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Investigation of vanadate as a pH sensitive analyte anion using capillary zone electrophoresis

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ABSTRACT

Inorganic anions were determined using capillary zone electrophoresis with UV detection. 1,2-Dihydroxybenzene-3,5-disulphonic acid disodium salt (Tiron) was used as the electrolyte. The determination of vanadate as the analyte ion resulted in two peaks which depended on the pH of the analyte solution. The two vanadate peaks showed opposite behaviour. A pH range of 2.3–11.8 was investigated. The relative standard deviation of the peak areas was 8%. Spectroscopic investigations showed that the vanadate anions interact with the electrolyte.

INTRODUCTION

Capillary zone electrophoresis (CZE) is being used to an increasing extent for the determination of inorganic cations and anions [1–8]. In addition to the determination of the ions of interest, the simultaneous determination of the pH of the analyte solution is desirable, especially in routine analysis, so that no additional pH measurement is necessary. When only small sample volumes are available, the determination of either the ions or the pH value is possible, but not both. Even when using microelectrodes, measurement of the pH of samples in a volume smaller than 1 μl is not feasible [9].

In principle, two approaches are possible to determine the pH of the analyte solution integrated in the electrophoretic process. The first is by the determination of the protons together with the analyte cations. However, problems arise because of the high ionic mobility of the protons in comparison with those of the electrolyte and the analyte cations. Further, the proton concentration will be changed by a factor of 10 per pH unit. This implies a system having low limits of detection for the protons (*e.g.*, 1 μM H^+ at pH 6.0).

On the other hand, the determination of pH is possible by adding a pH-sensitive species to the analyte solution. The species must fulfil the condition of showing a sensitive pH dependence of detector response (*e.g.*, absorbance), which must remain in a steady state until detection. Further, the species should not co-migrate with the ions of interest. Isopolyanions seem to be an interesting group of species that can be used as analyte additives for pH determination. Our experiments with different isopolyanions show that vanadate is the most pH-sensitive anion.

In aqueous solution, vanadates undergo a series of complex hydrolysis–polymerization reactions in a system of changing pH value [10,11]. The different pH-dependent species show different UV absorptivities [12].

In this work, the determination of vanadate at

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different pH values and concentrations was investigated. The peak area of the observed two vanadate signals depends on the pH of the analyte solution. 1,2-Dihydroxybenzene-3,5-disulphonic acid disodium salt (Tiron) was used as an electrolyte and UV detection was carried out.

EXPERIMENTAL

A Dionex (Idstein, Germany) CES 1 capillary electrophoresis instrument was used for analysis. UV detection was carried out at 293 nm. A high voltage of 25 kV with a negative voltage mode was applied. The separations were carried out using conventional fused-silica capillaries from Scientific Glass Engineering (Weiterstadt, Germany). The dimensions of the capillaries were 75 μm I.D. and 60 cm total length, with 55 cm from the point of injection to the detector cell. Before starting the separation the capillaries used were washed with the elecrolyte. Hydrostatic injection (10 cm, 30 s) and electrokinetic injection (3 kV, 10 s) were used for sample introduction.

All solutions were prepared each day from 50 m*M* stock solutions, filtered through a 0.22-μm membrane from Millipore (Eschborn, Germany) and degassed under vacuum for 10 min.

Spectroscopic investigations were carried out on a Hewlett-Packard model (Bad Homburg, Germany) 8451A diode-array spectrophotometer.

Chemicals

All solutions, standards and the electrolyte were prepared with purified water obtained from a Milli-Q system (Millipore). 1,2-Dihydroxybenzene-3,5-disulphonic acid disodium salt (99%), 2,5-dihydroxy-1,4-benzenedisulphonic acid dipotassium salt (98%) and 1,3-benzenedisulphonic acid disodium salt (90%) was obtained from Aldrich (Steinheim, Germany). Sodium metavanadate ($NaVO_3$) (98%) (Fluka, Neu-Ulm, Germany) was used for the preparation of the vanadate standards. All other reagents were of analytical-reagent grade from Merck (Darmstadt, Germany).

RESULTS AND DISCUSSION

Fig. 1 shows an electropherogram of an anion standard solution including vanadate. The van-

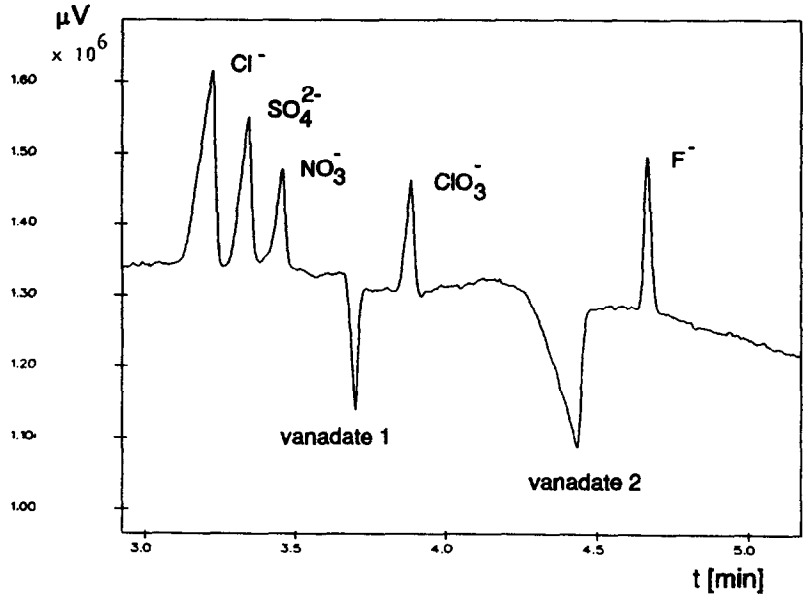

Fig. 1. Electropherogram of an anion standard including vanadate. Concentration of each anion = 0.1 m*M*, except for vanadate (0.4 m*M*). Electrolyte, 5 m*M* 1,2-dihydroxy-3,5-disulphonic acid disodium salt, (pH 5.3); capillary, fused silica (60 cm total length, 55 cm to the detector, 75 μm I.D.); voltage, −25 kV; detection, UV (293 nm), range 0.01 absorbance; injection hydrostatic (30 s, 10 cm).

adate anion gives two positive signals without co-migration with the main anions chloride, nitrate and sulphate. The second vanadate peak is extremly broad and shows fronting, possibly due to the adsorption of the vanadate species by the capillary surface, which results in peak distortion [13,14].

The vanadate samples were measured at different times after sample preparation. In Table I the ratio of the peak areas of the two vanadate peaks and the time after sample preparation (from 15 min to 10 h) are summarized. As can be seen, the ratio of the peak areas shows no significant changes. The same results were achieved by measurement of vanadate using different capillary lengths.

Experiments with molybdate and tungstate as analyte additives did not show the same significant pH dependence as vanadate. Niobate and tantalate, which also form isopolyanions, were not investigated because both species show precipitation at pH 7 [10].

Both vanadate peaks have an interesting dependence on the pH of the analyte solution, as demonstrated in Fig. 2a where three electropherograms of a vandate standard (0.4 m*M*) with different pH values of the analyte solution are shown.

At acidic pH (3.5) (Fig. 2a), the first vanadate peak shows a greater absorbance than the second peak. At higher pH (5.3), both peaks have nearly the same height (Fig. 2b). The second peak becomes dominant when the pH of the analyte solution is increased to 8.8 (Fig. 2c).

This pH-dependent behaviour is clearly demonstrated in Fig. 3, which shows the relationship between the pH of the analyte solution in the range 2.3–11.8 and the areas of the vanadate peaks. The two vanadate peaks show opposite behaviour.

The opposite behaviour of the vanadate peaks as demonstrated in Fig. 2 can be seen at pH 3.8. The first vanadate peak increases with decreasing pH whereas the second peak decreases. In the acidic pH region a stronger pH dependence of the peak areas can be observed than at pH > 5.0, where the peak areas are change only slightly with change in pH. At pH 11 only the second peak can be observed, the first peak having disappeared. The observed migration times of both peaks were constant over the pH range investigated.

The ratio of the peak areas (peak 2/peak 1) of the vanadate peaks as a function of pH is shown in Fig. 4. With increasing pH from pH 3.8 the peak-area ratio increases, whereas at lower pH the peak-area ratio decreases. This behaviour restricts the application of vanadate as an analyte additive at this concentration to samples with pH >4.0.

With fifteen consecutive injections by hydrostatic injection of a vanadate standard (0.4 m*M*), the relative standard deviation (R.S.D.) of the peak areas was 8%. The limit of detection for vanadate depends on the pH and is about 40 μM.

Measurements were also carried out with other Tiron and vanadate concentrations. Experiments with Tiron concentrations from 1 to 9 m*M* (at a constant pH of 5.3) and a vanadate

TABLE I

RATIO OF THE PEAK AREAS (VANDATE 2/VANADATE 1) DEPENDING ON THE TIME AFTER SAMPLE PREPARATION

pH of the vanadate solution = 6.5. Experimental conditions as in Fig. 1.

Time after sample preparation [min]	Ratio of peak areas
15	3.3
45	3.1
85	3.2
100	3.4
124	3.6
150	3.4
161	3.1
172	3.2
180	3.3
189	3.2
203	3.4
232	3.1
290	3.0
361	3.6
404	3.2
455	3.3
541	3.5
620	3.6
629	3.2

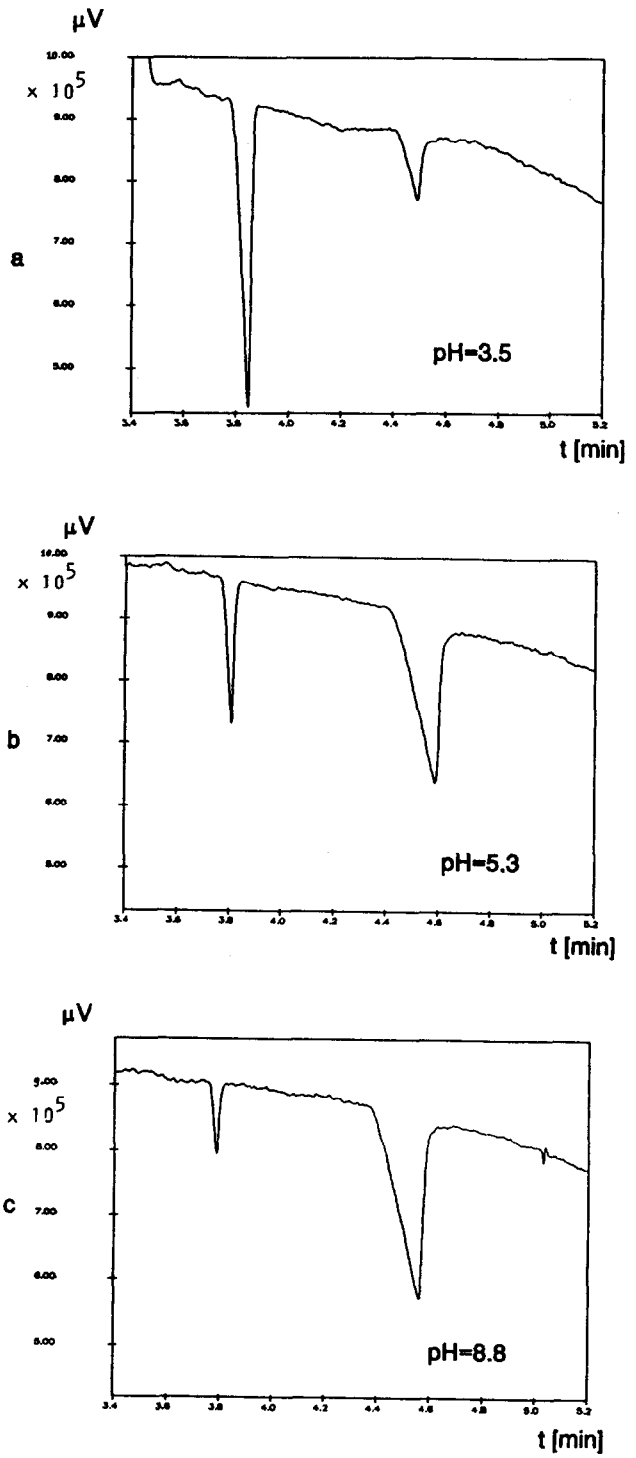

Fig. 2. Electropherograms of vanadate at different pH values of the analyte solution: (a) 3.5; (b) 5.3; (c) 8.8. Experimental conditions as in Fig. 1.

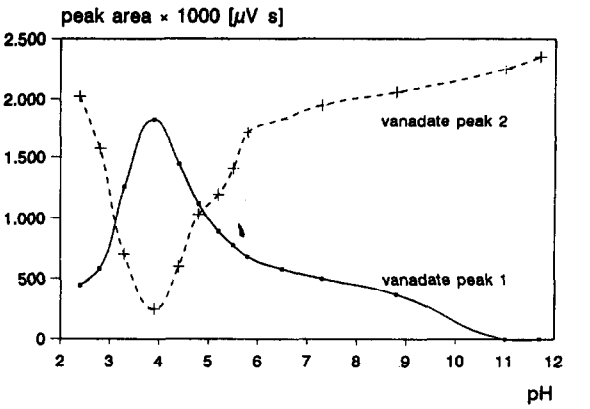

Fig. 3. Dependence of the areas of the two vanadate peaks on the pH of the analyte solution. Experimental conditions as in Fig. 1.

concentration of 0.4 m*M* showed no significant changes in the pH dependence of the two vanadate peaks, probably caused by the concentration excess of Tiron in all instances. For example, Fig. 5 shows the areas of the vanadate peaks as a function of the concentration of Tiron. The pH of the vandate solution was 7.0. Only the separation efficiency will be influenced by the concentration of Tiron. The best separation results were obtained with a Tiron concentration of about 5 m*M*.

Different vanadate concentrations (0.2–0.8 m*M*) with a fixed Tiron concentration (5 m*M*, pH 5.3) were also investigated. Fig. 6 shows the ratio of the areas of the two vanadate peaks

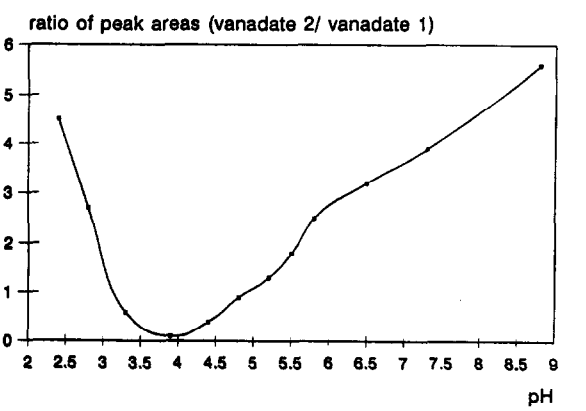

Fig. 4. Dependence of the ratio of the peak areas (vanadate 2/vanadate 1) on the pH of the analyte solution. Experimental conditions as in Fig. 1.

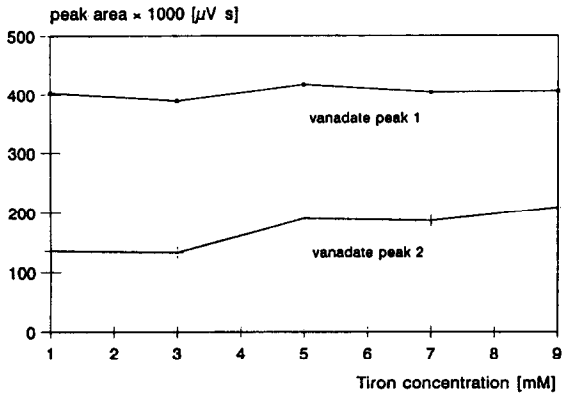

Fig. 5. Dependence of the areas of the two vanadate peaks on the concentration of Tiron. Vanadate concentration, 0.4 m*M* (pH 7.0); electrolyte, 1–9 m*M* 1,2-dihydroxy-3,5-disulphonic acid disodium salt (pH 5.3); other conditions as in Fig. 1.

using 0.2 and 0.8 m*M* vanadate at different pH values. The curve for 0.8 m*M* vanadate is comparable to that in Fig. 4, whereas the curve for 0.2 m*M* vanadate differs and shows great variations in the pH range 3–8. Therefore, this concentration is not useful for pH determination. Better results are obtained at higher vanadate concentrations (*e.g.*, 0.4 m*M*), but limited by problems of separating vanadate from the other anions of interest if the vanadate concentration is too high.

Investigations with different pH values of the electrolyte were also carried out. Using 5 m*M* Tiron, the pH range was varied from 3 to 7. The analyte solution was 0.4 m*M* vanadate at different pH values. The experiments showed that with electrolyte pH values below 5 the pH dependence of vanadate, as shown in Fig. 4, cannot be observed. Fig. 7 shows the ratio of the vanadate peak areas as a function of the pH of the vanadate solutions. The electrolyte pH value was 3.0.

In the pH range 5.0–7.0 the pH dependence for vanadate is comparable to that in Fig. 4. Above pH 7.0 the velocity of the electroosmotic flow is higher than the velocity of the vanadate ions in the opposite direction so that no measurements can be carried out.

The experiments were carried out with hydrostatic injection. With this injection mode, the ionic strength of the analyte solution shows no influence on the vanadate peaks. Electrokinetic injection with different vanadate concentrations and different pH values was also used.

Fig. 8 shows the ratio of the areas of the vanadate peaks as a function of pH using three vanadate concentrations and electrokinetic injection. The plots shows a maximum between pH 6 and 7. Hydrostatic injection is to be preferred because of discrimination effects when using the elektrokinetic injection mode. The amount of sample injected varies with the total ionic concentration of the sample [4]. Therefore, the use of internal standards is necessary [15].

Identification of the structures of the species

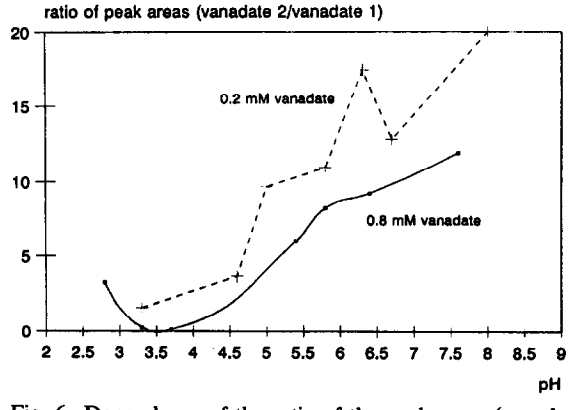

Fig. 6. Dependence of the ratio of the peak areas (vanadate 2/vanadate 1) on the pH of the analyte solution. Vanadate concentration 0.2 and 0.8 m*M*; other conditions as in Fig. 1.

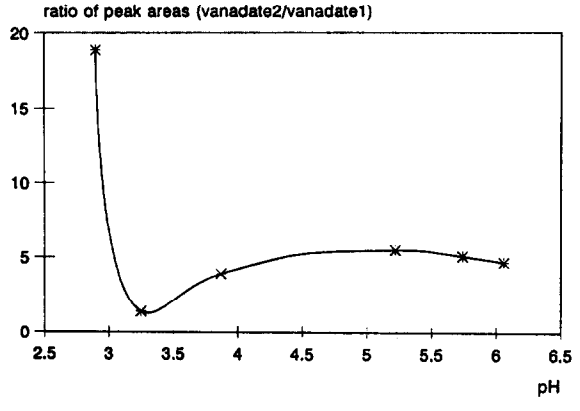

Fig. 7. Dependence of the ratio of the peak areas (vanadate 2/vanadate 1) on the pH of the analyte solution. Vanadate concentration, 0.4 m*M*; electrolyte, 5 m*M* 1,2-dihydroxy-3,5-disulphonic acid disodium salt (pH 3.0); other conditions as Fig. 1.

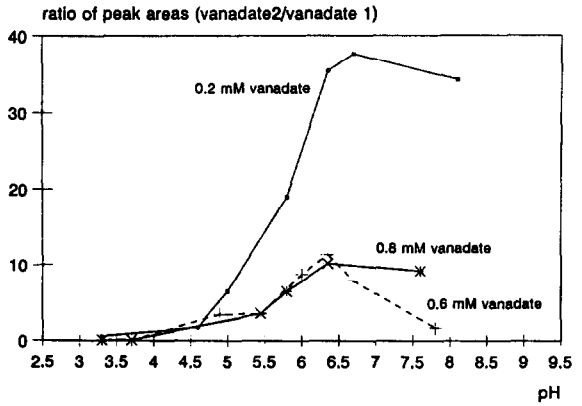

Fig. 8. Dependence of the ratio of the peak areas (vanadate 2/vanadate 1) on the pH of the analyte solution. Vanadate concentration, 0.2, 0.6 and 0.8 m*M*; injection, electrokinetic (10 s, 3 kV); other conditions as in Fig. 1.

giving the two vanadate peaks is difficult. The nature of the vanadate species existing in the acidic pH range is controversial and has not been clearly resolved [16]. The complex reactions (polymerization, condensation and protonation) of vanadates in aqueous solutions depend on pH and the vanadate concentration [12,17]. Further, the vanadate anions can be influenced by the electrolyte because two positive vanadate peaks were obtained. At 293 nm Tiron has a molar absorptivity of 5000 l mol^{-1} cm^{-1}. At the same wavelength the molar absorptivity of vanadate is only 1200 l mol^{-1} cm^{-1}. These results indicate that the vanadate anions interact with the aromatic electrolyte so that Tiron–vanadate species are formed with a higher UV absorbance than Tiron. This conclusion is supported by spectroscopic investigations because the absorbance of Tiron at 293 nm increases and the absorbance maximum will be shifted to longer wavelengths (from 293 to 299 nm) by adding vanadate to the aromatic compound.

Other electrolytes without the two hydroxy groups in the *ortho* position were also investigated. The experiments were carried out with 1,3-benzenedisulphonic acid disodium salt and 2,5-dihydroxy-1,4-benzenedisulphonic acid dipotassium salt. The results showed that only one vanadate peak will be observed. This peak shows a pH dependence similar to that of the first vanadate peak with the Tiron system. The second vanadate peak appears only with Tiron as electrolyte.

The application of vanadate for the pH determination of real samples and further spectroscopic investigations will be carried out.

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