

Determination of Selenium in Human Serum by LC/APCI-MS after Acid Digestion and Organic Derivatization using 2,3-Diaminonaphthalene

Masayuki Ando, Megumi Takizawa, Susumu Yamato, Kenji Shimada

Department of Analytical Chemistry, Niigata University of Pharmacy and Applied Life Sciences, 5-13-2 Kamishinei-cho, Niigata 950-2081, Japan

Measurement of serum selenium has recently been the subject of increasing interest because of the toxicological and nutritional importance of this element. Numerous reviews of current methodologies for determining selenium levels have been published. One of the most convenient and widely used methods for routine determination of serum selenium is the fluorometric method using 2,3-diaminonaphthalene (DAN), although this method lacks specificity.

We observed that 4,5-benzopiazselenol (BPS), which is a selenium derivative of DAN, is ionized with electron capture in an atmospheric pressure chemical ionization (APCI) interface, and we subsequently established a method for determining total human serum selenium by means of LC/APCI mass spectrometry.

EXPERIMENTAL

Apparatus and conditions. LC/MS analyses were performed on a Hitachi M-1000 LC/APCI mass spectrometer connected to a Hitachi L-6200 HPLC pump (Hitachi, Tokyo, Japan). The separation column was an Inertsil ODS-2 (150 mm \times 4.6 mm i.d., 5 μ m, GL Sciences, Tokyo, Japan) and methanol-water (85:15, v/v) used as the mobile phase (flow rate, 0.75 mL/min). The base molecular anion m/z 234 was monitored in SIM mode.

Determination of total serum selenium. Human serum (0.5 mL) was digested with mineral acids. The selenium(IV) derivative of 2,3-diaminonaphthalene was formed and was then extracted into 0.5 mL of cyclohexane. All procedures were carried out in a single test tube in order to minimize loss of selenium. An aliquot of 0.05 mL of the organic layer was injected into the LC-MS system.

RESULTS AND DISCUSSION

Some compounds were determined in LC/MS with electron capture ionization using an APCI interface in the negative mode.^{1,2} BPS formed a radical molecular ion in the APCI interface, and other pseudomolecular ions and fragment ions were not observed. The detection limit of BPS was equivalent to 0.2 ng of selenium. The lower limit of quantification of serum selenium was 10 ng/mL. The coefficient of variation of determined concentrations in control serum samples was less than 5%. The purity of the observed peak obtained from serum samples was checked using the ion cluster technique.

The proposed method allows the routine determination of selenium levels in serum samples with high specificity.

REFERENCES

- 1) G. Singh, A. Gutierrez, K. Xu and I. A. Blair, *Anal. Chem.*, 2000, 72, 3007.
 - 2) H. Hayen, N. Jachmann, M. Vogel and U. Karst, *Analyst*, 2002, 127, 1027.
-