

Phos-tag application data

Analysis of the phosphorylated species of β -casein
 ~ Mn^{2+} -Phos-tag SDS-PAGE with Laemmli system ~

● SAMPLE INFORMATION

		MW (kDa)
Protein	β -casein	24
Protein status	normal	-

● ELECTROPHORESIS CONDITION

Gel	10% polyacrylamide
Phos-tag conc.	100 μ M Mn^{2+} - Phos-tag
Metal complex	

Visualization	N/A
Antibody	N/A

● ASSAY FLOW

- 1 AP treatment of protein
- 2 Phos-tag electrophoresis

● RESULT

In Mn^{2+} -phos-tag gel, isoforms were separated by difference of phosphorylated site, not by degree of phosphorylation per one molecule.

● NOTE

Another data of simultaneous analysis : 002-2

● REFERENCE

Two-dimensional phosphate-affinity gel electrophoresis for the analysis of phosphoprotein isotypes. Kinoshita E, Kinoshita-Kikuta E, Matsubara M, Aoki Y, Ohie S, Mouri Y, Koike T. : *Electrophoresis* , **30**, 550, (2009)

key words : Mn^{2+} -Phos-tag, Laemmli, β -casein

Phos-tag application data

Analysis of the phosphorylated species of β -casein
 ~ 2-DE coupling urea-PAGE and Mn^{2+} -Phos-tag SDS-PAGE ~

● SAMPLE INFORMATION

		MW (kDa)
Protein	β -casein	24
Protein status	normal	-

● ELECTROPHORESIS CONDITION

Gel	1D: 4.0M urea + 6.0% polyacrylamide (without Phos-tag) 2D: 10% polyacrylamide
Phos-tag conc.	100 μ M Mn^{2+} - Phos-tag
Metal complex	

Visualization	N/A
Antibody	N/A

● ASSAY FLOW

- 1 AP treatment of protein
- 2 Urea-PAGE as the first dimension
- 3 Phos-tag electrophoresis as the second dimension

● RESULT

- The separation of phosphoprotein isotype should be improved relative to those by current gel electrophoresis method.
- Isoforms having same number of phosphorylation site were separated to different bands by this analysis.

● NOTE

Another data of simultaneous analysis : 002-1

● REFERENCE

Two-dimensional phosphate-affinity gel electrophoresis for the analysis of phosphoprotein isotypes. Kinoshita E, Kinoshita-Kikuta E, Matsubara M, Aoki Y, Ohie S, Mouri Y, Koike T. : *Electrophoresis* , **30**, 550, (2009)

key words : Mn^{2+} -Phos-tag, Laemmli, β -casein, Urea-PAGE, 2D-PAGE