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Development of an analytical method for beryllium in airborne dust by micellar electrokinetic chromatography

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Abstract

The separation of diacetylacetonato–beryllium [$\text{Be}(\text{acac})_2$] from acetylacetone was achieved with micellar electrokinetic chromatography (MEKC) followed by subsequent beryllium analysis carried out using capillary electrophoresis. Analysis using a separation and absorption detector, with a 50 μm capillary cell, could detect approximately 1000 ppb of beryllium in the final sample. $\text{Be}(\text{acac})_2$ is a promising reagent for absorption spectrophotometry, because its molar absorption coefficient, (32 000 $\text{l mol}^{-1} \text{cm}^{-1}$) is very large. As the complex and acetylacetone have a similar absorption wavelength, the excess free ligand must be removed from the sample to be measured. Acetylacetone is a weak acid with keto–enol tautomerism in aqueous solution. Acetylacetone and neutral $\text{Be}(\text{acac})_2$ should be separated with capillary zone electrophoresis (CZE) using a neutral or basic buffer solution as the mobile phase. Although the pH and temperature of the mobile phase were optimized, separation with CZE was interfered with by a portion of acetylacetone. This interfering portion seems to be a neutral keto-form with the same migration time as $\text{Be}(\text{acac})_2$. As a neutral species separation method, MEKC with sodium dodecyl sulfate was tried and the separation was completed. The optimum pH value and buffer temperature are pH 7.8 and 15°C, respectively. © 1999 Elsevier Science B.V. All rights reserved.

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1. Introduction

Beryllium (Be) is known as the most toxic element without radioactivity [1–3]. Occupational exposure to Be causes many kinds of diseases, such as berylliosis, which is a pneumoconiosis caused by beryllium-containing dust inhalation, lung cancer and immunological diseases by competitively interfering with magnesium. However, beryllium is an appealing material for many industries. Besides the chemically unstable element lithium, it is the most lightweight metal. It also has superior properties as an accessory ingredient of copper or nickel alloys. In spite of its high toxicity, beryllium has been used for many

purposes, such as the windows of X-ray tubes as Be metal, golf clubs and anti-static tools, as beryllium bronze and dental metal with nickel. All workers treating beryllium in the workplace face occupational exposure to toxic beryllium. However, the risks faced by dentists and dental laboratory technicians are greater due to their larger population, and the nature of their work [4–6]. The abrading and grinding of beryllium-containing dental compounds, produces inhalable dust hence increasing the risks.

To control the risks, beryllium concentration in workplace air is measured by fluorescence measurements in Japan [7]. The fluorescence method was the only method that certifies a required sensitivity when the current Japanese regulation of working environment measurement was established in 1975. How-

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ever, the method is so complex and has many problems. The first problem is the requirement of a highly skilled analyst in order to maintain accuracy. Secondly a lot of waste is produced during the analysis. As well as protection of the workers health, high risk beryllium pollution due to analysis should also be avoided. The last and most serious problem is the danger of exposure of toxic beryllium to industrial hygienists through analysis itself. Accordingly, I investigated a safe, clean and easy method for beryllium analysis. New methods such as flameless atomic absorption spectrometry (AAS) and/or inductively couple plasma atomic emission spectrometry (ICP-AES) could be employed in our regulation of beryllium in workplace air, but the author believes ion chromatography (IC) and capillary electrophoresis (CE) are more suitable methods to decrease the risk of beryllium exposure to the industrial hygienist. CE is operated with a small amount of electrophoresis buffer solution and it produces only small amounts of beryllium-containing toxic waste. Most of the procedure can be automated safely. In the present work, the author will report the first step of the development of a beryllium measurement method, with CE, for solution samples. This is a model for digested airborne dusts. Diacetylacetonato–beryllium complex $[\text{Be}(\text{acac})_2]$ is a promising reagent of absorption spectrophotometry, because it has suitable properties for analysis: (1) large molar absorption coefficient $32\,000\text{ l mol}^{-1}\text{ cm}^{-1}$ [8] and (2) large complexing coefficient 10^{18} M^{-1} [9].

Moreover the complexing coefficient is considerably larger than that of Be and ethylenediaminetetraacetic acid (EDTA) which is different from other metals. Therefore interfering metal cations would be masked by EDTA. However, there is a problem using $\text{Be}(\text{acac})_2$ for spectrophotometry. $\text{Be}(\text{acac})_2$ has the same absorption wavelength as free acetylacetonone. The excess free ligand should be removed completely from the measurement cell. Previously, the free acetylacetonone was extracted by a strong alkaline solution such as enol-form weak acid. The procedure is not practical, because it requires many operation steps, and the loss of the analyte is possible. Therefore, the author has studied the separation of $\text{Be}(\text{acac})_2$ from free excess acetylacetonone using CE. Two separation modes – capillary zone

electrophoresis (CZE) and micellar electrokinetic chromatography (MEKC) – were investigated, the reason being that we do not know whether acetylacetonone is mainly in the keto-form neutral molecule or the enol-form anion in the analytical cell.

2. Experimental

2.1. Reagents

All reagents were used without further purification. Running buffer for CE (pH 6 and pH 10) and sodium dodecyl sulfate (SDS) were purchased from Fluka (Neu-Ulm, Switzerland). Acetylacetonone was purchased from Fluka and Dojin Kagaku (Kumamoto, Japan) and all other reagents were purchased from Wako Pure Chemicals Industry (Osaka, Japan). Deionized, charcoal-treated and distilled water was obtained using a WF-12 (Yamato Kagaku, Tokyo, Japan) quartz boiler two-step distillation system.

2.2. Capillary electrophoresis

All CE experiments were carried out on a HP^{3D}CE G-1600 (Hewlett-Packard, Waldbronn, Germany) system. The capillary dimensions were of effective length $560\text{ mm} \times 50\text{ }\mu\text{m}$ I.D. The applied voltage was 30 kV. Other conditions will be described with each set of results. The samples were injected by pressure method. The applied pressure was 5 Pa and pressure adding time was 5 s except for huge sample load experiments.

2.3. Sample preparations

One handled ppm in 0.05 M nitric acid standard beryllium solution for AAS (Wako) was used as a start solution for model sample solution on the assumption that digestion by nitric acid and hydrogen peroxide was used for pretreatment method of workplace air sample. All sample solutions were prepared by dilution of the start solution with running buffer.

2.4. Buffer preparations

Prepared 3-cyclohexylamino-1-propanesulfonic acid (CAPS; 20 mM) buffer solution for pH 10 running buffer and 20 mM sodium citrate solution for pH 6.0 running buffer were purchased and used without further modification. The pH 7.8 running buffer solution was prepared by pH adjustment with injection of nitric acid to 0.1 M Tris solution.

3. Results and discussion

3.1. Separation with CZE

Fig. 1 shows the effects of pH in separating acetylacetonone and $\text{Be}(\text{acac})_2$ by CZE. At pH 6 and pH 10, acetylacetonone and $\text{Be}(\text{acac})_2$ were detected together as a single peak and at pH 7.8, though it was still insufficient, their peaks were detected separately. At pH 6, a sharp single peak was detected i.e., $\text{Be}(\text{acac})_2$ and acetylacetonone had a similar migra-

tion time and the transform equilibrium of acetylacetonone was one-sided. At pH 10, the peak shape broadened. Therefore acetylacetonone was in existence as the mixture of the keto and enol form and was passed through the capillary between the keto- and enol-form acetylacetonone. These results coincide with the trend of chemical equilibrium of keto-enol tautomerism of acetylacetonone and an increase in pH was expected to improve the separation. However, a higher pH accelerates the electroosmotic flow (EOF) resulting in a lower separation efficiency.

To improve separation of acetylacetonone and $\text{Be}(\text{acac})_2$, the capillary temperature was changed (Fig. 2). Fig. 3 shows the electropherogram in which the concentration of beryllium ranged from 0 to 10 ppm. Two peaks were observed in the samples without Be. To rule out suspicion of contamination of metals or organic substances, acetylacetonone from the two different companies was used in the additional experiments. But the same results appeared. As for Dojin-Kagaku's acetylacetonone which was

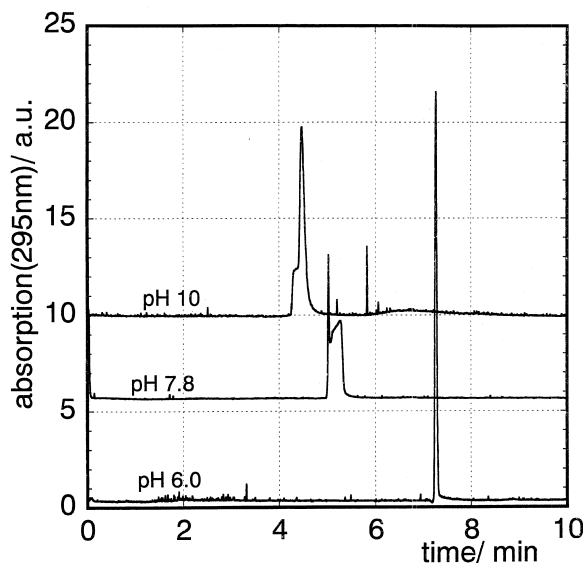


Fig. 1. The effect of running buffer pH on the separation of acetylacetonone and $\text{Be}(\text{acac})_2$. The buffers were 20 mM sodium citrate, pH 6, 0.1 M Tris–nitrate, pH 7.8 and 20 mM CAPS, pH 10. Capillary dimensions: effective length 560 mm \times 50 μm I.D.; capillary temperature, 20°C; applied voltage 30 kV. The samples were injected by pressure method. The applied pressure was 5 Pa and pressure adding time was 5 s.

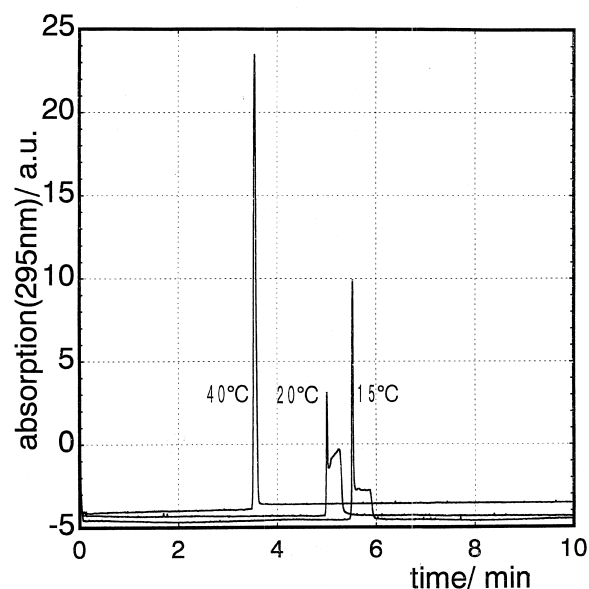


Fig. 2. The effect of the capillary on separation of acetylacetonone and $\text{Be}(\text{acac})_2$. Running buffer, 0.1 M Tris–nitrate, pH 7.8; capillary dimensions: effective length 560 mm \times 50 μm I.D.; applied voltage, 30 kV. The samples were injected by pressure method. The applied pressure was 5 Pa and pressure adding time was 5 s.

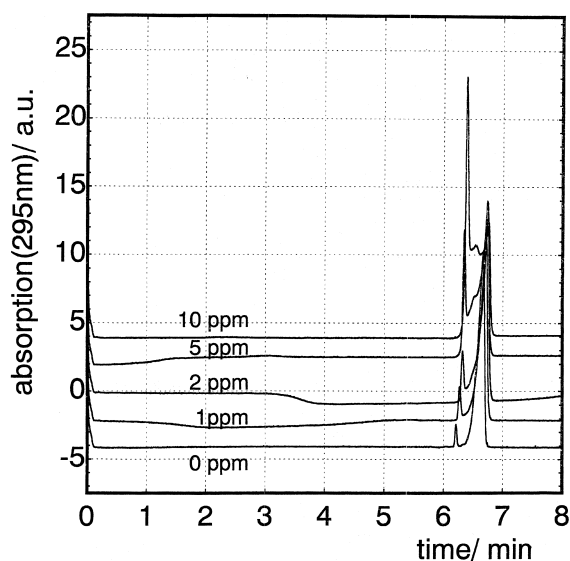


Fig. 3. Electropherograms of various concentrations of Be. Capillary temperature, 15°C; running buffer, 0.1 M Tris–nitrate, pH 7.8; capillary dimensions: effective length 560 mm×50 μm I.D.; applied voltage, 30 kV. The samples were injected by pressure method. The applied pressure was 5 Pa and pressure adding time was 5 s.

diluted to one-tenth with purified water, semi-qua ICP-AES analysis could not detect metals within the sensitivity measurement. It is likely that the double peaks detected at 0 ppm of Be are the keto- and enol-acetylacetonone isomers, respectively.

In the CZE separation method, some of the acac is extracted and detected at the same migration time as $\text{Be}(\text{acac})_2$. The peak height of extracted acetylacetonone was approximately similar to the peak height of 1 ppm Be. In accordance with an increase of the concentration of Be, the acetylacetonone peak lost its shape due to the imperfect separation. This means that CZE is unable to analyze Be although the reason is not known.

3.2. Separation with MEKC

In CZE, CE does not separate neutral molecules such as keto-acetylacetonone and $\text{Be}(\text{acac})_2$ from each other. Therefore, all acetylacetonone enol form is required to be kept for $\text{Be}(\text{acac})_2$ analysis with CZE. However, this is impossible. Accordingly, their neutral species should be separated from each other

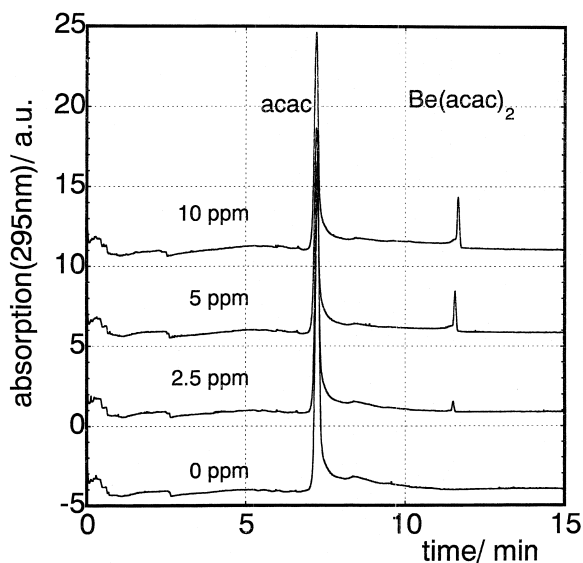


Fig. 4. Electropherograms of various concentrations of Be under optimized MEKC conditions. Capillary temperature, 10°C; running buffer, 0.1 M Tris–nitrate, 50 mM SDS, pH 7.8; capillary dimensions: effective length 560 mm×50 μm I.D.; applied voltage, 30 kV. The samples were injected by pressure method. The applied pressure was 5 Pa and pressure adding time was 5 s.

for $\text{Be}(\text{acac})_2$ detection. MEKC is the method of CE for neutral molecules. Fig. 4 shows the electropherograms of various concentrations of Be using MEKC separation with 50 mM SDS as micellar reagent under optimized pH and temperature. The optimum pH is 7.8 with lower temperatures resulting in good separation. Although the mechanisms of separation are different, the optimum values of pH and capillary temperature were the same. These results show simple coincidences, and could be controlled by the chemistry of acetylacetonone, such as the ratio of keto and enol isomers, transform rates, complexing rates, etc. This very interesting result could be used both for optimizing Be analysis by CE for practical use and for theoretical studies on acetylacetonone chemistry. With MEKC the separation is improved, particularly, because the migration times of $\text{Be}(\text{acac})_2$ and acetylacetonone are exceedingly different from each other. Therefore, some high sensitivity oriented to devices such as huge sample loading and using long cell detection could be applicable. The effect of sample load is shown in Fig. 5. With 10-times the normal levels of $\text{Be}(\text{acac})_2$

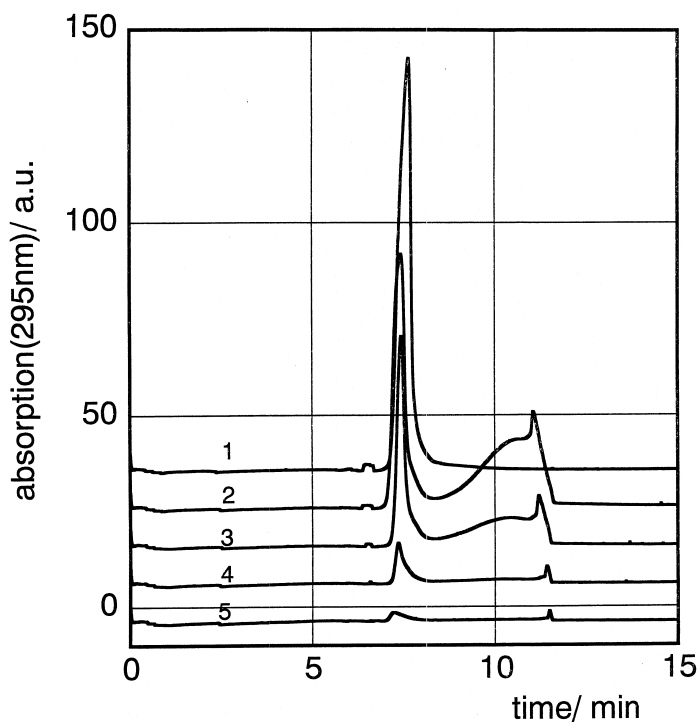


Fig. 5. Electropherograms of various amounts of loaded sample. Capillary temperature, 10°C; running buffer, 0.1 M Tris–nitrate, 50 mM SDS, pH 7.8; capillary dimensions: effective length 560 mm×50 μm I.D.; applied voltage, 30 kV. The samples were injected by pressure method. The applied pressure was 5 Pa. The pressure adding times were (5) 5 s, (4) 10 s, (3) 30 s, (2 and 1) 50 s. The sample concentrations were 10 ppm except curve 1. Curve 1e is blank solution.

overflowed from the SDS micellar phase. However, free acetylacetone shows a sharp peak. Therefore, low beryllium sample could be applicable to load huge sample. Fig. 6 shows the concentrations of various Be samples on condition that the volume injected was 10-times greater than before. An increased injection volume did not improve the limit of quantitative analysis. At the 100 ppb level no peak of Be(acac)₂ was observed at all while it was difficult to distinguish peaks from noise at 250 ppb and 500 ppb levels. At 1000 ppb a Be(acac)₂ peak was clearly detected. Japanese standards regulate the concentration of Be in workplace air should be under 0.002 mg/m³. Under the current authorized procedures of workplace air sampling and pretreatment methods, industrial hygienists would need 5 m³ air to obtain the desired concentration of 1000 ppb by diluting the sample solution. Moreover, the Japanese standards adopt the environmental concentration in controlling workplace environment instead of concentration of personal exposure. In the described procedure, given

the sampling time of 10 to 30 min the volume of 5 m³ cannot to be collected in such a short period. However the current procedures of measurement require the final sample solution volume of 100 ml while CE requires about 100 μl. So, as the current methods are improved it would be possible to raise the substantial sensitivity by 1000-fold and to apply CE to beryllium analysis in workplace air. Therefore to apply this method for the actual workplace, it needs to be further improved.

4. Conclusions

A clean, safe and easy beryllium measurement method is needed for industrial hygiene and CE is a promising technique. Be(acac)₂ can be separated from free acetylacetone by both CZE and MEKC. However, MEKC was superior to CZE for the separation of Be(acac)₂ and acetylacetone. This chemical system should be useful for not only

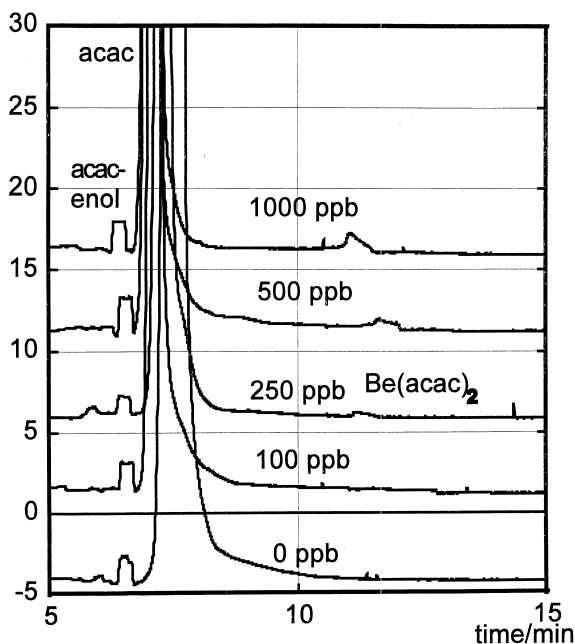


Fig. 6. Electropherograms of various concentrations of Be under optimized MEKC conditions with huge sample loading. The concentrations of samples are (a) 0, (b) 100, (c) 250, (d) 500 and (e) 1000 ppb. The samples were injected by pressure method, 5 Pa, 50 s. Capillary temperature, 10°C; capillary dimensions: effective length 560 mm×50 μm I.D.; applied voltage, 30 kV.

practical beryllium measurement, but also for the theoretical study of tautomerism equilibrium chemistry.

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