日本電気泳動学会 電子メール通信 No. 214

2024 年 11 月 20 日

【お知らせ__Journal of Electrophoresis Vo. 68 (2024) No.1　J-STAGE からの公開】

日本電気泳動学会会員の皆様

2024 年 9 月 4 日、Journal of Electrophoresis Vo. 68 (2024) No.1 (J-STAGE 電子版)に、以下の論文が掲載されましたのでお知らせ致します。ご連絡が遅れましたことお詫び申し上げます。
（https://www.jstage.jst.go.jp/browse/jelectroph）

J Electrophoresis. 2024; 68:1–6.

Full Paper

Title: Characterization of constitutively hyperphosphorylated lamin A in vivo using Phos-tag SDS-PAGE

Authors: Emiko Kinoshita-Kikuta, Kaku Shimoji, Kento Hiraishi, Kento Nishikawa, Kenichi Nagase, Tohru Koike, Eiji Kinoshita

Abstract: The phosphorylation states of proteins are specifically regulated by kinases and phosphatases. In general, the reactions are reversible, and the phosphorylation states are highly dynamic. However, there are many proteins that maintain highly phosphorylated states, and have multiple phosphorylated species. Among such proteins, we focused on human lamin A, an intermediate filament, and identified amino acid residues that contribute to the generation of multiple phosphorylated species. Using Phos-tag SDS-PAGE followed by immunoblotting, we analyzed a FLAG(DYKDDDDK)-tagged lamin A and 27 of its potential site-directed phosphorylation mutants transiently expressed in HeLa cells. This Phos-tag-based strategy, which exploits information on the phosphorylation sites deposited in the PhosphoSitePlus database, revealed that lamin A is constitutively phosphorylated at residues Ser-22, Thr-24, Ser-390, Ser-392, Ser-404, and Ser-406, and that combinations of phosphorylation at these residues generate at least six phosphorylated variants. This study showed that six residues, including the

well-studied mitotic phosphorylation site, are constitutively phosphorylated in interphase and that lamin is regulated by phosphorylation throughout the cell cycle. In contrast to proteomic discovery-mode MS strategies, Phos-tag SDS-PAGE is a method used to detect the species with relatively high phosphorylation stoichiometry. It also has a unique advantage: it allows us to identify variants with different phosphorylation states under biological conditions without fragmenting proteins into peptides and at the same time without losing important information on the molecular weights of these proteins. It is therefore useful for studying the characteristic functions of various native proteins and their quantitative changes in phosphorylation state during cellular processes.

J Electrophoresis. 2024; 68:7–13.

Full Paper

Title: Proteome analysis of murine organs using Auto2D

Authors: Takuya Ono, Yuto Ono, Tadashi Kondo

Abstract: Two-dimensional polyacrylamide gel electrophoresis (2D-PAGE) is a widely used proteomic tool that separates proteins based on the isoelectric points and molecular weights, allowing the observation of hundreds to thousands of proteins in a gel image. Because the protocol for the protein extraction and 2D-PAGE vary between laboratories, data comparison and integration across laboratories are challenging. By standardizing the protocol, collaborative research using the 2D-PAGE can be conducted across multiple laboratories. This study aimed to establish the standardized protocol for the protein extraction and 2D-PAGE. Using an automated 2D-PAGE machine, Auto2D, 620 common spots were identified across protein samples extracted from murine eight organs. Hierarchical cluster analysis based on the intensity of the spots revealed that samples formed groups according to the respective organs. Furthermore, correlation analysis yielded correlation coefficient of ≥0.75 for samples from the same organ. Principal component analysis (PCA) clearly separated samples into distinct clusters, which corresponded to different types of organs. Cluster analysis, correlation analysis, and PCA demonstrated that data was reliably reproducible using the same protein extraction protocol, indicating no need to change the protocol for the protein extraction for each organ. With the standardized protocol for the protein extraction and 2D-PAGE developed in this study, collaborative proteome analysis across multiple laboratories is feasible, enabling reliable data comparison and integration between laboratories and samples.

なお，日本電気泳動学会では学会英文機関誌（Journal of Electrophoresis）への論文投稿を広く募集しております。また Case Reports（英文誌）、症例報告（和文誌）の論文種目もございます。会員の皆様の積極的なご投稿を期待しております（会員であれば、投稿料は無料です）。

日本電気泳動学会　会長
亀山　昭彦

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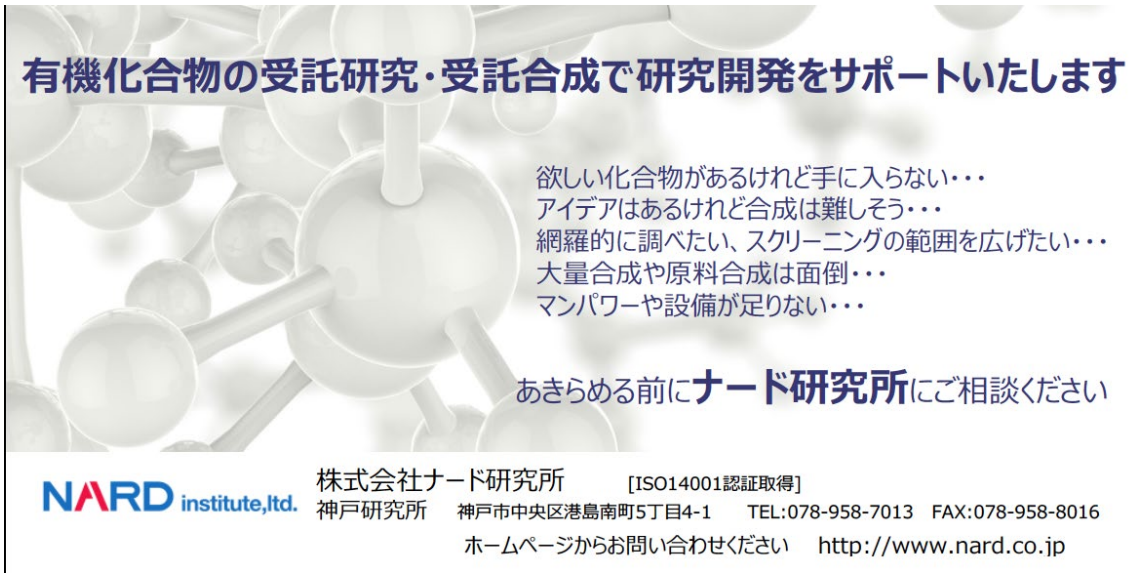

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