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Speciation of Some Biometals in Salmon Egg Cell Cytoplasm by Two-Dimensional HPLC/ICP-MS

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Abstract:

A two-dimensional (2D) HPLC/ICP-MS hyphenated system was developed and applied to speciation of biometals in salmon egg cell cytoplasm. The 2D HPLC was based on the combination of surfactant-mediated HPLC and size exclusion chromatography (SEC). In the present experiment, an ODS column coated with CHAPS (3-[(3-cholamidopropyl) dimethylammonio]-1-propanesulfonate), which is a zwitterionic bile acid derivative, was employed as a separation column in the first-dimensional surfactant-mediated HPLC. The surfactant-mediated HPLC allowed simultaneous separation of large and small molecules. In the second-dimensional HPLC, proteins thus separated from inorganic salts in the first dimension were further separated by the SEC column. The present 2D HPLC/ICP-MS system was available for the improvement of resolution in SEC separation of metalloproteins in salmon egg cell cytoplasm. Metalloproteins including Fe and Zn in salmon egg cell cytoplasm were detected as only one large peak at the volume of total exclusion limit of SEC column (>300 kDa), when analyzed by SEC/ICP-MS. However, these metalloproteins could be detected as two peaks in the present 2D HPLC/ICP-MS measurements; the molecular weights of Fe-proteins were >300 kDa and 100 kDa, and those of Zn-proteins were >300 kDa and 150 kDa.

Key words: <u>metalloprotein</u>, <u>salmon egg cell cytoplasm</u>, <u>speciation</u>, <u>two-dimensional</u> <u>HPLC</u>, <u>desalting</u>



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