MonoTip™ C18 Part Numbers

Description	Volume	Quantity	Cat.No.
MonoTip C18	200 µL	24 pcs	5010-21002
		96 pcs	5010-21000

\*Based on monolithic technology, Merck KGaA, Darmstadt, Germany

# Phosphorylated Protein Research

Researching reversible phosphorylation plays a very important role in understanding the cell regulation such as the cell cycles, cell growth, differentiation and apoptosis.

However, when the phosphopeptides in proteins are analyzed as indicators, it is hard to detect them as the amount of phosphopeptides and its ionization efficiency are extremely low.

Therefore it is critical to enrich and purify only phosphopeptides from the protein digestion.

How strong is the phosphopeptide of each signaling factor after stimulating the cells? How can you find the factors that emerge on the same time course?

Here is the solution.

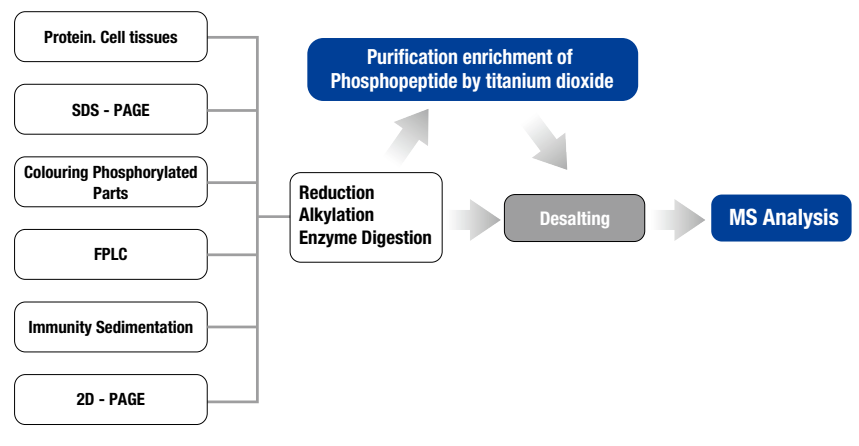
Titanium dioxide (titania) "Titansphere series", which is optimal for the enrichment and purification of phosphopeptides enables the dynamic state of phosphopeptides to be determined by easy centrifuge pretreatment method and mass spectrometry.

## Phosphorylated Protein Analyses by MS

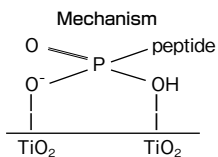
It is difficult to analyze phosphorylated proteins by MS

### Problems

- Amount of phosphorylated proteins in cells is very small
- Various numbers and parts where become phosphorylated
- Poor ionization efficiency



## Principal

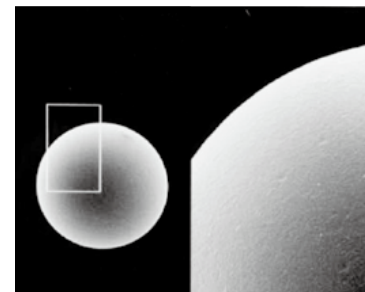


Phosphate groups are adsorbed on the surface of titanium dioxide under acidic conditions. A number of publications have been proving that titania and phosphopeptides selectively combine each other and we have developed the Titansphere series using titania.

Experience the satisfying performance of stable titanium beads modified and controlled by GL Sciences for the optimal purification and enrichment of phosphopeptides.

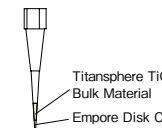
Bulk Materials for Purification & Enrichment of Phosphopeptides

# Titansphere™ TiO Bulk Material



Evenly spherical shaped porous particle, Titansphere TiO (Titanium Dioxide: TiO<sub>2</sub>) is a high-performing bulk material to selectively enrich and purify phosphopeptides.

### How to make a Titanium column



Cut the Empore Disk C8 and put it at the tip of a 200 μL pipette tip to be used as a filter

Stir and suspend the Titansphere TiO in CH<sub>3</sub>CN/H<sub>2</sub>O=80/20 v/v then load it to the tip (Adjust the amount of beads depending on the purpose)

Pressure the tip with a plastic syringe

### Purification/Enrichment Protocol

Only 5 steps by centrifuge treatment

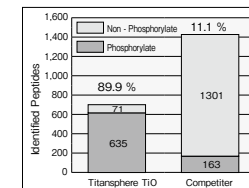


## Applications

### Efficient purification from HeLa Cell Lysate

The graph to the right shows the superior performance of Titansphere TiO using the HeLa Cell Lysate that consists of abundant non-phosphorylated peptides. Comparing the number of the identified phosphopeptides with the competitors' products, Titansphere TiO extracted 89.9 % of phosphopeptides even from cell lysate extract.

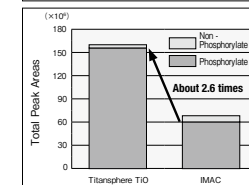
Sample : HeLa Cell Lysate, Sample volume : 50 μg, Titansphere TiO weight : 1 mg



### Comparison with IMAC

The graph to the right shows the performance comparison between Titansphere TiO and IMAC using Arabidopsis cell extract. Titansphere TiO showed 2.6 times the phosphopeptides peak area and 1.8 times the identified number of phosphopeptides compared to IMAC.

Sample : Arabidopsis Cell Extract, Sample volume : 100 μg, Titansphere TiO volume : 1 mg



### Identified Numbers of Phosphopeptides

	Phosphopeptides	Non - Phosphopeptides
Titansphere TiO	846	198
IMAC	474	379

### Specifications

Description	Specification
Particle Size	5 μm, 10 μm
Particle Shape	Spherical
Adsorption Spot	Titanium Dioxide Crystal
Pore Size	100 Å (10 μm)
pH Range	2 - 12
Gravity	1.74

### Titansphere™ TiO Part Numbers

Description	Volume	Cat.No.
Titansphere TiO 5 μm	500 mg	5020-75000
Titansphere TiO 10 μm	500 mg	5020-75010

## Purification & Enrichment of Phosphopeptides Titansphere™ Phos-TiO Kit



With the constant amount and optimized conditions of Titansphere bulk packed in the tip column, Titansphere Phos - TiO Kit offers an easy and stable operation to enrich phosphopeptides.

### Features

#### High selectivity

Due to the high affinity for the phosphopeptides, phosphopeptides can be detected by MS (Mass Spectrometry) even from the cell lysate extract that contains a trace amount of phosphopeptides.

#### Easy to Operate

Only 5 steps (only 40 minutes) to complete the operation. All steps can be done by centrifuge method that prevents possible human errors.

#### From few to large numbers of samples

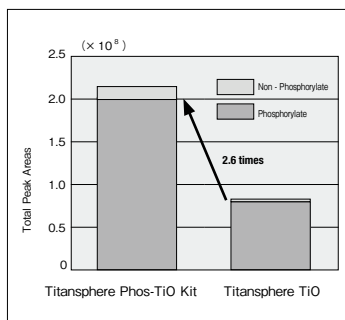
Each tip column is independent. With the special adaptor (available separately) any number of samples can be treated at once.

### Phosphopeptide Loading Capacity

Description	Content	
Sample	Tyr (PO <sub>3</sub> H <sub>2</sub> ) - Angiotensin II	
Tip Column	3 mg/200 μL	1 mg/10 μL
Loading Volume	3.5 μg	1.2 μg

### Performance

Optimal Titansphere TiO beads are used for Titansphere Phos - TiO Kit.



The existing Titansphere TiO beads were improved for better adsorption capacity of phosphopeptides. Compared to the existing Titansphere beads, Phos-TiO Kit showed 2.6 times more peak area and 1.6 times more identified phosphopeptides.

Sample : HeLa Cell Lysate  
Sample Volume : 50 μg  
Titansphere TiO beads : 1 mg

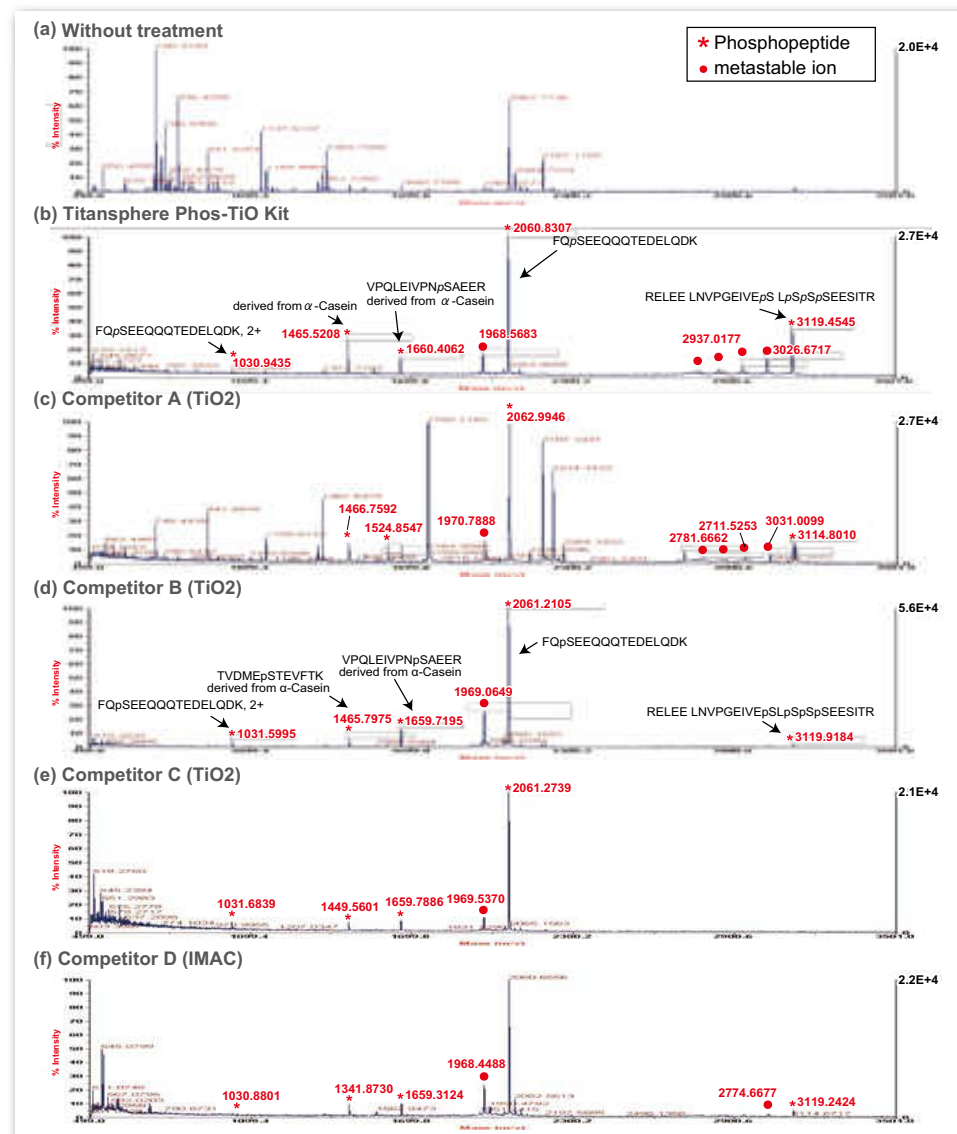
### Identified Number of Phosphopeptides

	Phosphopeptides	Non-Phosphopeptides
Titansphere Phos-TiO Kit	996	185
Titansphere TiO	635	71

Titansphere Phos-TiO Kit was developed based on the cooperation with Dr. Yasushi Ishihama from Graduation School of Pharm Sci, Kyoto University.

### Application

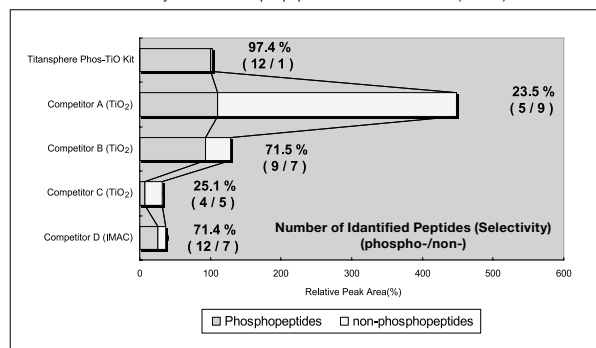
Fig.1 Comparison Between Titansphere Phos-TiO Kit and 4 Other Commercially Available Phosphopeptides Enrichment Methods (MALDI-TOF/MS)



The data above shows the purification efficiency of 2.5 μg of β-casein digestion using MALDI-TOF/MS. Compared to the untreated condition (a), phosphopeptides were selectively purified when using Titansphere Phos-TiO Kit. Also compared to other commercially available kits (c-e) Titansphere Phos-TiO Kit showed better selectivity. In general titanium dioxide is said to have poorer adsorption efficiency of multi-phosphopeptides than IMAC. However, Titansphere Phos-TiO Kit showed higher sensitivity and detection for 4-phosphopeptides than IMAC (f). Metastable ion is a dephosphorylated peak.

## Application

Fig. 2 Comparison Between Titansphere Phos-TiO Kit and 4 Other Commercially Available Phosphopeptide Enrichment Methods (LC-MS)



Tryptic digests of  $\alpha$ -casein, Futein and Phosvitin (2.5  $\mu$ g each) were used to compare the purification efficiency. The peak area value of phosphopeptides purified by Titansphere Phos-TiO Kit is shown as 100 % (n=3). The % shown in Fig. 2 is the ratio of phosphopeptides peak area value in the detected peptides peak area value. Also (%) in Fig. 2 shows the number of identified peptides (phosphopeptides/non-phosphopeptides).

## NEW Titansphere™ Phos-TiO for Large Volume Samples

GL Sciences now introduces larger versions of these spin columns as an extension of the Phos-TiO product line, including a 3 mL column containing 50 mg of our unique titanium dioxide (TiO<sub>2</sub>), and another column containing 100 mg of our TiO<sub>2</sub>.



### Specifications

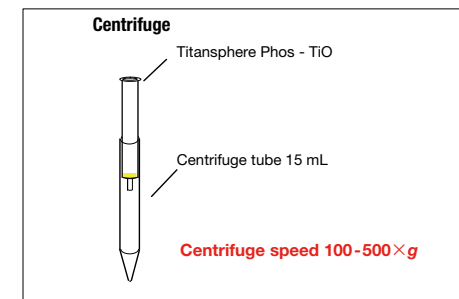
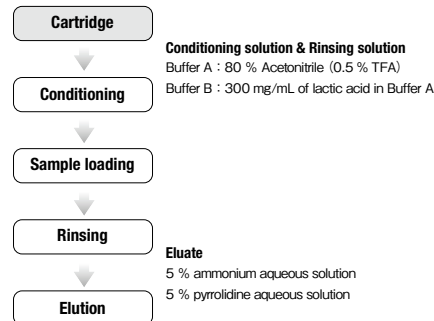
Sample	Tyr (PO <sub>3</sub> H <sub>2</sub> ) - Anagiotensin II	
Particle size	10 $\mu$ m	
Cartridge	50 mg/3 mL	100 mg/3 mL
Sample load volume	50 $\mu$ g	100 $\mu$ g

※ The maximum sample loading volume depends on the matrix condition.

## How to Operate

Phos-TiO columns are intended for use with a desktop or other centrifuge.

While some versions of Phos-TiO resemble pipette tips or SPE cartridges, these products are not intended for use with pipettes or SPE vacuum manifolds owing to mechanical design of the columns as well as the small particle size of the titanium dioxide media used.



**Purified & enriched phosphopeptides**

※ MonoSpin C18 is recommended for desalting.

### Titansphere™ Phos-TiO Kit Part Numbers

Description	Volume	Quantity	Cat.No.
Titansphere Phos-TiO Kit	1 mg/10 $\mu$ L	24 times	5010-21309
		96 times	5010-21310
	3 mg/200 $\mu$ L	24 times	5010-21311
		96 times	5010-21312

### Titansphere™ Phos-TiO Column Part Numbers

Description	Volume	Quantity	Cat.No.
Titansphere Phos-TiO Tip	10 $\mu$ L	24 pcs	5010-21302
		96 pcs	5010-21303
	200 $\mu$ L	24 pcs	5010-21307
		96 pcs	5010-21308
Titansphere Phos-TiO	50 mg/3 mL	25 (1 pcs)	5010-21290
	100 mg/3 mL	25 (1 pcs)	5010-21291

Description	Volume	Qty.(packed unit)	Cat.No.
Lactic Acid for Titansphere Phos-TiO	15 mL	1 pcs	5010-21295

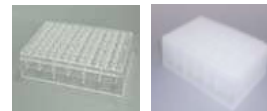
### Centrifuge adapter Part Numbers

Description	Quantity	Cat.No.
Centrifuge Adapter (10 $\mu$ L, 200 $\mu$ L Tips)	24 pcs	5010-21514
96well plate centrifuge adapter for 10 $\mu$ L Tips	1 pcs	5010-21340
	2 pcs	5010-21342
96well plate centrifuge adapter for 200 $\mu$ L Tips	1 pcs	5010-21341
	2 pcs	5010-21343

96well plate adapter is compatible with SBS standard plates.



Centrifuge Adapter      How to Use



96well plate centrifuge adapter for 10  $\mu$ L Tips      96well plate centrifuge adapter for 200  $\mu$ L Tips

## References

- Quantitative phosphoproteome analysis of a mouse liver cell line reveals specificity of phosphatase inhibitors.  
Pan C, Gnad F, Olsen JV, Mann M. *Proteomics*. 2008 Nov;8(21):4534-46.
- The Ser/Thr/Tyr phosphoproteome of *Lactococcus lactis* IL1403 reveals multiply phosphorylated proteins.  
Soufi B, Gnad F, Jensen PFR, Petranovic D, Mann M, Mijakovic I, Macek B. *Proteomics*. 2008 Nov;8(21):4534-46.
- Large-scale phosphorylation mapping reveals the extent of tyrosine phosphorylation in *Arabidopsis*.  
Sugiyama N, Nakagami H, Mochida K, Daudi A, Tomita M, Shirasu K, Ishihama Y, et al. *Syst Biol*. 2008;4:193. Epub 2008 May 6.
- TiO<sub>2</sub>-based phosphoproteomic analysis of the plasma membrane and the effects of phosphatase inhibitor treatment.  
Thingholm TE, Larsen MR, Ingrel CR, Kassem M, Jensen ON. *J Proteome Res*. 2008 Aug;7(8):3304-13. Epub 2008 Jun 26.
- The in vivo phosphorylation sites in multiple isoforms of amphiphysin I from rat brain nerve terminals.  
Craft GE, Graham ME, Bache N, Larsen MR, Robinson PJ. *Mol Cell Proteomics*. 2008 Jun;7(6):1146-61. Epub 2008 Mar 14.
- Phosphorylation Analysis of Primary Human T Lymphocytes Using Sequential IMAC and Titanium Oxide Enrichment.  
Carrascal M, Ovelheiro D, Casas V, Gay M, Abian J. *J Proteome Res*. 2008 Nov 6. [Epub ahead of print].
- Characterization of the human cerebrospinal fluid phosphoproteome by titanium dioxide affinity chromatography and mass spectrometry.  
Bahl JM, Jensen SS, Larsen MR, Heegaard NH. *Anal Chem*. 2008 Aug 15;80(16):6308-16.
- In vivo phosphorylation sites of barley tonoplast proteins identified by a phosphoproteomic approach.  
Ender A, Reiland S, Gerrits B, Schmidt UG, Baginsky S, Martinio E. *Proteomics*. 2009 Jan;9(2):310-21.
- The phosphorylation pattern of bovine heart complex I subunits.  
Palmasano G, Sardaneli AM, Signorile A, Papa S, Larsen MR. *Proteomics*. 2007 May;7(10):1575-83.
- Phosphoproteins of the chicken eggshell calcified layer  
Mann K, Olsen JV, Macek B, Gnad F, Mann M. *Proteomics*. 2007 Jan;7(1):106-15.
- Global, in vivo, and site-specific phosphorylation dynamics in signaling networks.  
Olsen JV, Blagoev B, Gnad F, Macek B, Kumar C, Mortensen P, Mann M. *Cell*. 2006 Nov 3;127(3):635-48.
- Phosphoproteome analysis of the human Chang liver cells using SCX and a complementary mass spectrometric strategy.  
Sui S, Wang J, Yang B, Song L, Zhang J, Chen M, Liu J, Lu Z, Cai Y, Chen S, Bi W, Zhu Y, He F, Qian X. *Proteomics*. 2008 May;8(10):2024-34.
- Phosphoproteome analysis of *E. coli* reveals evolutionary conservation of bacterial Ser/Thr/Tyr phosphorylation.  
Macek B, Gnad F, Soufi B, Kumar C, Olsen JV, Mijakovic I, Mann M. *Mol Cell Proteomics*. 2008 Feb;7(2):299-307. Epub 2007 Oct 15.
- Phosphopeptide enrichment by aliphatic hydroxy acid-modified metal oxide chromatography for nano-LC-MS/MS in proteomics applications  
Sugiyama N, Masuda T, Shinoda K, Nakamura A, Tomita M, Ishihama Y. *Mol Cell Proteomics*. 2007 Jun;6(6):1109-9. Epub 2007 Feb 23.
- Proteomic investigation of phosphorylation sites in poly (ADP-ribose) polymerase-1 and poly (ADP-ribose) glycohydrolase.  
Gagné JP, Moreel X, Gagné P, Labelle Y, Droit A, Chevalier-Paré M, Bourassa S, McDonald D, Hendzel MJ, Prigent C, Poirier GG. *J Proteome Res*. 2009 Feb;8(2):1014-29.
- Kinase-selective enrichment enables quantitative phosphoproteomics of the kinome across the cell cycle.  
Daub H, Olsen JV, Bairlein M, Gnad F, Oppermann FS, Köner R, Greff Z, Kéri G, Stemmann O, Mann M. *Mol Cell*. 2008 Aug 8;31(3):438-48.
- Phosphorylation of SUMO-1 occurs in vivo and is conserved through evolution.  
Matic I, Macek B, Hilger M, Walther TC, Mann M. *J Proteome Res*. 2008 Sep;7(9):4050-7.
- Distinction between human cytochrome P450 (CYP) isoforms and identification of new phosphorylation sites by mass spectrometry.  
Redlich G, Zanger UM, Riedmaier S, Bache N, Giessing AB, Eisenacher M, Stephan C, Meyer HE, Jensen ON, Marcus K. *J Proteome Res*. 2008 Nov;7(11):4678-88.
- Revealing the dynamics of the 20 S proteasome phosphoproteome: a combined CID and electron transfer dissociation approach.  
Lu H, Zong C, Wang Y, Young GW, Deng N, Souda P, Li X, Whitelegge J, Drows O, Yang PY, Ping P. *Mol Cell Proteomics*. 2008 Nov; 7(11):2073-89.
- Phosphoproteome analysis of fission yeast.  
Wilson-Grady JT, Villén J, Gygi SP. *J Proteome Res*. 2008 Mar;7(3):1088-97.
- Multiple phosphorylations in the C-terminal tail of plant plasma membrane aquaporins: role in subcellular trafficking of AtPIP2; 1 in response to salt stress.  
Prak S, Hem S, Boudet J, Viennois G, Sommerer N, Rossignol M, Maurel C, Santoni V. *Mol Cell Proteomics*. 2008 Jun;7(6):1019-30.
- Comparative phosphoproteomics of zebrafish *Fyn*/Yes morpholino knockdown embryos.  
Lemere S, Jopling C, Gouw J, Mohammad S, Heck AJ, Slijper M, den Hertog J. *Mol Cell Proteomics*. 2008 Nov;7(11): 2176-87.
- A quantitative atlas of mitotic phosphorylation.  
Dephoure N, Zhou C, Villén J, Beaussot SA, Bakalarski CE, Elledge SJ, Gygi SP. *Proc Natl Acad Sci U S A*. 2008 Aug 5; 105(31): 10762-7.
- Large scale phosphoproteome profiles comprehensive features of mouse embryonic stem cells  
Li QR, Xing XB, Chen TT, Li RX, Dai J, Sheng QH, Xin SM, Zhu LL, Jin Y, Pei G, Kang JH, Li YX, Zeng R. *Mol Cell Proteomics*. 2011 Apr;10(4):M110.001750. Epub 2010 Dec 13.
- Discovery of mouse spleen signaling responses to anthrax using label-free quantitative phosphoproteomics via mass spectrometry  
Manes NP, Dong L, Zhou W, Du X, Reghu N, Kool AC, Choi D, Bailey CL, Petrincio EF 3rd, Liotta LA, Popov SG. *Mol Cell Proteomics*. 2011 Mar;10(3):M110.000927. Epub 2010 Dec 28.
- Phosphoproteome analysis reveals regulatory sites in major pathways of cardiac mitochondria  
Deng N, Zhang J, Zong C, Wang Y, Lu H, Yang P, Wang W, Young GW, Wang Y, Korge P, Lotz C, Doran P, Liem DA, Apweiler R, Weiss JN, Duan H, Ping P. *Mol Cell Proteomics*. 2011 Feb;10(2):M110.000117. Epub 2010 May 22.
- Purification and identification of O-GlcNAc-modified peptides using phosphate-based alkyne CLICK chemistry in combination with titanium dioxide chromatography and mass spectrometry.  
Parker BL, Gupta P, Cordwell SJ, Laessen MR, Palmasano G. *J Proteome Res*. 2011 Apr 1; 10(4): 1449-58.
- Selective enrichment of sialic acid-containing glycopeptides using titanium dioxide chromatography with analysis by HILIC and mass spectrometry.  
Palmasano G, Lendal SE, Engholm-Keller K, Leth-Larsen, Parker BL, Larsen MR. *Nat Protoc*. Dec;5(12): 1974-82.
- Undesirable charge-enhancement of isobaric tagged phosphopeptides leads to reduced identification efficiency  
Thingholm TE, Palmasano G, Kjeldsen F, Larsen MR. *J Proteome Res*. 2010 Aug 6;9(8):4045-52.
- Glycoproteomic profile in wine: a 'sweet' molecular renaissance.  
Palmasano G, Antonacci D, Larsen MR. *J Proteome Res*. 2010 Dec 3;9(12):6148-59. Epub 2010 Oct 21.

## Purification & Enrichment of Phosphopeptides

# MonoTip™ TiO



MonoTip TiO is a tip packed with monolithic silica coated with titanium dioxide to selectively trap phosphopeptides to improve the detection range and reduce the time for analyses.

## Features

### Easy to Operate

An easy pipetting operation enables phosphopeptide enrichment. Unlike methods such as IMAC, MonoTip TiO does not need to coordinate the metal linkage.

### Large Sample Volume Available

MonoTip TiO is suitable for sample volumes from 50-200 µL. About 5 µg of phosphopeptide can be retained per tip, and also can be enriched by reducing the amount of solution.

## Application

Sample:  $\beta$ -Casein  
 1 phosphopeptide : FQpSEEQQTEDELQDK(MW=2061)  
 4 phosphopeptides : RFLEELNVPGEIVeSpSLpSpSpSpSEELSTR(MW=3122)

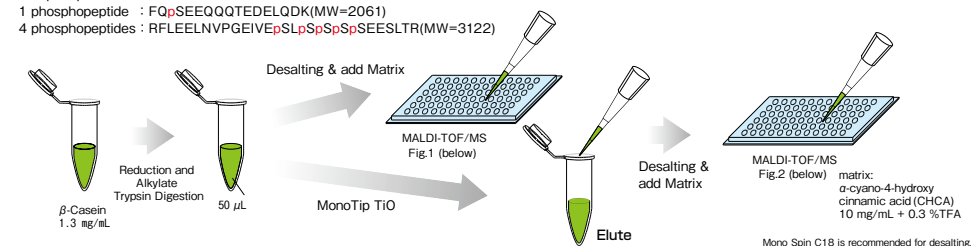


Fig. 1 : Before purification (Almost no phosphopeptides were enriched)

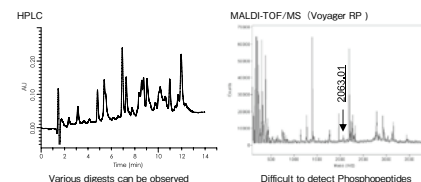
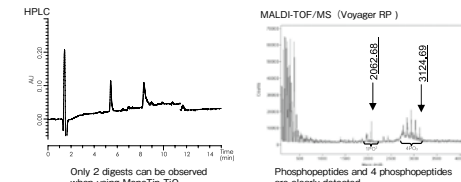


Fig. 2 : After purification (Phosphopeptides were selectively enriched)



## Specifications

Description	Specification
Operation time	Approx. 6 mins
Suitable sample volume	20 - 200 µL
Sample loading volume	up to 5 µg
Tip volume	200 µL
Packing material	Silica Monolith (Highly pure silica gel)
Through Pore Diameter	10 - 20 µm
Meso Pore Diameter	200 Å (20 nm)
Surface area	200 m <sup>2</sup> /g
Functional group	Titanium dioxide coating

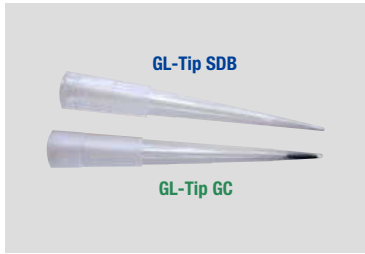
## MonoTip™ TiO Part Numbers

Description	Volume	Quantity	Cat.No.
MonoTip TiO	200 µL	24 pcs	5010-21007
		96 pcs	5010-21005

\*Based on monolithic technology, Merck KGaA, Darmstadt, Germany\*

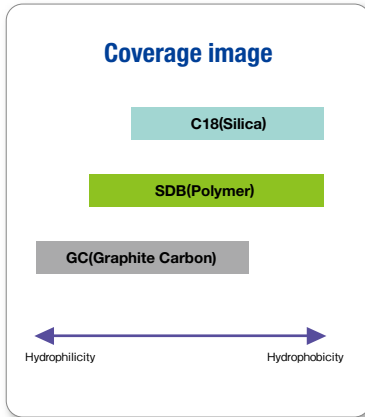
For Effective Desalting & Enrichment of Peptides

## GL-Tip SDB/GC



For proteome analysis, it is necessary to desalt and enrich peptides before introducing samples to MS. GL Sciences now introduces GL-Tip (200  $\mu$ L) series: GL-Tip SDB, tips packed with Empore DISK SDB (styrene divinylbenzen co-polymer) and GL-Tips GC, tips packed with graphite carbon.

GL-Tip SDB shows stronger retentivity than existing C18 tips. This strong retentivity enables more peptides (proteins) to be captured than trapped by existing C18 tips. GL-Tip GC enables the retention and recovery of highly hydrophilic peptides. Therefore the combination of GL-Tip SDB and GC realizes wider peptides coverage than ever. **GL-Tip adapts centrifuge method.**



### Features

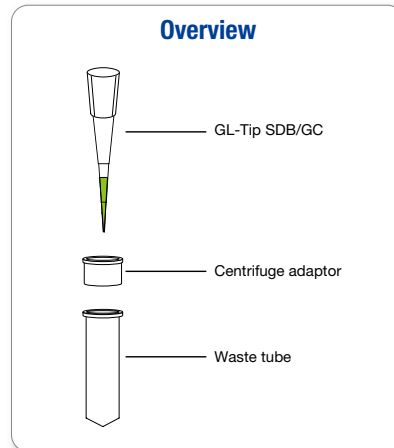
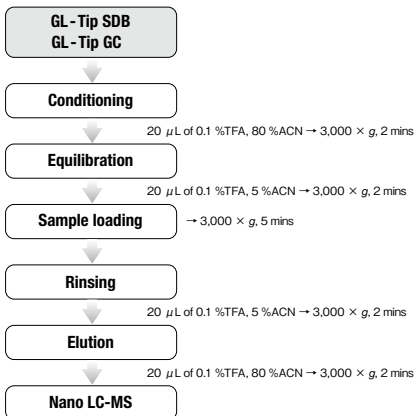
- Highly retentive
- Highly yield
- Easy to operate

### Specifications

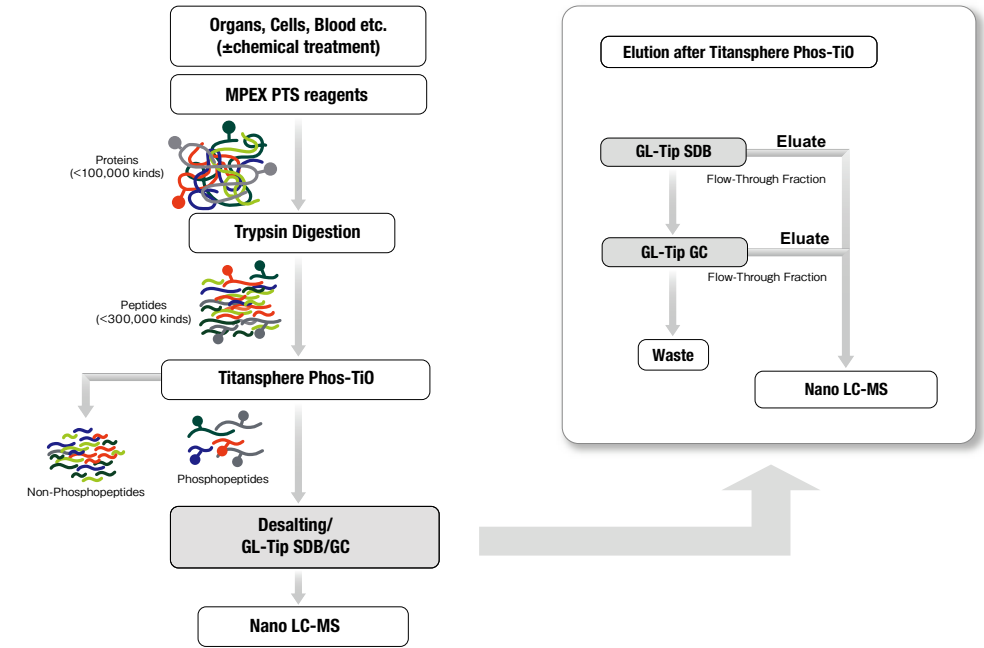
GL-Tip SDB	
Sample	Tyr (PO <sub>3</sub> H <sub>2</sub> )-AngiotensinII
Tip volume	200 $\mu$ L
Binding Capacity	60 $\mu$ g
GL-Tip GC	
Sample	Gly-Gly-Tyr-Arg
Tip volume	1 mg/200 $\mu$ L
Binding Capacity	30 $\mu$ g

\*The maximum sample loading volume depends on the matrix condition.

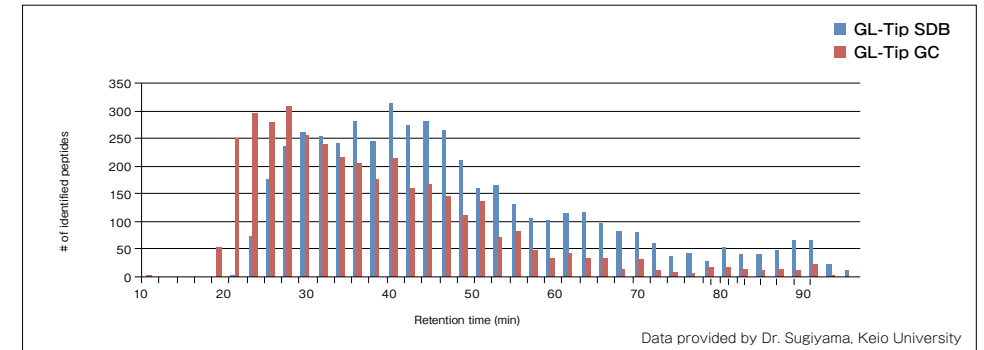
### Recommended Protocol



### Phosphopeptides Enrichment using GL-TIP & Titansphere Phos-TiO



### Retention Time Distribution



The solution containing phosphopeptides enriched by Titansphere Phos-TiO Kit from HeLa cell elution was desalted by GL-Tip SDB and GC. GL-Tip SDB traps a wide range of peptides while GL-Tip GC captures highly hydrophilic peptides.

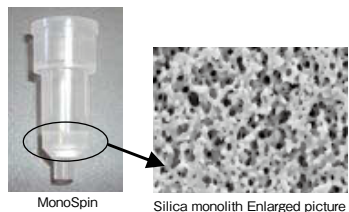
### GL-Tip SDB/GC Part Numbers

Description	Volume	Quantity	Cat.No.
GL-Tip SDB	200 $\mu$ L	96 pcs	7820-11200
GL-Tip GC	200 $\mu$ L	96 pcs	7820-11201
Centrifuge Adapter	—	24 pcs	5010-21514

Please note the adaptor is reusable

Monolithic SPE Column for the Purification and Enrichment of Small Sample Amounts

## MonoSpin™ Series



Optimal for sample purification and enrichment

MonoSpin is an SPE column packed with monolithic silica and is excellent for the sample pretreatment of small sample volume with easy and quick operation by centrifuge.

### Features

#### Easy to Operate

Easy operation by centrifuge

#### Fast

Speedy sample treatment with superb monolith silica through pore even for biological samples

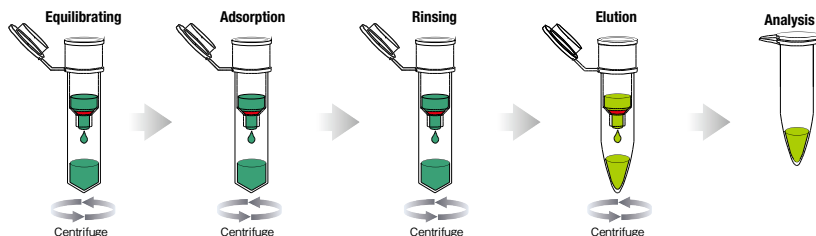
#### Small Sample Volume

Excellent for the pretreatment for samples of 50-800  $\mu$ L

#### Various Functional Groups

There are 8 kinds of functional groups for MonoSpin series.

### How to Operate



### Product Lineup

**MonoSpin C18**  
 Octadecyl functional group.  
 Optimal for drug extraction in biological samples, and desalting & enrichment of peptide samples.

**MonoSpin NH<sub>2</sub>**  
 Bonded with aminopropyl.  
 Optimal for the enrichment of sugar chain and/or hydrophilic compounds by HILIC mode.

**MonoSpin SCX**  
 Bonded with propyl benzene sulfonic acid combining both strong cation exchange & hydrophobic interaction. Optimal for the extraction of basic drugs.

**MonoSpin Amide**  
 Bonded with amide groups. Optimal for the extraction of sugar chains and various acidic and basic hydrophilic compounds by HILIC mode.

**MonoSpin SAX**  
 Bonded with Trimethyl aminopropyl combining both strong anion exchange & weak hydrophobic interaction. Optimal for the extraction of acidic drugs.

**MonoSpin PBA**  
 Specific column combined with phenyl boronic acid. Excellent for the selective extraction of cis diol compounds, such as catechol amines.

**MonoSpin TiO**  
 Monolith skeleton coated with titanium dioxide. Excellent for the enrichment of phosphopeptides

**MonoSpin CBA**  
 Bonded with carboxy acid combining both weak cation exchange. Optimal for the extraction of basic drugs.

### Application

#### Drugs in Urine (MonoSpin C18)

Various drugs can be purified and enriched by hydrophobic interaction of C18

**Solvents**  
 A (Rinsing) : CH<sub>3</sub>CN/Alkaline buffer (pH 12) = 10/90  
 B (Eluent) : CH<sub>3</sub>CN/0.1 % H<sub>3</sub>PO<sub>4</sub> (pH 3) 20 mM IPCC = 25/75 (IPCC-08 : Sodium 1-Octane sulfonate)

All the centrifugal processes at 3,000 rpm

**Conditioning**  
 ① Set the waste fluid tube and add 500  $\mu$ L of acetonitrile, then centrifuge (1 min)  
 ② Add 500  $\mu$ L of boiled water and centrifuge (1 min)  
 ③ Discard the waste fluid

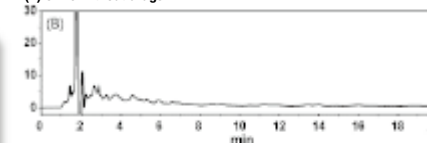
Sample: 500  $\mu$ L  
 Add alkali buffer (pH 12) 400  $\mu$ L  
 Add I.S. Methoxyphenamine 20  $\mu$ L

**MonoSpin C18 Sample loading**  
 Add the sample and centrifuge for 5 mins, discard the waste fluid

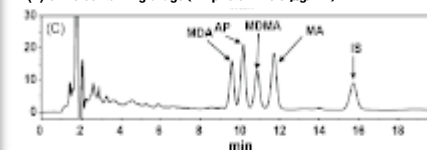
**Rinsing**  
 Add 500  $\mu$ L of A and centrifuge for 1 min, discard the waste fluid and set the recovery tubes

**Elution**  
 Add 200  $\mu$ L of B and centrifuge for 1 min to recover the eluent solution

#### (A) Urine without drugs



#### (B) Urine containing drugs (Amphetamine 5 $\mu$ g/mL)



#### Conditions

Column : C18 Column (150  $\times$  4.6 mm I.D.)  
 Eluent : CH<sub>3</sub>CN/0.1 % H<sub>3</sub>PO<sub>4</sub>, 20 mM IPCC-08 = 25/75  
 Flow Rate : 1 mL/min  
 Detection : UV 215 nm (PDA Detector)  
 Injection Vol: 10  $\mu$ L  
 Analyte : Methamphetamine (MA)  
 Amphetamine (AP)  
 3,4-Methylenedioxymethamphetamine (MDMA)  
 3,4-methylenedioxyamphetamine (MDA)

<J. Chromatogr A 1208 (2008) 71-75>

#### References

Extraction of amphetamines and methylenedioxyamphetamines from urine using a monolithic silica disk-packed spin column and high-performance liquid chromatography-diode array detection  
*J Chromatogr A*. 2008 Oct 24;1208(1-2) : 71-5

Simultaneous determination of dibucaine and naphazoline in human serum by monolithic silica spin column extraction and liquid chromatography-mass spectrometry.  
*J Chromatogr B Analyt Technol Biomed Life Sci*. 2008 Sep 1; 872(1-2) : 186-90

Simultaneous determination of amitraz and its metabolite in human serum by monolithic silica spin column extraction and liquid chromatography-mass spectrometry.  
*J Chromatogr B Analyt Technol Biomed Life Sci*. 2008 May 1;867(1) : 99-104.

#### Phosphorus-containing Amino Herbicides in Urine (MonoSpin TiO)

Phosphorus-containing amino herbicides can be selectively purified and enriched out of a heavily contaminated matrix such as urine.

**Solvents**  
 A (Rinsing) : 0.1% TFA in 80 % CH<sub>3</sub>CN aqueous solution  
 B (Rinsing) : 0.1% TFA in 50 % CH<sub>3</sub>CN aqueous solution  
 C (Eluent) : 2 % NH<sub>2</sub> aqueous solution

All the centrifugal processes at 5,200  $\times$  g

**Conditioning**  
 ① Set waste fluid tubes, put 20  $\mu$ L of A and centrifuge for 2 mins  
 ② Put 20  $\mu$ L of B and centrifuge for 2 mins

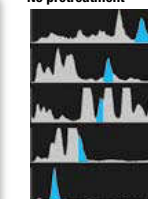
Sample : 10  $\mu$ L and I.S.  
 Dilute with 40  $\mu$ L of H<sub>2</sub>O  
 Add 100  $\mu$ L of B and mix

**MonoSpin TiO Sample loading**  
 ① Add the sample and centrifuge for 10 mins  
 ② Add the solution from column back into the column  
 Centrifuge for 10 mins and discard the waste fluid

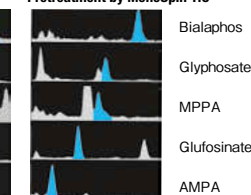
**Rinsing**  
 ① Add 20  $\mu$ L of B and centrifuge for 2 mins  
 ② Add 20  $\mu$ L of A and centrifuge for 2 mins  
 ③ Discard the waste fluid and set the recovery tubes

**Elution**  
 Add 20  $\mu$ L of C and centrifuge for 5 mins to recover the eluent solution

#### No pretreatment

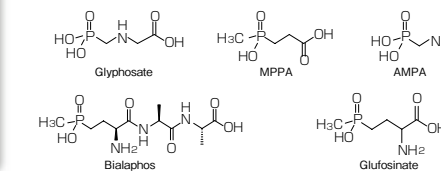


#### Pretreatment by MonoSpin TiO



#### Conditions

Column : C18 Column (150  $\times$  2.1 mm I.D.)  
 Eluent : CH<sub>3</sub>OH/20 mM HCO<sub>2</sub>NH<sub>4</sub> (pH 3.0) = 15/85  
 Flow Rate : 200  $\mu$ L/min  
 Detection : SIM  
 Injection Vol. : 5  $\mu$ L  
 Analyte : Bialaphos, Glufosinate, Glyphosate, AMPA (1ppm each)



## Recovery of basic drugs in serum (MonoSpin SCX)

Recovery of basic drugs in serum

**Solvents**  
 A (Coupling liquid) : 20 mM of potassium phosphate buffer (pH 7.0)  
 B (Eluent) : 5 % concentration ammonium water - methanol  
 C : A + drug : Propranolol hydrochloride (3.25 μL/mL)

All the centrifugal processes at 13,000 rpm

**Conditioning**  
 Set the waste fluid tube  
 Add 400 μL of A and centrifuge (1 min)

Sample : Serum 100 μL  
 + Add 200 μL of C

**MonoSpin SCX  
Sample loading**

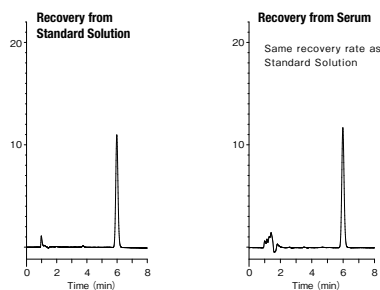
① Add the sample and centrifuge for 1 min  
 ② Discard the waste fluid

**Rinsing**

① Add 300 μL of A and centrifuge for 1 min  
 ② Discard the waste fluid and set the recovery tubes

**Elution**

Add 300 μL of B and centrifuge for 1 min to recover the eluent solution



**Conditions**  
 Column : Inertsil ODS-3 (150 × 4.6 mm I.D.)  
 Eluent : CH<sub>3</sub>CN / 5 mM KH<sub>2</sub>PO<sub>4</sub>, 52 mM SDS = 48/52  
 Flow Rate : 1.0 mL/min  
 Col. Temp : 40 °C  
 Detection : 230 nm  
 Injection Vol : 5 μL

## References

- Extraction of amphetamines and methylenedioxyamphetamines from urine using a monolithic silica disk-packed spin column and high-performance liquid chromatography-diode array detection. Namera A, Nakamoto A, Nishida M, Saito T, Yahata M, Yashiki M, Nagao M. *J Chromatogr A*. 24;1208(1-2):71-5.2008
- Monolithic spin column extraction and GC-MS for the simultaneous assay of diquat, paraquat, and fenitrothion in human serum and urine. Saito T, Morita S, Nakamoto A, Nishida M, Namera A, Nagao M, Inokuchi S. *J Chromatogr B Analyt Technol Biomed Life Sci*. 2008;872(1-2):186-90
- Simultaneous determination of amitraz and its metabolite in human serum by monolithic silica spin column extraction and liquid chromatography-mass spectrometry. Saito T, Yamamoto R, Inoue S, Nakamoto A, Nishida M, Namera A, Inokuchi S. *J Chromatogr B Analyt Technol Biomed Life Sci*. 2008;867(1):99-104.
- Monolithic silica spin column extraction and simultaneous derivatization of amphetamines and 3,4-methylenedioxyamphetamines in human urine for gas chromatographic-mass spectrometric detection. Nakamoto A, Nishida M, Saito T, Murakami K, Nagao M, Namura A. *Anal Chim Acta*. 2010 19;661(1):42-6
- Monolithic Spin-column Extraction and GC-MS Method for the Assay of Eperisone in Human Serum. Takeshi Saito, Takeshi Yamagiwa, Akihiro Nakamoto, Akira Namera, and Sadaki Inokuchi. *J. Health Sci.*, 56(5), 598-605, 2010
- Rapid Preparation Methods of Biological Samples for Ionic Compounds Using Ion Exchange Type Monolithic Silica Spin Column. Shigenori OHTA, Takeshi SAITO, Akira NAMERA and Masayoshi OHIR. *BUNSEKI KAGAKU* Vol.58 No.3., 213-218, 2010
- Monolithic spin column extraction and GC-MS for the simultaneous assay of diquat, paraquat, and fenitrothion in human serum and urine. Takeshi Saito, Tomokazu Fukushima, Akihiro Nakamoto, Akira Namera, Sadaki Inokuchi. *Anal Bioanal Chem*, 400(1), 25-31,2011
- Extraction of catecholamines from urine using a monolithic silica disk-packed spin column and high-performance liquid chromatography-electrochemical detection. Makoto Tsunoda, Takashi Funatsu. *Analytical Methods*, 3, 582-585, 2011
- Monolithic Spin Column: A New Extraction Device for Analysis of Drugs in Urine and Serum by GC/MS and HPLC/MS. Akira Namera, Masataka Nagao, Akihiro Nakamoto, Shota Takeshi Saito. *Journal of AOAC International* Vol. 94, No. 3, 2011
- A Convenient Purification Method for Pyridilamino Monosaccharides. Shunji Natsuka. *Biosci. Biotechnol. Biochem.* 75(7), 1405-1407, 2011

## Catecholamines in Urine (MonoSpin PBA)

Cis-diol compounds and catecholamines can be purified & enriched by MonoSpin PBA

**Solvents**  
 A (Rinsing) : 100 mM HEPES - NaOH (pH 8.5)  
 B (Coupling liquid) : 1.5 M HEPES - NaOH (pH 8.5)  
 C (Eluent) : 1 % acetic acid

All the centrifugal processes at 10,000 rpm

**Conditioning**  
 ① Set waste fluid tubes, put 500 μL of C and centrifuge for 1 min  
 ② Put 500 μL of A and centrifuge for 1 min  
 ③ Discard the waste fluid

Sample : 450 μL. Add 50 μL of B 3,4-Dihydroxybenzylamine (I.S.) 10 μL

**MonoSpin PBA  
Sample loading**

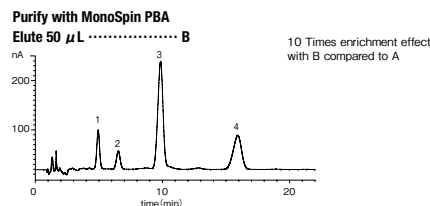
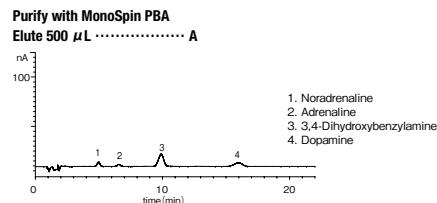
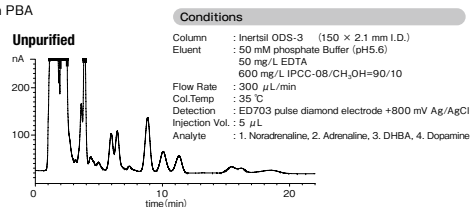
① Add the sample and centrifuge for 1 min  
 ② Discard the waste fluid

**Rinsing**

① Add 500 μL of A and centrifuge for 1 min  
 ② Discard the waste fluid and set the recovery tubes

**Elution**

Add 50 - 500 μL of C and centrifuge for 1 min to recover the eluent solution



## Specifications

Description	Specification
Packing material	Silica Monolith (Highly pure silica gel)
Through Pore Diameter	5 μm
Meso Pore Diameter	100 Å (10 nm)
Surface Area	350 m <sup>2</sup> /g
Sample Volume	50 - 800 μL

## MonoSpin™ Part Numbers

Description	Quantity	Cat.No.
MonoSpin C18	50 pcs	5010-21700
	100 pcs	5010-21701
MonoSpin Amide	50 pcs	5010-21727
	100 pcs	5010-21728
MonoSpin CBA	50 pcs	5010-21729
	100 pcs	5010-21730
MonoSpin NH2	50 pcs	5010-21710
	100 pcs	5010-21711
MonoSpin SCX	50 pcs	5010-21725
	100 pcs	5010-21726
MonoSpin SAX	50 pcs	5010-21720
	100 pcs	5010-21721
MonoSpin PBA	50 pcs	5010-21715
	100 pcs	5010-21716
MonoSpin TiO	50 pcs	5010-21705
	100 pcs	5010-21706

\*Based on monolithic technology, Merck KGaA, Darmstadt, Germany