Review

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Capillary electrophoresis for short-chain organic acids and inorganic anions in different samples

This review article is a comprehensive survey of capillary electrophoresis methods developed for the measurement of short-chain organic acids and inorganic anions in a wide variety of matrices, such as food and beverages, environmental, industry, and other applications, as well as clinical applications in body fluids such as urine, plasma or cerebrospinal fluid. Details of sample pretreatment and of electrophoretic conditions have been collected in tables, arranged by the type of matrix. Strategies employed for method development for the analysis of these compounds by capillary electrophoresis in real samples are discussed.

Keywords: Beverages / Body fluid / Carboxylic acids / Environmental / Food / Industry / Review
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Abbreviations: **BTA**, 1,2,4-benzenetricarboxylic acid; **CSF**, cerebrospinal fluid; **CTAH**, cetyltrimethylammonium hydroxide; **HDB**, hexadimethrine bromide; **HIA**, 5-hydroxyindolacetic acid; **HVA**, homovanillic acid; **IC**, ion chromatography; **LMW**, low molecular weight; **MTAB**, myristyltrimethylammonium bromide; **OFM**, OFM Anion-BT (Waters); **PAA**, polyacrylamide; ***p*-AB**, 4-aminobenzoic acid; **PDC**, 2,6-pyridinedicarboxylic acid; **PMA**, 1,2,4,5-benzene-tetracarboxylic acid (pyromellitic acid); **TMA**, trimellitic acid; **TTAB**, tetradecyltrimethylammonium bromide; **VMA**, vanillylmandelic acid

1 Introduction

Low-molecular-weight (LMW) organic acids are intermediates or final metabolites of many biochemical pathways in living organisms such as citric acid cycle, malolactic and carbohydrate fermentation, ethanol oxidation, as well as the product of certain industrial practices and, therefore, their measurement can serve as an indicator of the extent of several processes and for quality control. On the other hand, short-chain organic acids are intermediates or ultimate products in the degradative metabolic pathway of amino acids, fats and carbohydrates [1]. Several human diseases, in particular metabolic disorders, often lead to the accumulation of characteristic metabolites in body fluids including steroids, carbohydrates, aminoacids, purines and pyrimidines, and organic acids [2]. In metabolic disorders, the diagnostic metabolites accumulate as a result of genetic effects causing decreased enzyme activity. Combined with clinical information, the accurate identification of these metabolites can aid in the diagnosis of the disease. Organic acids have been determined in urine and serum in order to diagnose numerous inborn errors of metabolism known as organic acidurias [3]. Central nervous system diseases, neuroblastoma, nephrolithiasis, and other pathologies are also related to organic acids increase in body fluids.

The usual methods for LMW organic acid analysis include capillary gas chromatography (GC) with or without mass spectrometry (MS) after solvent extraction and derivatiza-

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tion. GC-MS has been used routinely as a screening method for the analysis of patient urine for the diagnosis of metabolic disorders [4]. In spite of its unquestionable sensitivity, selectivity and identification ability, two significant drawbacks of the GC-MS technique are the long time of sample preparation and analysis and the need of trained personal. That has hindered its use with general screening purposes. On the other hand, its use can be fully unnecessary for the monitoring of diagnosed diseases.

Another routine method for analyzing LMW organic acids is high-performance liquid chromatography (HPLC). Organic acids have been analyzed using normal-phase silica separation, but more frequently they have been separated underivatized or as their phenacyl derivatives in reversed-phase HPLC. Anion-exchange chromatography with suppressed conductivity detection is a well-established technique for the simultaneous determination of many inorganic and organic acids in various matrices. However, in many cases this technique also needs sample preparation and certain carboxylic acids may be coeluted. UV detection at 210 nm in line with the conductivity detector has also been applied for complementary information. Other methods of organic acid analysis include plasmaspray liquid chromatography and ion-exclusion chromatography. Related to clinical laboratory, the more common tool for specific assays are enzymatic methods. But enzymatic kits are expensive and that is more significant if a big number of samples are daily measured as can be the case with oxalate and citrate in kidney stone formers.

Capillary electrophoresis (CE) has been proved to be an extraordinary tool for the measurement of LMW organic acids. The main features of the technique related to the problem are: (i) the ability to separate small molecules from complex matrices without sample pretreatment. This is because these molecules run faster and then the capillary is completely emptied and washed after each run. (ii) The possibility of measuring the absorbance at 200 nm or below, where the carboxylic group absorbs, because it works in aqueous media. (iii) The low consumption of reactives: a few milliliters of an aqueous buffer are enough for one day.

The main drawbacks are related with the detection systems. Since the sample volume employed is very small (nanoliters), the limits of detection (LODs) in UV are not the best quality of the technique. It can be improved around 10^3 times with laser-induced fluorescence detectors, but then derivatization is needed, with all the problems associated. Finally, MS detector could give similar identification capability to GC-MS, but the coupling is still at the beginning. Chemically short-chain organic acids are small water-soluble molecules that get negatively ionized at pH values around 3–6 and do not present other chromophore more than the carboxylic group that absorbs weakly and presents its maximum absorbance around 200 nm. That wavelength only can be employed in aqueous media. Coelectroosmotic conditions are usually employed in the analysis of LMW carboxylic acids, which are accomplished by locating the anode at the capillary outlet and with the addition to the background electrolyte (BGE) of an electroosmotic flow (EOF) modifier that suppresses or reverses the EOF.

The objective of the present paper is to summarize and discuss the methods employed for LMW organic acid analysis by CE in food and beverages, environmental and industrial samples, and body fluids. All these methods are basically very similar and will be discussed altogether, but the review of the articles has been organized attending to the nature of the matrix because in this way it is easier to find the approach more similar to one concrete problem. The heterogeneous group of compounds related with body fluids presents similar CE behavior independently of the disease they are related to. That is the cause why in the present paper very different pathologies are included and many times physiological compounds not associated with a pathological increase, but appearing in the electrophoretic profile in the analytical conditions.

Table 1 summarizes the works published on food and beverages [5–37], Table 2 on environmental samples [38–63], Table 3 on industrial processes [64–82], Table 4 on miscellaneous samples of diverse origin [83–90], and Table 5 on body fluids [2, 15, 43, 91–123] (see Addendum pp. 1964–1981). Comprehensive surveys related to the application of CE to the analysis of carboxylic acids and related acids in one specific matrix have been published by Klampfl *et al.* [124] and Lindeberg [125] in food and beverages and by Craston and Saeed [126] in environment. A previous text of clinical applications of CE for short-chain organic acids analysis was written by Nuttall and Guzman [127].

2 Sample pretreatment

It is important to highlight that simplicity in this step is one of the main contributions of the technique and therefore most of the methods developed do not employ other sample pretreatment more than dilution and filtration or centrifugation [5–22, 25, 28, 31–37, 40–43, 48, 50, 53–56, 59–63, 66, 67, 72, 73, 75, 77–81, 83, 87]. Karlsson *et al.* [46] studied the influence of filtration, preservation and storing on the analysis of LMW organic acids in natural waters. Some authors added EDTA to liberate the

organic acids from its possible complexes with metals [24, 57, 58]. In dairy products, acidification with sulfuric acid has been employed to facilitate coagulation [10, 11]. In latex, two modes of coagulation have been employed: acidic media and freezing [78]. Gaseous samples such as some beers, soft drinks and wines have to be degassed, generally it is done by sonication [5–8, 12]. Special treatment deserved ascorbic acid analysis in vegetables, which employed 2% thiourea in 10 mM HCl acid [27]. In some cases, solid-phase extraction (SPE) has been employed on C_{18} [49] or cation exchangers [74] and even on-line dialysis in a flow injection analysis (FIA) arrangement [88] has been coupled to the CE equipment. Recently, solid-phase microextraction with polyacrylate-coated fiber has been employed but only for aromatic acids in soils [128].

In the case of body fluids, the best fluid to analyze for organic acids is urine because: (i) organic acids are concentrated in the urine so that most of them are present in the urine in much higher concentration than in the blood; (ii) the virtual lack of protein facilitates the analysis of the sample, (iii) a specimen consisting of a simple voiding is easy to obtain and is adequate for analysis. Therefore, urine is the body fluid more frequently analyzed for short chain organic acids [2, 15, 94, 98, 100, 102–106, 108–110, 112–115, 117–123, 129–131]. Other fluids also analyzed for organic acids by CE are saliva [91], cerebrospinal fluid (CSF) [92,113], serum [94–96, 99, 100, 107], and amniotic fluid [113, 116].

One of the advantages of CE is the ability to separate small molecules in complexes matrices without sample pretreatment. That is so, because once the analytes pass through the detector the capillary can be emptied and washed and it is ready for a new analysis. That is why many authors measure organic acids in urine or saliva without any other sample treatment more than dilution and filtration or centrifugation to eliminate solid matter [2, 91, 100, 102, 106, 108, 109, 115, 119, 120, 122, 129]. Nevertheless, although protein content in urine is low when uncoated capillaries are employed, proteins can interfere because they get adsorbed on the capillary wall and they must be eliminated prior to the analysis. It can be done with SPE on C_{18} cartridges [93, 104] although Willetts *et al.* [103] detected selective retention of certain organic acids such as lactate, or with cationic exchange resins for orotic acid in urine [105]; by liquid-liquid extraction with ethyl acetate [98]; or by acidification alone [117] or followed by thermic treatment [114]. Deproteinization with acetonitrile (two volumes of acetonitrile to one volume of sample) seems to offer a simultaneous method of stacking for small molecules, which is a mechanism of analyte concentration on the capillary itself [132].

Purification has also been developed in-line by isotachophoresis [110] prior to CZE. On conditions that a suitable electrolyte system is selected for ITP step, performed in the first preseparation capillary of a higher internal diameter, sample components create correct and stable isotachophoretic zones with sharp boundaries. Only a well-defined fraction of the sample containing the stacked analyte is transferred into the second analytical capillary and analyzed by CE.

Serum with higher protein content than urine is usually deproteinized because proteins can precipitate in the capillary or get adsorbed to the wall in uncoated capillaries and migration times, in that cases, vary broadly. It has been done by ultrafiltration [95] not only in serum, but also in cerebrospinal liquid [92] or by precipitation with cold methanol [100]. However, some authors have succeeded to measure directly in serum with polyacrylamide(PAA)-coated capillaries and a careful selection of the BGE components [96]. Derivatization for including a group that facilitates detection is another way of sample pretreatment used by some authors, but it will be described in more detail in the Section 3.5.

3 Method development parameters

Parameters to be optimized during method development include separation mode, type of capillary, BGE, injection mode, and detection. They are discussed briefly below.

3.1 Separation mode

Separation mode is the first election to develop an analytical method for short-chain organic acids. Since their electrophoretic mobility towards the anode is usually higher than the EOF towards the cathode, the most common mode of analysis is the mode called reversed polarity, which means that the injection is performed at the negative end (cathode) while the detector is placed in the positive end (anode).

The analysis of LMW organic acids with the opposite polarity is not frequent, because these compounds present high electrophoretic mobility towards the anode and could exit by the injection end without passing through the detector. Nevertheless, benzoic acid [12], ascorbic acid [17] alone or with isoascorbic acid [49], and a group of compounds produced as effluents in a distillery [69] or in the production of sugar [71] have been measured with normal polarity. In the case of clinical analysis, vanillylmandelic acid (VMA) and homovanillic acid (HVA), which contain an aromatic ring and therefore bigger size, normal

polarity is the most common mode, but reversed polarity has also been employed [122]. It must be pointed that in some papers it is not possible to work out that reversed polarity was employed, but the migration orders of the analytes make think in that sense [33, 74, 88].

3.2 Type of capillary

Reversal of the polarity needs the EOF (towards the cathode in the standard configuration) to be suppressed or even reverted. Flow reversal is achieved by two basic methods, use of coated capillaries or uncoated capillaries with a surfactant added to the BGE. Most of the methods developed for LMW organic acids employed uncoated silica capillaries with a surfactant added, but the adsorption of compounds to the wall make the reproducibility not always as good as it would be desiderable. Some authors employed different capillary types such as eCAP [9], fluorinated ethylene propylene (FEP) capillary [16], and PAA-coated capillary [19].

Fourteen short-chain organic acids were studied by CE with indirect UV detection in three different capillary conditions: polyacrylamide-coated, myristyltrimethylammonium bromide dynamically coated and uncoated. Actually, dynamical coating consists of a surfactant added to the background buffer and it cannot be considered a type of capillary. The best performance regarding precision in migration time, highest column efficiency, and better LODs was obtained by using the PAA-coated capillary. Nevertheless, when the method was applied to clinical urine samples, several interferences appeared and the authors recognize that method needed further study for real samples [100]. Our experience, as much with standards as with many different biological samples [108, 109, 113, 117, 122], is also that PAA-coated capillaries performances related to reproducibility are the best. Moreover, if capillaries are adequately treated, their usable period can be very long (to our experience even more than two years working daily) and that compensates the initial higher costs.

Orotic acid was analyzed in capillaries coated with polyvinyl alcohol [104]. Following the authors, these capillaries performed well, were stable and required little conditioning to give reproducible migration times. However, it was necessary to employ relative complex specimen preparation steps to achieve good assay precision, which eliminates the major advantage of the technique. FEP and fused silica was employed for the ITP-CZE measurement of orotic acid in children urine [110]. On the other hand, Nutku and Erim [35] employed a polyethyleneimine-coated capillary that generated an anodic EOF and thus favored the separation speed of organic acids.

3.3 Background electrolyte

The nature, concentration and pH of the BGE in CE are the most important parameters for resolution and detection. Theoretically, once the organic acids are fully ionized, which happens at pH values two units over their pK_a, the pH value is not very important and it ought to be adjusted considering the maximum buffer capacity of the electrolytes, but under this limit, small variations even in the second decimal figure significantly affect the separation. Buffering electrolytes of sufficient capacity are needed to control buffer-ion depletion caused by electrolysis.

Harrold *et al.* [97] demonstrated the ability to modify electrophoretic mobility and selectivity as a function of temperature and electrolyte ionic strength for inorganic and short chain organic acids. Although it is a work developed only for standards and focussed in the application of a particular mode of detection, it provides with strategies to be applied for the separation of these compounds. It is well known that as electrolyte ionic strength decreases, the inherent electrophoretic mobility of the anions toward the anode decreases while the EOF toward the cathode increases, resulting in an overall run time decrease. The important observation is that the electrophoretic mobilities of the anions change at different rates and that permits to manipulate the selectivity. Nonabsorbing electrolytes have been employed for direct detection such as tetraborate at pH 9.3 [21, 28], phosphate at pH 10.2 [69], pH 7.5 [19], pH 6.5 [33], 6.25 [78], and pH 6 [59]. Regarding concentration, it affects EOF, electrodispersion of the analyte bands, and current generated at a given potential. An equilibrium ought to be found between the stacking effect and current generated, nevertheless, concentrations in the BGE as high as 500 mM have been employed without any problem, with equipment that refrigerate the capillary [78].

For indirect detection, the BGE is even more critical not only the pH and wavelength of the chromophore, but also its mobility related to these of the analytes and the concentration to give a maximum range for measuring. Wu *et al.* [22] investigated and discussed the suitability of several absorbance providers, additives and pH, affecting the selectivity and resolution of CE for mono-, di- and tricarboxylic acids, as much as, hydroxy acids. Although their work was only applied to standards, the information can be very useful to work with indirect detection of short-chain organic acids. Moreover, these factors have been reviewed for carboxylic acids by Doble and Haddad [133].

As chromophore and buffer have been employed phtalate [11, 15, 18, 62, 67, 72, 73, 91, 98, 100], benzoate [9, 36, 81, 82, 85, 86], PDC [5, 6, 10, 14, 20, 25, 38, 61, 75],

TMA [22, 45, 54, 57, 58, 74], MES [16, 31], PMA [36, 37, 41, 82], 2,4-dihydroxybenzoic acid with ε-aminocaproic acid [35, 96], 1,2-dimethylimidazole and trimellitic acid [13], *p*-hydroxybenzoate [40, 43, 47, 63], *p*-anisate [84], 3,5-dinitrobenzoic acid [42], salicylic acid with Tris [48, 52], benzoic acid and Tris [107], NDC [39], *p*-AB [32, 38, 51], BTA [46], phenylhydroxyacetic or mandelic acids [96], glutamic acid plus spermine [110], and 5-sulfosalicylic acid [44]. The sensitivity obtained with 2,6-naphthalenedicarboxylic acid was reported as five times higher than with phthalate, which is commonly used. On the other hand, inorganic chromophores have also been employed such as chromate [23, 24, 26, 65, 66, 68, 70, 76, 77, 80, 88, 89, 114] and molybdate [53].

With uncoated capillaries and revesed polarity a surfactant must be added to decrease, suppress or even reverse the EOF. Cationic surfactants such as CTAB [5, 6, 10, 11, 18, 23, 26, 38, 40, 42, 55, 72, 73, 76, 78, 79, 88, 134], CTAH [14, 20, 53, 75], TTAB [7, 8, 22, 24, 31, 32, 39, 45, 51, 54, 57–61, 66, 74, 81, 82, 85–87,89], TTAOH [21, 28], and MTAB [33, 34, 62] have traditionally been employed. Surfactants with different nature have also been employed such as HDM [25, 50, 80, 82]. The more hydrophobic the surfactant, the more effective is the reversal of EOF and the faster the migration time of the acids [39, 135]. Volgger *et al.* [90] compared the effect of CTAB, TTAB and HDB. For these authors HDB rendered the best overall results in terms of separation speed and resolution of relevant acids for their problem. Various alkylamines have been more recently investigated as EOF suppressors and tetraethylenepentamine was selected by Fung and Tung [83] to obtain a nice separation of 25 organic and inorganic anions. In a previous work, Arellano *et al.* [36] employed EDTA for the separation of seven organic acids and four inorganic anions in wine and fruit juices. Generally, a single-surfactant species has been used, to reverse the EOF, but Haddad *et al.* [66] noted certain selectivity effects in the separation of inorganic and organic anions when a binary mixture of surfactants is used.

Another method for manipulating selectivity is the addition of alkali-earth metals, mainly Ca^{2+}, to the BGE [21, 28, 43, 45, 48, 51, 54, 63, 67, 103]. In the capillary these ions interact with the organic analytes through the formation of complexes with different stabilities, which affects the electrophoretic mobility of the ligands. Small amounts of organic solvents can also be added to the BGE for improving resolution. Methanol has been added at 5 [33, 34, 62, 122], 10 [108, 109, 117], 20 [18, 83], 30 [31], and even 50% [84]. Acetonitrile has been employed at 5 [88], 15 [59], 20 [60], and 30% [98]. Even mixtures of both [94] have been employed as organic modifiers to improve resolution in some cases. Some authors employed a commercial BGE of undescribed composition [102].

Special mention must be made of CE-MS buffers, that should contain mainly volatile compounds to work at an optimum level. Thus, ammonium bicarbonate [112], ammonium acetate [2] or an aqueous solution of naphthalene disulfonate, PMA, and methanol with diethylenetriamine as EOF modifier have been employed [111]. Compounds such as orotic acid with a protonable nitrogen can also be measured at low pH, positively charged [104].

3.4 Injection mode

In hydrodinamic injection, the loaded sample volume is nearly independent of the sample matrix, although it depends on the viscosity. However, in electrokinetic injection, the amount loaded is dependent on the EOF, conductivity and viscosity of BGE, and sample and electrophoretic mobilities of the analytes. Thus, injection bias exists with the more mobile species being loaded to a greater extent. Levart *et al.* [38] found preconcentration factors ranging from 14 (chloride) to 3 (propanoate) by using electrokinetic injection. Electrokinetic sample introduction with transient isotachophoretic preconcentration was optimized for peak height, peak area, peak asymmetry, efficiency, peak resolution, and reproducibility of migration time and peak area for the ultratrace determination of anions on silicon wafer surfaces [82]. The detection limit was 10 $nmol \cdot L^{-1}$ and results agreed with those obtained by ion chromatography (IC). Nevertheless, except for samples with a constant matrix, the use of electrokinetic injection enhances CE sensitivity, but it suffers from matrix bias and poorer precision and, therefore, it is not recommended for quantification.

3.5 Detection

As previously described, organic acids can be separated in aqueous buffers and if nonabsorbing electrolytes are employed, direct measurement at 200 nm or below is a good option for the carboxylic group [92, 93, 95, 101–104, 106, 108, 109, 122, 129]. Obviously, when organic acids with a characteristic spectra are measured, different wavelengths can be employed, for example, orotic acid was analyzed at 280 nm [105]. Positive identification of these compounds can be enhanced by the use of diode-array detection and spectral matching.

As can be seen in Tables 1–5, in 79 proposals out of 124 indirect detection was employed. Indirect detection is achieved by including an absorbing ion (UV-absorbing

or fluorescence-absorbing chromophore) in the buffer, which provides a high background absorbance. Displacement of the absorbing ion by analytes within the sample produces negative absorbance peaks, which can be turned in positive by the software of the equipment. The choice of the indirect chromophore is dictated by the mobility of the ions to be analyzed, since the best resolution occurs when the mobility of the anion in the buffer is close to that of the sample ions. In some cases, a derivatizating agent, such as pirenyldiazomethane has been bonded to the molecules to increase the UV absorbance [94, 99].

Nevertheless, controversy exits about direct or indirect detection for providing better LODs. Generally, the latter is considered a mode more sensible than direct detection, and it can be so for standards, but when dirty or complex samples have to be measured, such as biological fluids, high dilution rates have to be employed to avoid very noisy baselines and overlapping peaks and, then, the result is not so good. Tables 1–4 include the LODs reported for the different methods and when two values appear they are the range (minimum and maximum LODs) for the different acids. Castiñeira *et al.* [34] concluded that the sensitivity of the analysis in wine samples carried out using direct detection was from 35–80-folds higher than for the indirect procedure. Similarly, for saturated carboxylic acids which are intermediates and reaction products in the conversion pathway of citric acid and itaconic acid in hot, LOD of 100 ppb for direct detection and 25 ppb for indirect detection have been found [90]. Probably, that is in part due to the different behavior of standards and samples. In general, indirect detection provides higher sensitivity for standards but, as every compound with lower absorption than the BGE gives a peak, samples have to be more diluted to avoid interferences than samples measured with direct detection and final LODs in real samples are similar or even lower.

Figure 1 shows an example of a CE separation with indirect UV detection for 25 standards and Fig. 2 the corresponding application to herb extracts. For comparison, Fig. 3 shows an example of a CE separation with direct UV detection for standards and a sample of natural rubber latex serum and Fig. 4, an example of a CE separation with direct UV detection for standards related to clinical samples.

Since the first commercially available CE device offering the possibility of conductivity detection, only a small number of papers dealing with the applicability of this system for the analysis of real samples has been published. LMW ionic compounds have been measured in electrodeposition coatings by CE with conductivity detection [136]. Authors found an excellent agreement with the results achieved by IC and shorter analysis time. Nonaqueous capillary electrophoresis is best suited for fluorescence detection. Merocyanine 540 has been employed as a

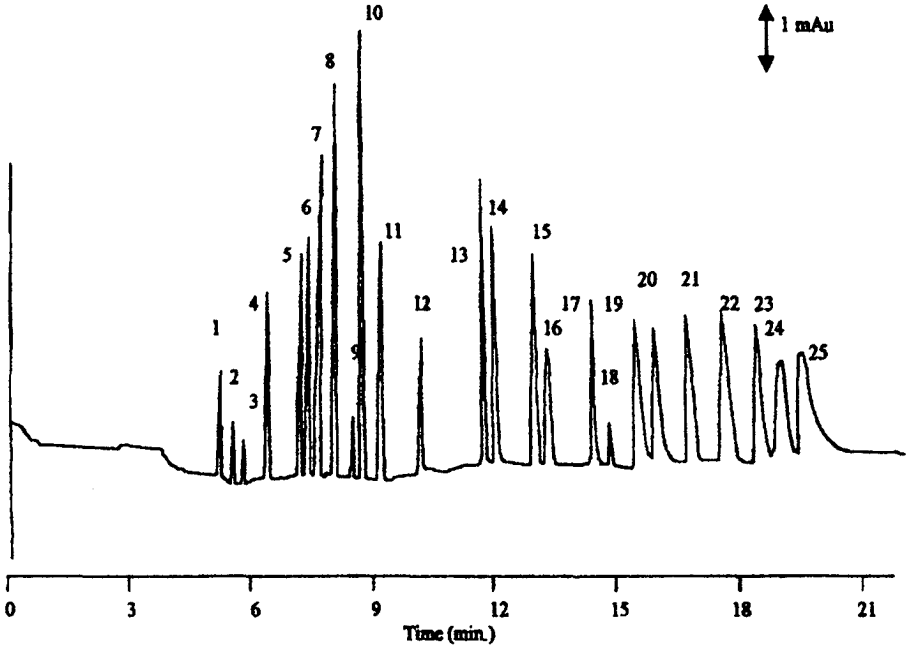

Figure 1. Electropherogram of a standard mixture of organic anions. Conditions: buffer, 15.0 mM tetraethylenepentamine (TEPA), 20.0% methanol at pH 8.4; capillary, 65 cm × 75 μm ID fused silica; voltage, −18 kV; current, 6 μA; injection, 8 cm for 25 s; detection wavelength, 254 nm; concentration, each anion 0.1 mM. Peaks: 1, chloride; 2, nitrate; 3, sulfate; 4, oxalate; 5, malonate; 6, formate; 7, fumarate; 8, tartrate; 9, malate; 10, succinate; 11, glutarate; 12, adipate; 13, citrate; 14, acetate; 15, propionate; 16, lactate; 17, *n*-butyrate; 18, 2-hydroxyisobutyrate; 19, valerate; 20, chlorovalerate; 21, capronate; 22, glutamate; 23, octanoate; 24, quinate; 25, glucuronate. Reprinted from [83], with permission.

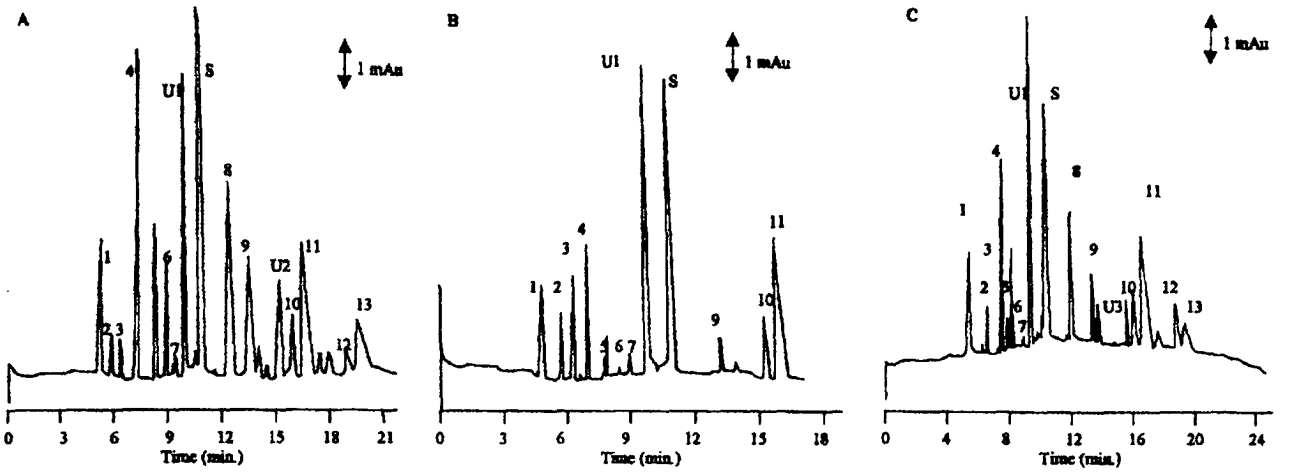

Figure 2. Electropherograms of typical herb samples colleted from a local pharmaceutical shop. (A) *Flos chrysthemi*, (B) *Spica prunellae*, (C) *Folium mori*. Conditions as in Fig. 1, except injection, 8 cm for 30 s. Peaks: 1, chloride; 2, sulfate; 3, oxalate; 4, malonate; 5, tartrate; 6, succinate; 7, glutarate; 8, citrate; 9, lactate; 10, valerate; 11, chlorovalerate; 12, quinate; 13, glucoronate; S, system peak; U1–U3, unidentifiend peaks. Reprinted from [83], with permission.

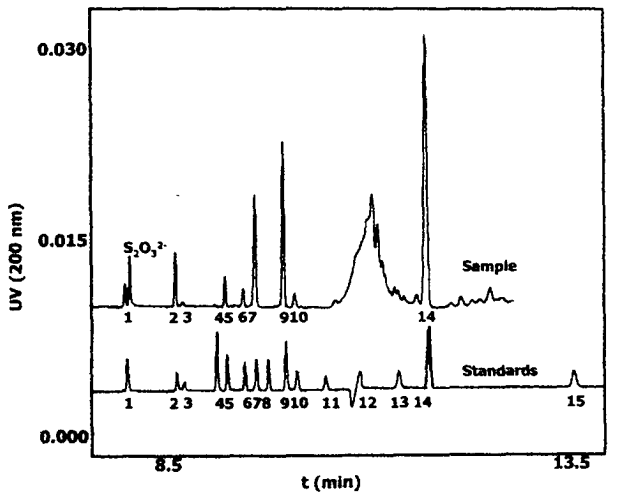

Figure 3. Electropherogram of a standard mixture and sample of natural rubber latex. Conditions: buffer, 500 mM H_3PO_4, 0.5 mM CTAB, pH 6.25; capillary, 57 cm × 50 μm ID uncoated fused silica; voltage, −10 kV; current, 118 μA; injection for 5 s; detection wavelength, 200 nm; Peaks: 1, nitrate (0.25 mM); 2, oxalate (0.5 mM); 3, formate (4 mM); 4, fumarate (0.25 mM); 5, aconitate (0.25 mM); 6, succinate (2.0 mM); 7, malate (2.0 mM); 8, glutarate (2.0 mM); 9, citrate (2.0 mM); 10, acetate (3.0 mM); 11, glycolate (3.0 mM); 12, propionate (2.0 mM); 13, furanoic (0.5 mM); 14, pyroglutamate (0.5 mM); 15, quinate (2.0 mM).

fluorophore for indirect laser-induced fluorescence detection of ascorbic acid and its stereoisomer isoascorbic acid, but LODs were not better that 0.30 and 0.17 μM, respectively [137]. Fluorescence has also been employed after derivatizating the carboxylic acids, but derivatization of short-chain organic acids in aqueous solution is the most challenging because of the low reactivity of the carboxylic group in water. Usually, it requires several reaction steps. Nevertheless, some derivatizating agents have been employed in biological samples: 5-bromofluorescein for C_8–C_{11} carboxylic acids to be detected with the argon laser at 488 nm [107]; 1-pirenyldiazomethane for dicarboxylic acids to be detected with He-Cd laser [94, 99].

Methods of describing LODs are usually very confusing because many authors report absolute masses in the capillary and it must be borne in mind that there are only a few nanoliters of sample into the capillary; other authors give concentration in the vial, but samples have to be diluted during the treatment; Schneede *et al.* [94] clearly describe 40 nM for the methylmalonic-pirenyldiazomethane derivative and under 1 μM of methylmalonic in human serum. An exhaustive study of parameters that influence on separation was developed by these authors, but there are no data about the quantitativity or reproducibility of the derivatization reaction.

Electrochemical detection has also been employed in some cases, but the problem of measuring very small currents at the end of a capillary with high voltage is well known, as are the problems related to the extreme potentials needed for obtaining a redox response from carboxylic acids. Fu *et al.* [115] employed a graphite-paste electrode modified with cobalt phthalocianine, but they only measured oxalate, ascorbate and uric acids, those with known electrochemical properties, and recently Li *et al.* [123] employed a carbon-fiber microdisk for measuring VMA and HVA.

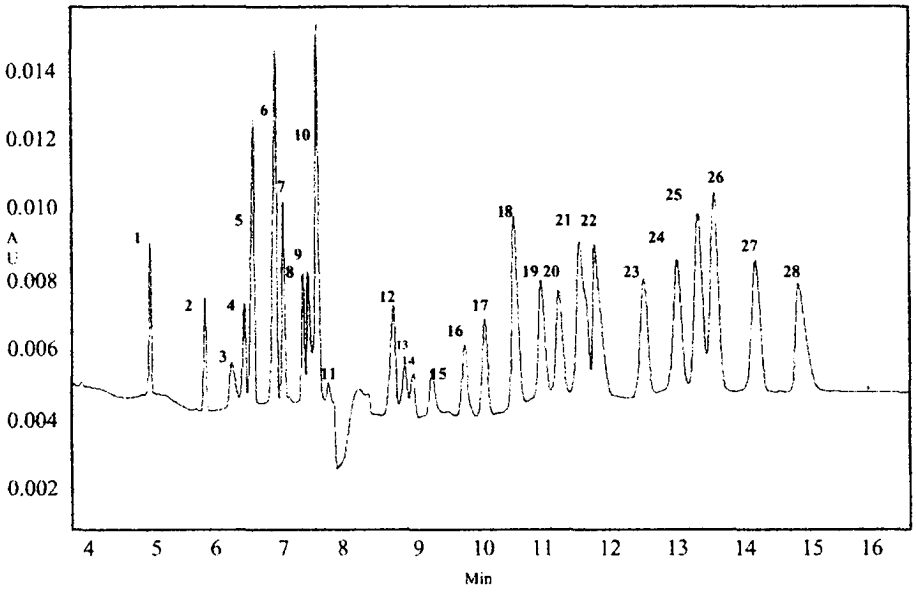

Figure 4. Electropherogram of a standard mixture of short-chain organic acids. Conditions: buffer, 200 mM phosphate, pH 6.0, with 10% methanol added, neutral coated capillary, 37 cm length, injection by pressure of 0.5 psi for 5 s, voltage, −10 kV; detection at 200 nm. Peaks: 1, oxalic; 2, fumaric; 3, ketoglutaric; 4, malic; 5, methylmalonic; 6, glutaric; 7, citric; 8, adipic; 9, methylcitric; 10, *N*-acetylaspartic; 11, glycolic; 12, acetoacetic; 13, propionic; 14, lactic; 15, ketoisovaleric; 16, glyceric; 17, 2-hydroxybutiric; 18, 3-hydroxybutiric; 19, 2-hydroxyisovaleric; 20, 3-hydroxyisovaleric; 21, propionylglycine; 22, 4-methylvaleric; 23, phenyllactic; 24, homogentisic; 25, hypuric; 26, uric; 27, 4-hydroxyphenyllactic; 28, aminoadipic acids.

Related to MS detection, to date only two works have been published dealing with the analysis of small carboxylic acids [138, 139]. In the first study, succinic, maleic, malonic, and glutaric acids were separated, meanwhile in the second one, 11 LMW organic acids were determined and parameters optimized to achieve the highest sensitivity. Applicability to an ale sample was included. Dealing with the analysis of diagnostic metabolites by CE-MS two works have also been published. The first one by He *et al.* [112] includes glutathione, pyroglutamate, adenylosuccinate, ornithine, histidine, and homogentisic acid measurement in normal and spiked urine samples. The second one was developed by Jellum *et al.* [130] and is devoted to the analysis of urine and blood samples from patients with known metabolic disorders (galactosemia, neuroblastoma, Zellweger syndrome, propionic academia, and alcaptonuria) by CE-tandem MS. The authors say that although the results are promising, there is still a long way to go. Libraries of urinary metabolites must be created for automated identification and the potential of these techniques must be further evaluated by running patient samples routinely.

4 Chiral analysis

Chiral analysis of short-chain organic acids is complicated because their short chains make difficult the three-point interaction generally accepted as necessary for chiral recognition. Moreover, they lack a powerful UV-absorbing chromophore. That is why many methods for chiral short-chain organic acids analysis have been developed with derivatization to diastereomers. Relatively recent works have shown the possibility of their direct chiral separation in CE by different mechanism: Ligand-exchange CE, macrocyclic antibiotics and cyclodextrins.

The presence of organic acid racemates in food products can indicate their use as additives, which are not always permitted and needs to be controlled. On the other hand, different isomers of the same acid can present different flavor or taste and their analysis can be of interest for quality control. Lactic acid in sake, as well as in wine, is a major organic acid and it is thought to have a great influence on the taste. Whereas naturally occurring lactic acid bacteria are used in the traditional brewing of sake, the use of lactic acid addition has recently been predominant in order to simplify sake brewing [140]. Authors say that although sensory studies are needed, D-lactic acid in water has a different sour taste from L-lactic.

On the other hand, most biochemical reactions have enantiomeric selectivity. Different enantiomers of the same compound can activate different metabolic pathways [141]. Enantiomeric ratio of chiral metabolites is an important parameter for the understanding of metabolic processes and in many cases it can have diagnostic purposes. From this point of view it is possible to determinate the origin of several pathologies by an enantiomeric

analysis of selected metabolites. D-Enantiomers usually have a bacteriological origin while L-enantiomers have predominantly an endogenous one. CE has demonstrated to be a good choice for enantiomeric resolutions using chiral selectors in the separation buffer which can provide very simple and automated method development.

The optimization of the separation conditions in CE of the two optical isomers of lactic acid by a factorial design has been reported. The method, which does not require any other sample pretreatment more than dilution and filtration, was applied to the identification of both isomers in body fluids as plasma, urine, amniotic fluid, and CSF [113]. This very complex area exceeds the intention of the present paper, but to mention that a comprehensive review on the subject has been recently published [142].

5 Comparison of CE with other separation techniques

Many of the authors have compared CE analysis with IC, being the general conclusion that CE offers several advantages over IC: enhanced separation efficiency, tolerance of complex matrices without laborious sample pretreatment, shorter analysis time, and lower cost [7, 73, 82]. Results obtained by CE during the determination of LMW ionic compounds in electrodeposition coatings [136], a wide variety of samples, ranging from simple aqueous solutions to complex plant organic streams [73], silicon wafer surfaces [82] showed excellent agreement with those achieved by IC; in a systematic approach to the separation of mono- and hydroxycarboxylic acids in environmental samples by IC and CE. Souza and coworkers [42] concluded that a complete characterization of all analytes could not be achieved by IC due to coelution of certain analytes. On the other hand, in CE it was possible to discriminate all analytes, but the method lacked concentration sensitivity. Similar conclusions were obtained by Roselló *et al.* [25] when comparing a CE method for organic acids involved in tomato flavor analysis with routine HPLC methods.

6 Applications

The applications related to food, beverages, environment, and industry are clearly summarized in Tables 1–4 and they do not deserve further comments. Those applications related to body fluids will be briefly discussed to establish their clinical relevance.

6.1 Short-chain organic acids profiling

Individually, many disorders of organic acid, fatty acid metabolism and other aminoacidopathies excluding phenylketonuria (PKU) are rare, but collectively they are probably of an equal incidence to PKU at about 1:5000–1:10 000 live births [143]. The analysis of short-chain organic acids in urine is a well-established procedure for the diagnosis of inherited errors of metabolism [144, 145]. Currently, GC-MS is the most reliable technique for this purpose, nevertheless, it is also expensive, laborious and limited to referral laboratories. On the other hand, CE can provide a simple and rapid alternative [127]. CE is limited at the present time to the analysis of a relatively short number of acids and identification is performed by migration time as compared with standards and by spiking and therefore it has not the structural elucidation ability of GC-MS, but the benefits of a method such as CE that provides rapid analysis is apparent in such situations as the critically ill newborn presenting coma and metabolic acidosis. In such cases, rapid diagnosis facilitates appropriate treatment.

Nowadays, when done, screening of inborn errors of metabolism including phenylketonuria is developed by MS/MS in blood samples [146]. This is a very expensive technique and it measures carnitines and glycines more than organic acids. Thus, it does not allow one to differentiate propionic from methylmalonic aciduria. Both derive from the metabolism of propionate, and the same conjugated compounds are increased, but methylmalonic acid is the essential marker of methylmalonic aciduria [147]. In this case, CE could be a complementary diagnostic tool. Clinical management of methylmalonic aciduria is considered to be most critical during the early years of life [148].

On the other hand, CE with a very different separation mechanism can be also a complementary analytical tool for compounds such as propionic or oxalic acids poorly detected by GC-MS, due to their low recovery after sample pretreatment, and for compounds such as pyroglutamic and pipecolic acids with the same masses and therefore interfering. Chen *et al.* [100] described an indirect detection assay for 14 short-chain organic acids in serum and urine. Wu *et al.* [22] also separated 14 short-chain organic acids with indirect UV detection, but only preliminary results with body fluids were provided. Shirao *et al.* [93] described an assay for 12 short-chain organic acids in urine and Jariego and Hernanz [102] also described an assay for 10 short-chain organic acids in urine both with direct detection at 185 nm. Hiraoka *et al.* [92] described a similar method for CSF. Petucci *et al.* [95] reported a method for screening 19 metabolites in uremic serum with direct detection and normal polarity. Barbas *et al.*

[108] developed and validated a method for measuring 10 organic acids in urine and, then, with small variations, it was applied to identify 27 organic acids [109]. Simultaneously, a second buffer at a lower pH was developed with confirmatory purposes. This is the method with the largest number of biologically relevant organic acids identified in urine by CE. Figure 4 shows the separation obtained for the standards with this method. Moreover, the urine sample collection in filter paper to facilitate the collection and sending to the clinical laboratory has been studied by these authors [131] and 20 organic acids can be detected. In opinion of Seymour *et al.* [4] general screening programs would be recommended for glutaric aciduria, because prevention is possible and it is included in the study. At least 15 acids can be measured with this method. That could facilitate massive screening programs.

6.2 Nephrolithiasis markers

The evaluation of risk factors for calculi formation is a common clinical test in developed countries. The majority of stones, 70–80%, are composed mainly of calcium oxalate crystals [149]. Thus, elevated oxalic acid excretion is a risk factor, meanwhile, elevated citric acid excretion is a protective factor that tends to prevent calcium from precipitation. A comprehensive review on the subject has been recently published by García *et al.* [150]. The classical clinical tests are enzymatic assays which measure each acid in a separate probe, these methods are expensive and need manual work. CE permits the simultaneous and automated measurement of both acids, and many times other related compounds, in a short time and without any other sample pretreatment more than dilution and filtration. Holmes [114] described a method with indirect detection that permits to detect related anions such as glycolate and urate. Samples need 100-fold dilution, mainly to reduce chloride concentration. That affects LODs, and it may be variable depending on the concentrations. García *et al.* [122] validated the method previously described for profiling short-chain organic acids for quantifying oxalate and citrate. These compounds are also present in the separations obtained by other authors previously quoted. Accuracy was established by comparing with the enzymatic assays in 29 urine samples with very good results.

6.3 Homovanillic and vanillylmandelic acid and related compounds

HVA and VMA, the major metabolites of catecholamines, are often tested in urine for neurologic diagnosis and for monitoring the response to therapy in illnesses like phaeochromocytoma and neuroblastoma [151–153]. The latter is the second most frequent disease, leukemia being the first, seen in children with malignant tumors [120]. Neuroblastoma, neuroblastic tumor, is the most frequent extracranial solid tumor in early childhood [154]. In USA, the incidence is one out of 7000 children younger than the age of five [155]. In England, the incidence of neuroblastoma in the northern region is one in 10580 live births [156]. This disease is one of the few malignant tumours that excrete unambiguous markers for diagnosis. About 95% of the patients studied were reported to excrete abnormal levels of either or both VMA and HVA in their urine [157]. If detected in the early stages, before the age of 1 year, the disease may be perfectly cured [158].

In view of this, low-cost methods are necessary to satisfy the rising demand for mass screening in childhood [159]. Moreover, a second area of clinical pathology that involves the cardiovascular system (hypertension, hypotension) is also related with these metabolites [160] and so the demand for their measurement is increasing. On the other hand, intestinal tumors, which secrete large amounts of serotonin, are often discovered by the enhanced urinary excretion of 5-hydroxyindolacetic acid (HIA) [161]. In a strict definition, these compounds are not short-chain organic acids, but they can be considered included in a wider sense of the term.

Isaaq *et al.* [118] described a CZE method for HVA and VMA measurement in infant urine samples after extraction with ethyl acetate. Since the concentration in the urine samples of healthy infants is less than the detection limit, a concentration step is also necessary with this method. Caslavska *et al.* [119] developed a method based in MEKC for urinary indole derivatives and catecholamine metabolites with fluorescence detector by direct injection of plain or diluted samples. On the other hand, Shirao *et al.* [120] recently published another micellar electrokinetic chromatographic method including VMA and HVA, but not HIA. As only spiked samples are treated the method is intended for detecting clearly pathological situations during routine mass screening of pheochromocytoma and neuroblastoma.

García *et al.* [122] developed separation conditions in CE, with a neutral-coated capillary and reversed polarity. The method was optimized to make direct measurement of VMA, HVA and HIA possible in urine samples without pre-treatment. The method developed was validated, presenting adequate parameters for linearity, accuracy and precision. Detection limits range from 0.03 to 2.5 μM. It was applied to urine samples taken from patients both adults and children in hospital. Some of them were also measured by immunoassay and with HPLC with electrochemical detection (ED) and results compared well.

7 Concluding remarks

CE is already a mature technique to be implemented in routine analysis of short-chain organic acids with various advantages, with reference to other more classical techniques. The success of the technique arises from its ability to provide simple, efficient and low-cost separations in a short time with minimum consumption of reactives. With regard to organic acidurias, this technique provides an interesting tool for screening programs in newborns. For prenatal diagnosis or when the patient is acutely ill, the procedures utilized must be capable of providing not only accurate but also rapid results.

The CE method with a higher number of organic acids separated and identified permits analysis of 27 compounds in a 15 min run [109], out of near 200 that might exist. Although this is not many, it is important to consider that included in this list are some of the more frequent pathologies, such as methylmalonic, propionic, Canavan disease, pyroglutamic aciduria, hyperoxaluria, orotic, fumaric, isovaleric, alkaptonuria, lactic aciduria, ketosis, and even more important glutaric aciduria. In the last instance, if it is detected and treated at an early stage, development can be normal.

On the other hand, CE is a technique with a mechanism of separation orthogonal to GC-MS and, therefore, it can be a complementary analytical tool. CE would be the best option for monitoring previously diagnosed diseases, when the structural identification of unknown diagnostic metabolites is not necessary. A possible role of CE in the routine system for metabolic disorders might be following the diagnosed samples and pretesting all urine samples. Samples with abnormal CE-profiles would subsequently be given high priority for more elaborate analysis with GC-MS or MS/ MS.

Finally, related to quantitative aspects, several authors have validated the methods previously described and when these methods have been compared with other well-established separation techniques there was a good agreement in the results. Moreover, interlaboratory assays are being run to submit methods to regulatory authorities to be considered as official methods. The main drawbacks are the lack of sufficient concentration sensitivity in some demanding analysis and of CE-trained personnel in many quality control laboratories. The first point relays on new technical developments for its solution while the second is just a question of detecting the need.

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8 References

[1] Lehotay, D. C., *Biomed. Chromatogr.* 1991, *5*, 113–121.

[2] Presto Elgstoen, K. B., Zhao, J. Y., Anacleto, J. F., Jellum, E., *J. Chromatogr. A* 2001, *914*, 265–275.

[3] Sweetman, L., Sweetman, L. in: Hommes, F. A. (Eds.), *Techniques in Diagnostic Human Biochemical Genetics: a Laboratory Manual*, Wiley-Liss, New York 1991, p. 143.

[4] Seymour, C. A., Thomason, M. J., Chalmers, R. A., Addison, G. M., Bain, M. D., Cockburn, F., Littlejohns, P., Lord, J., Wilcox, A. H., *Health Technol. Assess.* 1997, *1*, 1–95.

[5] Soga, T., Wakaura, M., *J. Am. Soc. Brew. Chem.* 1997, *55*, 44–46.

[6] Soga, T., Ross, G. A., *J. Chromatogr. A* 1997, *767*, 223–230.

[7] Klampfl, C. W., Katzmayr, M. U., *J. Chromatogr. A* 1998, *822*, 117–123.

[8] Klampfl, C. W., *J. Agric. Food Chem.* 1999, *47*, 987–990.

[9] Ackermans, M. T., Ackermans-Loonen, J. C. J. M., Beckers, J. L., *J. Chromatogr.* 1992, *627*, 273–279.

[10] Izco, J. M., Tormo, M., Jimenez-Flores, R., *J. Dairy Sci.* 2002, *85*, 2122–2129.

[11] Izco, J. M., Tormo, M., Jimenez-Flores, R., *J. Agric. Food Chem.* 2002, *50*, 1765–1773.

[12] Jimidar, M., Hamoir, T. P., Foriers, A., Massart, D. L., *J. Chromatogr.* 1993, *636*, 179–186.

[13] Xiong, X., Li, S. F. Y., *J. Chromatogr. A* 1998, *822*, 125–136.

[14] Soga, T., Ross, G. A., *J. Chromatogr. A* 1999, *837*, 231–239.

[15] Kenney, B. F., *J. Chromatogr.* 1991, *546*, 423–430.

[16] Kaniansky, D., Masár, M., Madajová, V., Marák, J., *J. Chromatogr. A* 1994, *677*, 179–185.

[17] Choi, O.-K., Jo, J.-S., *J. Chromatogr. A* 1997, *781*, 435–443.

[18] López Martinez, N., Rodrigues Roldán, A., *Alimentaria* 1993, *6*, 71–74.

[19] Saavedra, L., García, A., Barbas, C., *J. Chromatogr. A* 2000, *881*, 395–401.

[20] Soga, T., Imaizumi, M., *Electrophoresis* 2001, *22*, 3418–3425.

[21] Castro, R., Moreno, M. V. G., Natera, R., Garcia-Rowe, F., Hernandez, M. J., Barroso, C. G., *Chromatographia* 2002, *56*, 57–61.

[22] Wu, C. H., Lo, Y. S., Lee, Y.-H., Lin, T.-I., *J. Chromatogr. A* 1995, *716*, 291–301.

[23] O'Flaherty, B., Yang, W.-P., Sengupta, S., Cholli, A. L., *Food Chem.* 2001, *74*, 111–118.

[24] Horie, H., Yamauchi, Y., Kohata, K., *J. Chromatogr. A* 1998, *817*, 139–144.

[25] Roselló, S., Galiana-Balaguer, L., Herrero-Martínez, J. M., Maquieira, A., Nuez, F., *J. Sci. Food. Agric.* 2002, *82*, 1101–1106.

[26] Jimidar, M., Hartmann, C., Cousement, N., Massart, D. L., *J. Chromatogr. A* 1995, *706*, 479–492.

[27] Fukushi, K., Takeda, S., Wakida, S., Yamana, M., Higasshi, K., Hiiro, K., *J. Chromatogr. A* 1997, *772*, 313–320.

[28] García-Moreno, M. D. V., Jurado-Campoy, C. J., García-Barroso, C., *Eur. Food Res. Technol.* 2001, *213*, 381–385.

[29] Padarauskas, A., Olsauskaite, V., Schwedt, G., *J. Chromatogr. A* 1998, *800*, 369–375.

[30] Haumann, I., Boden, J., Mainka, A., Jegle, U., *J. Chromatogr. A* 2000, *895*, 269–277.

[31] Huang, X., Luckey, J. A., Gordon, M. J., Zare, R. N., *Anal. Chem.* 1989, *61*, 766–770.

[32] Klampfl, C. W., Katzmayr, M. U., Buchberger, W., *Electrophoresis* 1998, *19*, 2459–2464.

[33] Castiñeira, A., Pena, R. M., Herrero, C., Garcia-Martin, S., *J. High Resolut. Chromatogr.* 2000, *23*, 647–652.

[34] Castiñeira, A., Pena, R. M., Herrero, C., Garcia-Martin, S., *J. Food Comp. Anal.* 2002, *15*, 319–331.

[35] Nutku, M. S., Erim, F. B., *J. Microcol. Sep.* 1999, *11*, 541–543.

[36] Arellano, M., Couderc, F., Puig, P., *Am. J. Enol. Vitic.* 1997, *48*, 408–412.

[37] Arellano, M., Andrianary, J., Dedieu, F., Couderc, F., Puig, P., *J. Chromatogr. A* 1997, *765*, 321–328.

[38] Levart, A., Gucek, M., Pihlar, B., Veber, M., *Chromatographia* 2000, *51*, S321–S324.

[39] Dabek-Zlotorzynska, E., Dlouhy, J. F., *J. Chromatogr. A* 1994, *685*, 145–153.

[40] Ahumada, I., Mendoza, J., Escudero, P., Mossert, K., Ascar, L., *J. AOAC Int.* 2001, *84*, 1057–1064.

[41] Dabek-Zlotorzynska, E., Dlouhy, J. F., Houle, N., Piechowski, M., Ritchie, S., *J. Chromatogr. A* 1995, *706*, 469–478.

[42] Souza, S. R., Tavares, M. F. M., Carvalho, L. R. F., *J. Chromatogr. A* 1998, *796*, 335–346.

[43] Devêvre, O., Putra, D. P., Botton, B., Garbaye, J., *J. Chromatogr. A* 1994, *679*, 349–357.

[44] Xu, X., de Bruyn, P. C. A. M., de Koeijer, J. A., Logteberg, H., *J. Chromatogr. A* 1999, *830*, 439–451.

[45] Dahlen, J., Hagberg, J., Karlsson, S., *Fresenius' J. Anal. Chem.* 2000, *366*, 488–493.

[46] Karlsson, S., Wolrath, H., Dahlen, J., *Water Res.* 1999, *33*, 2569–2578.

[47] Zhou, L., Dovletoglou, A., *J. Chromatogr. A* 1997, *763*, 279–284.

[48] Bazzanella, A., Lochmann, H., Mainka, A., Bachmann, K., *Chromatographia* 1997, *45*, 59–62.

[49] Davey, M. W., Bauw, G., Montagu, M. V., *Anal. Biochem.* 1996, *239*, 8–19.

[50] Herrero-Martínez, J. M., Simó-Alfonso, E., Deltoro, V. I., Calatayud, A., Ramis-Ramos, G., *Anal. Biochem.* 1998, *265*, 275–281.

[51] Tenberken, B., Ebert, P., Hartmann, M., Kibeler, M., Mainka, A., Prokop, T., Roder, A., Bachmann, K., *J. Chromatogr. A* 1996, *745*, 209–215.

[52] Tenberken, B., Bachmann, K., *J. Chromatogr. A* 1997, *775*, 372–377.

[53] Fung, Y.-S., Lau, K.-M., *Talanta* 1998, *45*, 641–656.

[54] Hagberg, J., Dahlen, J., Karlsson, S., Allard, B., *Int. J. Environ. Anal. Chem.* 2000, *78*, 385–396.

[55] Barbas, C., Garcia, J. A. L., Mañero, F. J. G., *Phytochem. Anal.* 1999, *10*, 55–59.

[56] Garcia, J. A. L., Barbas, C., Probanza, A., Mañero Gutierrez, F. J., *Phytochem. Anal.* 2001, *12*, 305–311.

[57] Westergaard, B., Hansen, H. C. B., Borggaard, O. K., *Analyst* 1998, *123*, 721–724.

[58] Westergaard Strobel, B., Bernhoft, I., Borggaard, O. K., *Plant Soil* 1999, *212*, 115–121.

[59] Naidu, R., Chen, Z. L., *Chromatographia* 2001, *54*, 495–500.

[60] Xu, J., Chen, Z., Yu, J. C., Tang, C., *J. Chromatogr. A* 2002, *942*, 289–294.

[61] Pantsar-Kallio, M., Kuitunen, M., Manninen, P. K. G., *Chemosphere* 1997, *35*, 1509–1518.

[62] Chen, Z., Tang, C., Yu, J. C., *J. High Resolut. Chromatogr.* 1999, *22*, 379–385.

[63] Prima-Putra, D., Botton, B., *J. Plant Physiol.* 1998, *153*, 670–676.

[64] Schrickx, J. M., Raedts, M. J., Stouthamer, A. H., van Verseveld, H. W., *Anal. Biochem.* 1995, *231*, 175–181.

[65] Krivácsy, Z., Molnár, Á., Tarjányi, E., Gelencsér, A., Kiss, G., Hlavay, J., *J. Chromatogr. A* 1997, *781*, 223–231.

[66] Haddad, P. R., Harakuwe, A. H., Buchberger, W. W., *J. Chromatogr. A* 1995, *706*, 571–578.

[67] Lalljie, S. P., Vindevogel, J., Sandra, P., *J. Chromatogr.* 1993, *652*, 563–569.

[68] Kelly, R. G., Weyant, C. M., Lewis, K. S., *J. Chromatogr. A* 1999, *834*, 433–444.

[69] Desauziers, V., Avezac, M., Fanlo, J. L., *Analusis* 2000, *28*, 163–167.

[70] Oehrle, S. A., *J. Chromatogr. A* 1996, *745*, 81–85.

[71] Sirén, H., Määttänen, A., Riekkola, M.-L., *J. Chromatogr. A* 1997, *767*, 293–301.

[72] Saeed, M., Depala, M., Craston, D. H., *Chromatographia* 1998, *47*, 709–715.

[73] Chen, J., Preston, B. P., Zimmerman, M. J., *J. Chromatogr. A* 1997, *781*, 205–213.

[74] Fiehn, O., Wegener, G., Jekel, M., *Int. J. Environ. Anal. Chem.* 1998, *69*, 257–271.

[75] Soga, T., Ross, G. A., *J. Chromatogr. A* 1999, *834*, 67–71.

[76] Farré, J., Borrull, F., Calull, M., *Chromatographia* 1997, *44*, 235–239.

[77] Swallow, K. W., Low, N. H., *J. Agric. Food Chem.* 1994, *42*, 2808–2811.

[78] Galli, V., Olmo, N., Barbas, C., *J. Chromatogr. A* 2000, *894*, 135–144.

[79] Galli, V., Olmo, N., Barbas, C., *J. Chromatogr. A* 2002, *949*, 367–372.

[80] Volgger, D., Zemmann, A., Bonn, G., *J. High Resolut. Chromatogr.* 1998, *21*, 3–10.

[81] Mallet, S., Arellano, M., Boulet, J. C., Couderc, F., *J. Chromatogr. A* 1999, *853*, 181–184.

[82] Ehmann, T., Bächmann, K., Fabry, L., Rüfer, H., Pahlke, S., Kotz, L., *Chromatographia* 1997, *45*, 301–311.

[83] Fung, Y. S., Tung, H. S., *Electrophoresis* 2001, *22*, 2242–2250.

[84] Roldan-Assad, R., Gareil, P., *J. Chromatogr. A* 1995, *708*, 339–350.

[85] Arellano, M., El Kaddouri, S., Roques, C., Couderc, F., Puig, P., *J. Chromatogr. A* 1997, *781*, 497–501.

[86] Arellano, M., Jomard, P., El Kaddouri, S., Roques, C., Nepveu, F., Couderc, F., *J. Chromatogr. B* 2000, *741*, 89–100.

[87] Orta, D., Mudgett, P. D., Drybread, M., Schultz, J. R., Sauer, R. L., *J. Chromatogr. A* 1998, *804*, 295–304.

[88] Kuban, P., Karlberg, B., *Anal. Chem.* 1997, *69*, 1169–1173.

[89] Nelson, B. C., Uden, P. C., Rockwell, G. F., Gorski, K. M., Aguilera, Z. B., *J. Chromatogr. A* 1997, *771*, 285–299.

[90] Volgger, D., Zemann, A. J., Bonn, G. K., Antal, M. J. Jr., *J. Chromatogr. A* 1997, *758*, 263–276.

[91] Romano, J., Jandik, P., Jones, W. R., Jackson, P. E., *J. Chromatogr.* 1991, *546*, 411–421.

[92] Hiraoka, A., Miura, I., Tominaga, I., Hattori, M., Sasaki, H., *Seibutsu Butsuri Kagaku* 1994, *38*, 233–236.

[93] Shirao, M., Furuta, R., Suzuki, S., Nakazawa, H., Fujita, S., Maruyama, T., *J. Chromatogr. A* 1994, *680*, 247–251.

[94] Schneede, J., Mortensen, J. H., Kvalheim, G., Ueland, P. M., *J. Chromatogr. A* 1994, *669*, 185–193.

[95] Petucci, C. J., Kantes, H. L., Strein, T. G., Veening, H., *J. Chromatogr. B* 1995, *668*, 241–251.

[96] Dolník, V., Dolníkova, J., *J. Chromatogr. A* 1995, *716*, 269–277.

[97] Harrold, M., Stillian, J., Bao, L., Rocklin, R., Avdalovic, N., *J. Chromatogr. A* 1995, *717*, 371–383.

[98] Marsh, D. B., Nuttall, K. L., *J. Capil. Electrophor.* 1995, *2*, 63–67.

[99] Schneede, J., Ueland, P. M., *Anal. Chem.*, 1995, *67*, 812–819.

[100] Chen, H., Xu, Y., Van Lente, F., Jp, M. P. C., *J. Chromatogr. B* 1996, *679*, 49–59.

[101] Chiari, M., Dell'Orto, N., Casella, L., *J. Chromatogr. A* 1996, *745*, 93–101.

[102] Jariego, C. M., Hernanz, A., *Clin. Chem.* 1996, *42*, 477–478.

[103] Willetts, M., Clarkson, P., Cooke, M., *Chromatographia* 1996, *23*, 671–674.

[104] Franke, D. R., Nuttall, K. L., *J. Capil. Electrophor.* 1996, *3*, 309–312.

[105] Sevcik, J., Adam, T., Sazel, V., *Clin. Chim. Acta* 1997, *259*, 73–81.

[106] Jellum, E., Dollekamp, H., Brunsvig, A., Gislefoss, R., *J. Chromatogr. B* 1997, *689*, 155–164.

[107] Zuriguel, V., Causse, E., Bounery, J. D., Nouadje, G., Simeon, N., Nertz, M., Salvayre, R., Couderc, F., *J. Chromatogr. A* 1997, *781*, 233–238.

[108] Barbas, C., Adeva, N., Aguilar, R., Rosillo, M., Rubio, T., Castro, M., *Clin. Chem.* 1998, *44*, 1340–1342.

[109] García, A., Barbas, C., Aguilar, R., Castro, M., *Clin. Chem.* 1998, *44*, 1905–1911.

[110] Procházková, A., Kriványková, L., Boček, P., *J. Chromatogr. A* 1999, *838*, 213–221.

[111] Johnson, S. K., Houk, L. L., Johnson, D. C., Houk, R. S., *Anal. Chim. Acta* 1999, *389*, 1–8.

[112] He, T., Quinn, D., Fu, E., Wang, Y. K., *J. Chromatogr. B* 1999, *727*, 43–52.

[113] Saavedra, L., Barbas, C., *J. Chromatogr. B* 2002, *766*, 235–242.

[114] Holmes, R. P., *Clin. Chem.* 1995, *41*, 1297–1301.

[115] Fu, C., Wang, L., Fang, Y., *Talanta* 1999, *50*, 953–958.

[116] Nelson, B. C., Rockwell, G. F., Campfiel, T., O'Grady, P., Hernandez, R. M., Wise, S. A., *Anal. Chim. Acta* 2000, *410*, 1–10.

[117] García, A., Muros, M., Barbas, C., *J. Chromatogr. B* 2001, *755*, 287–295.

[118] Issaq, H. J., Delviks, K., Janini, G. M., Muschik, G. M., *J. Liq. Chromatogr.* 1992, *15*, 3193–3201.

[119] Caslavska, J., Gassmann, E., Thormann, W., *J. Chromatogr. A* 1995, *709*, 147–156.

[120] Shirao, M. K., Suzuki, S., Kobayashi, J., HiroyukiN., Mochizuki, E., *J. Chromatogr. B* 1997, *693*, 463–467.

[121] Siren, H., Karjalainen, U., *J. Chromatogr. A* 1999, *853*, 527–533.

[122] García, A. H. M., Jiménez, L. M., Barbas, C., *J. Chromatogr. A* 2000, *871*, 341–350.

[123] Li, X., Jin, W., Weng, Q., *Anal. Chim. Acta* 2002, *461*, 123–130.

[124] Klampfl, C. W., Buchberger, W., Haddad, P. R., *J. Chromatogr. A* 2000, *881*, 357–364.

[125] Lindeberg, J., *Food Chem.* 1996, *55*, 73–94.

[126] Craston, D. H., Saeed, M., *J. Chromatogr. A* 1998, *827*, 1–12.

[127] Nuttall, K. L., Guzman, N. A., *Pathology and Laboratory Medicine. Clinical and Forensic Applications of Capillary Electrophoresis*, Humana Press, Totowa, NJ 2001, p. 193.

[128] Fan, X., Deng, Y., *J. Chromatogr. A* 2002, *979*, 417–424.

[129] Jellum, E., Dollekamp, H., Blessum, C., *J. Chromatogr. B* 1996, *683*, 55–65.

[130] Presto Elgstoen, K. B., Jellum, E., *Electrophoresis* 1997, *18*, 1857–1860.

[131] Barbas, C., García, A., Miguel, L., Simó, C., *J. Chromatogr. B* 2002, *780*, 73–82.

[132] Shihabi, Z. K., Friedberg, M. A., *Electrophoresis* 1997, *18*, 1724–1732.

[133] Doble, P., Haddad, P. R., *J. Chromatogr. A* 1999, *834*, 189–212.

[134] Changquing, L., Ting, M., Xiaolin, F., Bowen, M., *SEPU* 1997, *15*, 173–175.

[135] Dabek-Zlotorzynska, E., Dlouhy, J. F., *J. Chromatogr. A* 1994, *685*, 145–153.

[136] Klampfl, C. W., Katzmayr, M. U., Buchberger, W., Basener, N., *J. Chromatogr. A* 1998, *804*, 357–362.

[137] Chen, M.-J., Chen, H.-S., Lin, C.-Y., Chang, H.-T., *J. Chromatogr. A* 1999, *853*, 171–180.

[138] Jonson, S. K., Houk, L., Johnson, D. C., Houk, R. S., *Anal. Chim. Acta* 1999, *389*, 1–8.

[139] Hagberg, J., *J. Chromatog. A* 2003, *988*, 127–133.

[140] Kodama, S., Yamamoto, A., Matsunaga, A., Matsui, K., Nakagomi, K., Hayakawa, K., *J. Agric. Food Chem.* 2002, ACS ASAP.

[141] Sewell, A. C., Heil, M., Podebrad, F., Mosandl, A., *Eur. J. Pediatr.* 1998, *157*, 185–191.

[142] Barbas, C., Saavedra, L., *J. Sep. Sci.* 2002, *25*, 1190–1196.

[143] Durand-Zaleski, I., Saudubray, J. M., Kamoun, P. P., Blum-Boisgard, C., *Int. J. Technol. Assess. Health Care* 1992, *8*, 471–478.

[144] Chalmers, R. A., Purkiss, P., Watts, R. W., Lawson, A. M., *J. Inher. Metab. Dis.* 1980, *3*, 27–43.

[145] Chalmers, R., Lawson, A., *Organic Acids in Man.*, Chapman and Hall, New York 1982.

[146] Millington, D. S., Kodo, N., Norwood, D. L., Roe, C. R., *J. Inher. Metab. Dis.* 1990, *13*, 321–324.

[147] Kuhara, T., Shinka, T., Inoue, Y., Ohse, M., Zhen-wei, X., Yoshida, I., Inokuchi, T., Yamaguchi, S., Takayanagi, M., Matsumoto, I., *J. Chromatogr. B* 1999, *731*, 141–147.

[148] Van den Bergh, F. A., del Canho, H., Duran, M., *J. Inher. Metab. Dis.* 1992, *15*, 897–898.

[149] Collins, J. G., *Vital and Health Statistics-Prevalence of Selected Chronic Conditions*, Department of health and human services. Centers for disease control and prevention/National center for health statistics, Maryland (USA) 1997, *Vol. 10 (194)*.

[150] Barbas, C., García, A., Saavedra, L., Muros, M., *J. Chromatogr. B* 2002, *781*, 433–455.

[151] Tuchman, M., Ulstrom, R. A., *Year Book Medical Publishers, Inc.* 1985, 469–507.

[152] Schweisguth, O., *J. Pediatr. Surg.* 1968, *3*, 118–120.

[153] Young, J. L., Ries, L. G., Silverber, E., Horm, J. W., Miller, R. W., *Cancer* 1986, *58*, 598–602.

[154] Fauler, G., Leis, H. J., Huber, E., Schellauf, C., Kerbl, R., UrbanC., Gleispach, H., *J. Mass Spectrom.* 1997, *32*, 507–514.

[155] Tuchman, M., Lemieux, B., Auray-Blais, C., Robison, L. L., Giguere, R., McCann, M. T., Woods, W. G., *Pediatrics* 1990, *86*, 765–773.

[156] Seviour, J. A., McGill, A. C., Craft, A. W., Parker, L., Bell, S., Cole, M., Smith, J., Hawkins, E., Brown, J., Gordon, A. D., *Am. J. Pediatr. Hematol./Oncol.* 1992, *14*, 332–336.

[157] Tokuda, T., Tokieda, T., Anazawa, A., Yoshioka, M., *J. Chromatogr.* 1990, *530*, 418–423.

[158] Kawaguchi, S., Hirachi, N., Fukamachi, M., *J. Chromatogr.* 1991, *567*, 11–19.

[159] Seviour, J. A., McGill, A. C., Dale, G., Craft, A. W., *J. Chromatogr.* 1988, *432*, 273–277.

[160] Clauson, R. C., *Research Methods in Neurochemistry*, Plenum Publishing, New York 1985, *Vol. 6*.

[161] Wielders, J. P. M., Mink, J. K., *J. Chromatogr.* 1984, *310*, 379–385.

9 Addendum

Table 1. Determination of short-chain organic acids and inorganic anions in food and beverages by CE

Sample	Analytes	Sample pretreatment	Electrophoretic conditions	Time (t_{anal}, min)	LOD	Ref.
Beer	Oxalic, formic, malic, citric, succinic, pyruvic, acetic, lactic, and pyroglutamic acid, Cl^-, SO_4^{2-}, PO_4^{3-}	Degassing by sonication and dilution 1:5 with water	Fused-silica capillary (72 cm × 75 μm ID) Indirect UV detection at 350 nm with reference at 200 nm −25 kV potential t_{inj} = 2 s T^a = 20°C 5 mM PDC, 0.5 mM CTAB, pH 5.6	7	0.6–1.6 mg/L	[5]
Beer	Oxalic, formic, malic, citric, succinic, pyruvic, acetic, lactic, and pyroglutamic acid, Cl^-, SO_4^{2-}, PO_4^{3-}	Degassing by sonication and dilution 1:5 with water	Fused-silica capillary (72 cm × 75 μm ID) Indirect UV detection at 350 mm with reference at 200 nm −25 kV potential t_{inj} = 2 s T^a = 20°C 5 mM PDC, 0.5 mM CTAB, pH 5.6	8	0.9–2.5 mg/L with 3:1 signal-to-noise ratio	[6]
Beer	Oxalic, formic, citric, malic, succinic, acetic, lactic, pyroglutamic, and pyruvic acid, Cl^-, SO_4^{2-}, PO_4^{3-}	Dilution 1:10 and degassification	Fused-silica capillary (48 cm × 50 μm ID) for UV detection and (60 cm × 50 μm ID) for conductivity detection (a) Indirect UV detection at 254 nm; (b) conductivity detection −30 kV potential t_{inj} = 0.2 min 7.5 mM *p*-AB containing 0.12 mM TTAB, pH 5.75 with His	10	(a) 0.117–0.229 mg/L (b) 0.034–0.667 mg/L	[7, 8]
Bread	Propionic acid	Dilution with water and sonication	eCAP capillary tubing (40 cm × 75 μm ID) Indirect UV detection at 214 nm 10 kV potential t_{inj} = 3 s 5 mM Tris, pH 4.6 with benzoic acid	15	0.03–0.08 mM	[9]
Cheese and yogurt	Oxalic, formic, citric, succinic, orotic, uric, pyruvic, acetic, propionic, lactic, sulfuric, and butyric acid	Acidification with H_2SO_4, centrifugation and filtration with 0.45 μm	Fused-silica capillary (105 cm × 75 μm ID) Indirect UV detection at 230 and 300 nm −25 kV potential t_{inj} = 10 s T^a = 25°C 20 mM PDC, 0.5 mM CTAB, pH 12.15	20	0.2–5.7 × 10^{-2} mM	[10]
Dairy products (cheddar cheese and plain liquid yogurt)	Oxalic, citric, formic, succinic, orotic, uric, pyruvic, acetic, propionic, lactic, sulfuric, and butyric acid	Acidification with H_2SO_4, centrifugation and filtration with 0.45 μm	Fused-silica capillary (105 cm × 75 μm ID) Indirect UV detection at 200 nm −25 kV potential t_{inj} = 2 s T^a = 30°C 4.4 mM potassium hydrogen phthalate, 0.27 mM CTAB, pH 11.2	18		[11]

Table 1. Continued

Sample	Analytes	Sample pretreatment	Electrophoretic conditions	Time (t_{anal}, min)	LOD	Ref.
Diet cola soft drinks and artificial sweetening powders	Benzoic acid	Degassing, and dilution with water	Fused-silica capillary (52 cm × 75 μm ID) Direct UV detection at 214 mm 15 kV potential t_{inj} = 30 s Phosphate buffer μ = 0.025, pH 11	9	2–5 mg/L with 3:1 signal-to-noise ratio	[12]
Fruit juices	Ascorbic, sorbic, benzoic, malic, tartaric, maleic, lactic, acetic, malonic, and oxalic acid, ClO_4^-	Filtration with 0.45 μm or dilution	Fused-silica capillary (38 cm × 50 μm ID) Indirect photometric detection at 210 mm −20 kV potential t_{inj} = 15 s T^a = 25°C 4 mM 1,2 dimethylimidazole, 1 mM TMA, 2.86 mM 18-crown-6, pH 7.5	6	0.08–5 mg/L	[13]
Fruit juices, nutrient tonic and soy sauce	Oxalic, formic, malic, citric, succinic, pyroglutamic, acetic, and lactic acid, Br^-, Cl^-, NO_2^-, NO_3^-, SO_4^{2-}, F^-, $P_2O_7^{4-}$, PO_4^{3-} BO_3^{3-}	Dilution with water and centrifugation	Fused-silica capillary (104 cm × 50 μm ID) Indirect UV detection at 350 nm with reference at 230 nm −30 kV potential t_{inj} = 6 s T^a = 15°C 20 mM PDC, 0.5 mM CTAH, pH 12.1	18	6–12 mg/L with 3:1 signal-to-noise ratio	[14]
Fruit juices, soy sauce and wines	Acetic, malic, succinic, lactic, citric, butyric, and tartaric acid	Microfiltration, dilution with water and filtration, 0.45 μm Millex HV	Fused-silica capillary (100 cm × 75 μm ID) Indirect UV detection at 254 nm −20 kV potential t_{inj} = 45 s 5 mM potassium phthalate, 0.5 mM OFM, pH 7.0	15	1 mg/L with 3:1 signal-to-noise ratio	[15]
Fruit juices, wine, margarine and marmalade	Sorbic acid	Dilution and filtration	Fluorinated ethylene-propylene capillary (20 cm × 30 μm ID) Indirect UV detection at 254 nm 100 mM MES, 10 mM Bris-Tris, 0.2% PEG, pH 5.2	5	5×10^{-4} mM	[16]
Fruits, vegetables, juice and drinks	Ascorbic acid	Centrifugation and filtration, 0.45 μm	Fused-silica capillary (27 cm × 57 μm ID) Indirect UV detection at 254 and 265 nm 10–30 kV potential t_{inj} = 5 s T^a = 25°C 100 mM sodium borate, pH 8.0	2	0.06 mg/L with 3:1 signal-to-noise ratio	[17]
Juices	Citric, isocitric, and tartaric acid	Filtration 0.20 μm	Fused-silica capillary (50 cm × 75 μm ID) Indirect UV detection at 254 nm −10 kV potential t_{inj} = 1 s 50 mM phthalic acid, 0.5 mM CTAB, pH 7.0 and 20% methanol	12	60 mg/L	[18]

Table 1. Continued

Sample	Analytes	Sample pretreatment	Electrophoretic conditions	Time (t_{anal}, min)	LOD	Ref.
Orange juices	Citric, tartaric, isocitric, and malic acid	Dilution and filtration 0.45 μm	Neutral polyacrylamide-coated capillary (57 cm × 50 μm ID) Direct UV detection at 200 nm −14 kV potential t_{inj} = 5 s 200 mM phosohate, pH 7.50	11	2–9 mg/L	[19]
Sea urchin and sake	Malic, succinic, acetic, lactic, pyroglutamic, and citric acid, Cl^-, SO_4^{2-}, PO_4^{3-}	Dilution 1:40 v/v with water and ultra-centrifugation	Fused-silica capillary (104 cm × 50 μm ID) Indirect UV detection at 350 nm with reference at 230 and 275 nm −30 kV potential t_{inj} = 6 s T^a = 15°C 20 mM PDC, 0.5 mM CTAH, pH 12.1	20	6–12 mg/L with 3:1 signal-to-noise ratio	[20]
Sherry wine vinegar	Citric, tartaric, malic, succinic, lactic, and acetic acid	Dilution with water	Fused-silica capillary (53 cm × 75 μm ID) Direct UV detection at 185 nm −7 kV potential t_{inj} = 1 s T^a = 20°C 10 mM tetraborate, 0.5 mM TTAOH, 100 mg/L Ca^{2-} and Mg^{2-}, pH 9.3	20	1.3–64.1 mg/L	[21]
Sports drinks, nutrients-added drink, fruit juice, and tea	Citric, oxalic, succinic, acetic, tartaric, malic, lactic, aspartic, glutamic, ascorbic, and gluconic acid	Dilution 10-fold and filtration	Fused-silica capillary (70 cm × 75 μm ID) Indirect UV detection at 220 nm −20 kV potential t_{inj} = 3 s T^a = 25°C (a) 5 mM TMA, 1 mM TTAB, pH 9.0 (b) 5 mM TMA, 1 mM TTAB, pH 5.5	(a) 10 (b) 5	2.0×10^{-3} mM	[22]
Sugar and wine samples	Oxalic, citric, malic, lactic, formic, acetic, and pyroglutamic acid, Cl^-, NO_2^-, NO_3^-, SO_4^{2-}, F^-, $H_2PO_4^-$, HCO_3^-		Fused-silica capillary (17 cm × 50 μm ID) Indirect UV detection at 214/254 nm t_{inj} = 6 s (electrokinetic) at 0.5 kV 0.7 mL of 270 mM sodium chromate, 3.75 mL of 20 mM CTAB and 1.2 mL ACN to 30 mL total volume, pH adjusted by addition of 100 mM NaOH	1	0.08–0.3 mg/L (for anions)	[23]
Tea infusions	Oxalic, citric, malic, aspartic, glutamic, and quinic acid, F^-	Dilution, addition of Na_2EDTA and microfiltration, 0.45 μm	Fused-silica capillary (57 cm × 75 μm ID) Indirect UV detection at 254 nm −20 kV potential t_{inj} = 5 s T^a = 20°C 10 mM sodium chromate, 0.5 mM TTAB, 0.1 mM Na_2 EDTA			[24]

Table 1. Continued

Sample	Analytes	Sample pretreatment	Electrophoretic conditions	Time (t_{anal}, min)	LOD	Ref.
Tomato	Oxalic, malic, and citric acid	Centrifugation and dilution, 0.2 μm	Fused-silica capillary (60 cm × 50 μm ID) Direct UV detection at 200 nm −25 kV potential t_{inj} = 20 s T^a = 20°C 20 mM PDC acid, 0.1% HDM, pH 12.1	18	0.8–1.6 mg/L	[25]
Various vegetables	Oxalic, succinic, citric, formic, acetic, propionic, and butyric acid, Br^-, Cl^-, NO_3^-, NO_2^-, SO_4^{2-}, HPO_4^{2-}, CO_3^{2-}		Fused-silica capillary (52 cm × 75 μm ID) Indirect UV detection at 254 nm −20 kV potential t_{inj} = 10 s 10 mM chromate, 2.30 mM CTAB, pH 11.5	8	0.05 mg/L (for NO_3^- and NO_2^-)	[26]
Vegetables	Ascorbic acid	Homogenization with 2% thiourea-10 mM HCl	Fused-silica capillary (50 cm × 50 μm ID) Direct UV detection at 270 nm −20 kV potential t_{inj} = 3 s T^a = 35°C 20 mM sodium tetraborate, pH 9.2	5	0.35 mg/L with 3:1 signal-to-noice ratio	[27]
Vinic sample	Formic, fumaric, succinic, oxalic, malic, tartaric, acetic, lactic, and citric acid	Desalinization by dilution	Fused-silica capillary (53 cm × 75 μm ID) Direct UV detection at 185 nm −7 kV potential t_{inj} = 30 s 10 mM tetraborate, 0.5 mM TTAOH, 100 mg/L Ca^{2-} and Mg^{2+}, pH 9.3	15	0.08–4.75 mg/L	[28]
Water samples	Cl^-, SO_4^{2-} and HCO_3^-		Fused-silica capillary (50 cm × 75 μm ID) Indirect UV detection at 214 nm −25 kV potential t_{inj} = 5 s (electrokinetic) at 5 kV T^a = 25°C (a) 5 mM imidazole, 2 mM HNO_3, pH 4.0 adjusted with fumaric acid (b) 2.5 mM $Cu(NO_3)_2$, 5 mM ethylenediamine, 1 mM fumaric acid, pH 8.5 adjusted with TEAOH	5		[29]
Drinking water	Oxalic acid, Cl^-, NO_3^-, SO_4^{2-}, ClO_3^-, F^-, Br^-, $S_2O_3^{2-}$		Fused-silica capillary (different measures cm × 75 μm ID) Indirect UV detection at 220 nm Different kV potential t_{inj}, hydrodynamic and electrokinetic (a) 5 mM imidazole, 5 mM thiocyanate, 2 mM citric acid, 1 mM 18-crown-6 (b) 12 mM DIPP, 4 mM TMA, 1.5 mM HIBA, 2.3 mM 18-crown-6, pH 4.8	10	5×10^{-3} mM	[30]

Table 1. Continued

Sample	Analytes	Sample pretreatment	Electrophoretic conditions	Time (t_{anal}, min)	LOD	Ref.
Wine	Tartaric, malic, succinic, acetic, and lactic acid	Dilution in electrolyte by a factor of 10–100	Fused-silica capillary (66 cm × 75 μm ID) Conductivity detection −25 kV potential 7 mM MES/His, 0.5 mM TTAB and 30% methanol, pH 6.0		1×10^{-3} mM with 2:1 signal-to-noise ratio	[31]
Wine	Oxalic, tartaric, citric, malic, succinic, adipic, glutaric, acetic, lactic, butyric, valeric, and shikimic acid, Cl^-, SO_4^{2-}	Dilution 1:100 and filtration, 0.45 μm	Fused-silica capillary (48 cm × 50 μm ID) for UV detection and (60 cm × 50 μm ID) for conductivity detection (a) Indirect UV detection at 254 nm; (b) conductivity detection −30 kV potential t_{inj} = 0.2 min 7.5 mM *p*-AB, 10.5 mM Bis-Tris containing 0.1 mM TTAB, pH 7.0 with LiOH	10	(a) 0.131–0.510 mg/L (b) 0.054–2.750 mg/L	[32]
Wine	Tartaric, malic, succinic, acetic, and lactic acid	Dilution 1:40 with water and filtration, 0.45 μm	Fused-silica capillary (60 cm total length × 75 μm ID) (a) Direct UV detection at 185 nm; (b) Indirect UV detection at 254 mm (a) 20 kV potential, (b) 15 kV potential t_{inj} = 30 s T^a = 25°C (a) 3 mM phosphate, 0.5 mM MTAB, pH 6.5 (b) 7 mM phthalic acid, 2 mM MTAB, 5% v/v methanol, pH 6.1	6	(a) 0.015–0.054 mg/L (b) 1.407–2.296 mg/L	[33, 34]
Wine and fruit juices	Tartaric, malic, citric, lactic, succinic, and acetic acid	Dilution with water and filtration, 0.45 μm	Polyethyleneimine (PEI)-coated silica capillary (45.5 cm/57.3 cm × 75 μm ID) Indirect UV detection at 249 nm −28 kV potential t_{inj} = 0.1 s T^a = 30°C 20 mM 6-aminohexanoic acid, 5 mM 2,4-dihydroxybenzoic acid, pH 4.9	11	3–9 mg/L	[35]
Wines and fruit juices	Oxalic, tartaric, malic, succinic, citric, acetic, and lactic acid Cl^-, NO_3^-, SO_4^{2-}, PO_4^{3-}	Dilution with water and filtration, 0.45 μm	Fused-silica capillary (44 cm × 75 μm ID) Indirect UV detection at 220 nm −20 kV potential t_{inj} = 2 s T^a = 30°C 3 mM PMA, 3 mM EDTA, pH 7.5	11	0.06–1.07 mg/L	[36, 37]

DIPP, dimethyldiphenylphosphonium iodide; HIBA, hydroxyisobutyric acid; TEAOH, tetrabutylammonium hydroxide; TTAOH, tetradecyltrimethylammonium hydroxide

Table 2. Determination of short-chain organic acids and inorganic anions in environmental samples by CE

Sample	Analytes	Sample pretreatment	Electrophoretic conditions	Time (t_{anal}, min)	LOD	Ref.
Air	Oxalic, formic, malonic, glutaric, glycolic, acetic, lactic, and propionic acid, Cl^-, SO_4^{2-}		Fused-silica capillary (50 cm × 50 μm ID) Indirect UV detection at 200 nm −20/−25 kV potential t_{inj} = 5 s (hydrodynamic inj.) and t_{inj} = 5 s (electrokinetic inj. at −5/−15 kV) 5 mM PDC, 0.5 mM CTAB, pH 5.6	5	0.04–0.6 mg/L with 3:1 signal-to-nose ratio	[38]
Air extracts (solid and liquid)	Formic, fumaric, glutaric, adipic, pimelic, suberic, azelic, sebacic, phthalic, methane-sulfonic, carbonic, cetric, chloroacetic, dichloroacetic, propionic, butyric, and benzoic acid		Fused-silica capillary (50 cm × 75 μm ID) Indirect UV detection at 254 nm −20 kV potential t_{inj} = 10 s T^a = 25°C 2 mM NDC, 0.5 mM TTAB and 5 mM NaOH, pH 11	5	100 mg/L	[39]
Aqueous extract of soil	Oxalic, formic, tartaric, pyruvic, citric, lactic, succinic, and acetic acid, Cl^-, NO_3^-, PO_4^{3-}, SO_4^{2-}	Dilution, centrifugation and filtration, 0.2 μm	Fused-silica capillary (52 cm × 75 μm ID) Indirect UV detection at 254 nm −30 kV potential t_{inj} = 30 s T^a = 25°C 10 mM *p*-hydroxybenzoate, 0.5 mM CTAB, pH 4.5	5	0.13–2.67 mg/L	[40]
Atmospheric aerosols	Oxalic acid, Br^-, Cl^-, NO_3^-, NO_2^- and SO_4^{2-}	Filtration, 0.22 μm	Fused-silica capillary (50 cm × 75 μm ID) Indirect UV detection at 254 nm −30 kV potential t_{inj} = 10 s 2.25 mM PMA, 6.5 mM NaOH, 0.75 mM hexamathonium hydroxide, 1.6 mM TEA, pH 7.7–7.9	4	0.035–0.154 mg/L	[41]
Atmospheric particulate matter	β-Hydroxybutyric, acetic, lactic, formic, glycolic, butyric, and propionic acid	Filtration, 0.22 μm	Fused-silica capillary (50 cm × 50 μm ID) Indirect UV detection at 254 nm −15 kV potential t_{inj} = 10 s T^a = 25°C 10 mM 3,5-dinitrobenzoic acid, 0.1 mM CTAB, pH 5–6	7	0.050–0.36 mg/L	[42]
Culture filtrates of soil fungi	Formic, oxalic, pyruvic, maleic, aspartic, glucuronic, acetic, ascorbic, shikimic, gallic, propionic, butyric, fumaric, citric, malic, lactic, succinic, and gluconic acid	Filtration, 0.45 μm	Fused-silica capillary (52.4 × 75 μm ID) Indirect UV detection at 254 nm −20 kV potential t_{inj} = 10 s T^a = 25°C *p*-Hydroxybenzoate, 0.4 mM Ca^{2+}, 2.5% OFM, pH 4.75	8		[43]

Table 2. Continued

Sample	Analytes	Sample pretreatment	Electrophoretic conditions	Time (t_{anal}, min)	LOD	Ref.
Forensic environmental samples	Oxalic, citric, malic, tartaric, formic, acetic, propionic, trichloroacetic, butyric, valeric, and $S_2O_3^{2-}$, Br^-, Cl^-, I^-, NO_2^-, NO_3^-, SO_4^{2-}, F^-, SCN^-, ClO_3^-, HPO_4^{2-}, HCO_3^-, SO_3^{2-}, PO_4^{3-}		Fused-silica capillary (70 cm × 50 μm ID) Indirect UV detection at 210 nm −30 kV potential t_{inj} = 22 s (hydrodynamic)/16 s (electrokinetic injection) at −2 kV T^a = 30°C 3 mM 5-sulfosalicylic acid, 21 mM Tris, pH 8.6	7	1.5×10^{-4}–1×10^{-3} mM (hydodynamic injection)/ 2×10^{-6}–1.3×10^{-6} mM (electrokinetic injection)	[44]
Fresh snow sample (water)	Formic, acetic, propionic, butyric, valeric, oxalic, malonic, succinic, glutaric, adipic, fumaric, maleic, citric, and tartaric acid, Cl^-, NO_3^- and SO_4^{2-}		Fused-silica capillary (75 cm × 75 μm ID) Indirect UV detection at 254 nm −30 kV potential t_{inj} = 30 s (hydrodynamic) and t_{inj} = 45 s (electrokinetic at −5 kV) 5 mM Tris, 2 mM TMA, 0.2 mM TTAB, 0.6 mM Ca^{2-}, pH 8.5	11	0.1–0.2 mg/L (hydrodynamic inj.) and 0.001–0.02 mg/L (electrokinetic inj.) with 2:1 signal to-noise ratio	[45]
Natural waters	Acetic, butyric, β-hydroxybutyric, formic, lactic, maleic, methylsuccinic, malonic, oxalic, succinic, valeric acid, MoO_4^{2-}, HCO_3^-		Fused-silica capillary (43 cm × 75 μm ID) Indirect UV detection at 254 nm −15 kV potential t_{inj} = 45 s (electrokinetic) at 5 kV 5 mM BTA solution (Jassen), 0.5 mM OFM, pH 8	15–20	10^{-3} mg/L	[46]
Novel antifungal lipopeptide	Acetic acid		Fused-silica capillary (56 cm × 75 μm ID) Indirect UV detection at 450 nm with reference at 220 nm −20 kV potential t_{inj} = 3 s T^a = 25°C 4 mM *p*-hydroxybenzoic acid, OFM, pH 6.0 with LiOH	5	0.1 mg/L	[47]
Plant matrices	Oxalic, malonic, fumaric, formic, succinic, tartaric, malic, glutaric, pyruvic, lactic, citric, and ascorbic acid; Cl^-, NO_3^-, PO_4^{3-}, CO_3^{2-}	Dilution with water and filtration, 0.45 μm	Fused-silica capillary (55 cm × 75 μm ID) Indirect UV detection at 232 nm t_{inj} = 3 s 7.5 mM salicylic acid, 15 mM Tris, 500 μM DoTAOH, 180 μM mM $Ca(OH)_2$, pH 8.3	6	5×10^{-4}–2×10^{-3} mM with signal-to-noise ratio of 3	[48]
Plant tissue	Ascorbic and isoascorbic acid	C_{18} SPE (samples are injected in 3% MPA, 1 mM EDTA)	Fused-silica capillary (50 cm × 75 μm ID) Direct UV detection at 260 nm 25 kV potential t_{inj} = 3–10 s T^a = 25°C 200 mM borate, pH 9	10	84 fmol	[49]

Table 2. Continued

Sample	Analytes	Sample pretreatment	Electrophoretic conditions	Time (t_{anal}, min)	LOD	Ref.
Plants	Ascorbic acid	Centrifugation and filtration, 5 μm	Fused-silica capillary (33.5 cm × 50 μm ID) Indirect UV detection at 265 nm −15 kV potential t_{inj} = 20 s T^a = 23°C 60 mM sodium chloride, 60 mM sodium dihydrogen phosphate, 1×10^{-4}% HDM, pH 7	4	0.2 mg/L	[50]
Rain drop	Fomic, acetic, and oxalic acid, Cl^-, NO_3^-, SO_4^{2-}, $C_2O_4^{2-}$, CO_4^{3-}		Fused-silica capillary (63 cm × 75 μm ID) Indirect UV detection at 264 nm −28 kV potential t_{inj} = 30 s 7.5 mM *p*-AB, 750 μM barium hydroxide, 100 μm TTAB(H), pH 9.4	10	fmol	[51]
Rain drop	Oxalic, formic, acetic, propionic, malonic, maleic, azelic, butyric, valeric, and pelargonic acid, Cl^-, NO_3^-, SO_4^{2-}		Fused-silica capillary (50 cm × 50 μm ID) Indirect UV detection at 220 nm −25 kV potential t_{inj} = 45 s T^a = 25°C 20 mM salicylic acid, 32 mM tris-(hydroxy-methyl)amino-methane, 0.001% hexa-dimethrion bromide, pH 8.1	6	32–72 fmol	[52]
Rainwater	Formic and acetic acid Cl^-, NO_3^-, SO_4^{2-}, F^-, HPO_4^{2-}, HCO_3^-	Filtration, 0.45 μm	Fused-silica capillary (65 cm × 75 μm ID) Indirect UV detection at 230 nm −20 kV potential t_{inj} = 60 s (hydrodynamic)/20 s (electrokinetic) at −3 kV T^a = 24°C 5 mM molybdate, 0.15 mM CTAH, 0.01% polyvinyl alcohol, 5 mM Tris, pH 7.9	7	0.5–20 mg/L (hydrodynamic injection)/ 0.1–3 mg/L (electrokinetic injection)	[53]
Rainwater samples and soil extracts	Malonic, oxalic, fumaric, maleic, formic, succinic, tartaric, glutaric, adipic, propionic, butyric, valeric, and citric acid		Fused-silica capillary (76 cm × 75 μm ID) Indirect UV detection at 254 nm −30 kV potential t_{inj} = 45 s at −5 kV (electrokinetic) 5 mM Tris, 2 mM TMA, 0.6 mM TTAB, 0.6 mM $Ca(OH)_2$, pH 8.5	8	5×10^{-4}–5×10^{-3} mg/L with 2:1 signal-to-noise ratio	[54]
Root exudates	Oxalic, formic, fumaric, acetic, malic, citric, succinic, and lactic acid, NO_3^-	Filtration with 0.22 μm and dilution with water	Fused-silica capillary (57 cm × 75 μm ID) Direct UV detection at 200 nm −10 kV potential t_{inj} = 10 s T^a = 25°C 200 mM phosphate, 0.5 mM CTAB, pH 6	14		[55, 56]

Table 2. Continued

Sample	Analytes	Sample pretreatment	Electrophoretic conditions	Time (t_{anal}, min)	LOD	Ref.
Soil	Oxalic, malonic, tartaric, malic, succinic, citric, formic, acetic, propionic, valeric, and lactic acid, Cl^-, NO_3^-, SO_4^{2-}	Centrifugation, addition of Na_4EDTA and filtration, 0.45 μm	Fused-silica capillary (50/70 cm × 75 μm ID) Indirect UV detection at 254 nm −30 kV potential T^a = 20°C (a) 3 mM TMA, 0.02% v/v EDTA, pH 5.8 (b) 8 mM Tris, 2 mM TMA, 0.3 mM TTAB, pH 7.6	10	2.6×10^{-4}–1.77×10^{-3} mM	[57, 58]
Soil and plant extract	Oxalic, formic, fumaric, tartaric, malonic, malic, citric, succinic, maleic, acetic, and lactic acid, Cl^-, NO_3^-, NO_2^-, SO_4^{2-}	Dilution with water, centrifugation and filtration, 0.45 μm	Fused-silica capillary (95.5 cm × 50 μm ID) Direct UV detection at 185 nm −20 kV potential t_{inj} = 30 s T^a = 25°C 25 mM sodium phosphate, 0.5 mM TTAB, 15% CAN, pH 6	12	1×10^{-3}–9×10^{-3} mM with 3:1 signal-to-noise ratio	[59]
Soil and plant tissue extract	Oxalic, fumaric, tartaric, malonic, malic, citric, maleic, phthalic, acetic, benzoic, salicylic, *p*-hrydroxybenzoic, *p*-coumaric, ferulic, and sinapinic acid, NO_3^-, NO_2^-, SO_4^{2-}	Shaking and centrifugation	Fused-silica capillary (70.4 cm × 50 μm ID) Direct UV detection at 190 nm −20 kV voltage t_{inj} = 10 s T^a = 25°C 30 mM phosphate, 1.0 mM TTAB, 20% v/v ACN, pH 6.5	10	1×10^{-3}–8×10^{-3} mM	[60]
Waste water	Formic, acetic, propionic, butyric, oxalic, malonic, succinic, phthalic, and maleic acid, Cl^-, NO_3^-, NO_2^-, F^-, $H_2PO_4^-$, CO_3^{2-}, SO_4^{2-}	Filtration and dilution, 0.45 μm	Fused-silica capillary (60 cm × 50 μm ID) Indirect UV detection at 254 nm −20 kV potential t_{inj} = 30 s 9 mM PDC, 0.5 mM TTAB, pH 7.8	7	0.3–0.6 mg/L	[61]
Water, soil and plant extract	Oxalic, formic, tartaric, aconitic, malic, citric, pyruvic, succinic, acetic, and ascorbic acid, Cl^-, PO_4^{3-}, SO_4^{2-}	Shaking and centrifugation	Fused-silica capillary (70.4 cm × 50 μm ID) Indirect detection at 215 nm −20 kV voltage t_{inj} = 3 s T^a = 20°C 10 mM phthalic acid, MTAB, 5% methanol, pH 5.6	15	5×10^{-3}–0.03 mM	[62]
Xylem exudates	Fumaric, aspartic, glutamic, tartaric, malic, citric, and succinic acid, Cl^-, PO_3^{2-}, SO_4^{2-}	Dilution	Fused-silica capillary (52.5 cm × 75 μm ID) Indirect UV detection at 254 nm −20 kV potential t_{inj} = 20 s 5 mM *p*-hydroxybenzoate containing 0.1 mM Ca^{2+}	5		[63]

DoTAOH, dodecyltrimethylammonium hydroxide; MPA, metaphosphoric acid; TEA, triethanolamine

Table 3. Determination of short-chain organic acids and inorganic anions in industrial processes by CE

Sample	Analytes	Sample pretreatment	Electrophoretic conditions	Time (t_{anal}, min)	LOD	Ref.
Aspergillus niger in recycling culture	Oxalic, formic, acetic, propionic, pyrrolodonic, valeric, capronic, and gluconic acid, Br^-, Cl^-, SO_4^{2-}, PO_4^{3-}		Fused-silica capillary (40 cm × 50 μm ID) Indirect UV detection at 230 mm t_{inj} = 3 s T^a = 30°C Prototype wide-range anion analysis electrolyte containing trimesic acid (Perkin-Elmer/ABD)	9	0.1 mg/L	[64]
Atmospheric aerosol	Oxalic, malonic, formic, succinic, and acetic acid, Cl^-, NO_3^-, SO_4^{2-}		Fused-silica capillary (52 cm × 75 μm ID) Indirect UV detection at 254 nm −20 kV potential t_{inj} = 30 s T^a = 25°C 6 mM chromate, 2.5 cm^3 OFM in 100 cm^3 solution	3.5	0.088– 0.119 mg/L with 2:1 signal-to-noise ratio	[65]
Bayer liquor	Malonic, acetic, citric, tartaric, succinic, formic, and oxalic acid, Cl^-, SO_4^{2-}, F^-, PO_4^{3-}, CO_3^{2-}	Dilution with water	Fused-silica capillary (52 cm × 75 μm ID) Indirect UV detection at 254 nm −20 kV potential t_{inj} = 45 s (a) 3 mM TTAB, 3 mM DTAB, 7.5 mM chromate, pH 9 (b) 5 mM TTAB, 1 mM DTAB, 5.5 mM chromate, pH 9	5	(a) 0.09–0.34 mg/L (b) 0.16–0.88 mg/L	[66]
Chicory root thick juice and beet sugar	Formic, tartaric, malic, citric, succinic, glycolic, acetic,and lactic acid	Thawing where necessary and dilution with water	Fused-silica capillary (53 cm × 75 μm ID) Indirect UV detection at 254 nm −20 kV potential t_{inj} = 20 s 5 mM phthalate, 0.2–0.6 mM Ca^{2+}, 2% OFM, pH 5.6	6		[67]
Corrosion	Acetic and formic acid, Cl^-, NO_2^-, NO_3^-, SO_4^{2-}, HPO_4^{2-}, HCO_3^-, $Cr_2O_4^{2-}$		Fused-silica capillary (53 cm × 75 μm ID) Indirect UV detection at 350 nm with reference at 230 nm −17 kV potential t_{inj} = 30 s (a) 5 mM sodium chromate tetrahydrate and 0.5 mM OFM-OH, pH 8 (b) 12.5 mM potassium phosphate monobasic, 14.8 mM sodium phosphate dibasic, 1 mM OFM-OH, pH 8	6		[68]
Distillery effluents	Acetic, propionic, butyric, and valeric acid		Fused-silica capillary (80 cm × 75/100 μm ID) Direct UV detection at 185 nm 25 kV potential t_{inj} = 45 s (hydrodynamic)/t_{inj} = 45 s (electrokinetic) at 5 kV 7.5 mM Na_2HPO_4, 1 mM OFM-OH, pH 10.2		0.22–0.38 mg/L	[69]

Table 3. Continued

Sample	Analytes	Sample pretreatment	Electrophoretic conditions	Time (t_{anal}, min)	LOD	Ref.
Electronic components	Oxalic acid, Cl^-, NO_3^-, SO_4^{2-}, F^-, HPO_4^{2-}		Fused-silica capillary (60 cm × 75 μm ID) Indirect UV detection at 254 nm −15 kV potential t_{inj} = 30 s (electrokinetic) at −5 kV (a) 10 mM chromate, 1.5 mM OFM-OH, pH 11 (b) 7 mM chromate, 0.5 mM OFM-OH, pH 8		(a) 2×10^{-4}–6.5×10^{-4} mg/L (b) 2.3×10^{-4}–1.16×10^{-3} mg/L with 3:1 signal-to-noise ratio	[70]
Fluids formed in the production of sugar	Hippuric, isovaleric, butyric, propionic, acetic, malonic, pyruvic, and oxalic acid, Cl^-, NO_2^-, NO_3^-, SO_4^{2-}, F^-, PO_3^{3-}, PO_4^{3-}		Fused-silica capillary (60/80 cm × 50 μm ID) Indirect UV detection at 254/260 mm 20–25 kV potential t_{inj} = 3/20 s T^a = 25°C 0.5 mM sulfonated nitronaphthols, pH 8.0	15	1 mg/L with 2:1 signal-to-noise ratio	[71]
Industrial process streams	Succinic and levulinic acid	Dilution with water or water-ACN (30:70 v/v)	Fused-silica capillary (56 cm × 75 μm ID) Indirect UV detection at 310 nm with reference at 210 nm −20 kV potential t_{inj} = 3 s (electrokinetic) at −5 kV T^a = 20°C 5 mM potassium hydrogen phthalate, 2.5% KOH, 0.25 mM CTAB	8	0.5 mg/L	[72]
Industrial samples	Oxalic, malonic, formic, acetic, isovaleric, valeric, isocaproic, phthalic, propionic, butyric, and benzoic, Br^-, Cl^-, NO_2^-, NO_3^-, SO_4^{2-}, F^-, PO_4^{3-}	Dilution	Fused-silica capillary (40 cm × 50 μm ID) Indirect UV detection at 340 nm with reference at 210 nm −20 kV potential t_{inj} = 10 s 5 mM phthalate, 0.25 mM CTAB, pH 7.0	4	0.1–0.2 mg/L	[73]
Industrial wastewater	Oxalic, malonic, succinic, glutaric, adipic, formic, acetic, fumaric, maleic, tartaric, malic, and citric acid	Cation exchangers	Fused-silica capillary (45/50 cm × 50 μm ID) Indirect UV detection at 285 nm 10 kV potential t_{inj} = 10 s 0.6 mM TTAB, 3 mM TMA, pH 10.15	10		[74]
Nickel plating bath sample	Oxalic, formic, lactic, tartaric, malic, citric, acetic, succinic, and oxalic acid, Br^-, Cl^-, NO_3^-, SO_4^{2-}, PO_2^{3-}, PO_3^{3-}, PO_4^{3-}	Dilution with water	Fused-silica capillary (104 cm × 50 μm ID) Indirect UV detection at 350 nm with reference at 230 nm −30 kV potential t_{inj} = 6 s T^a = 15°C 20 mM PDC, 5 mM CTAH, pH 5.7	15	0.8–1.9 mg/L	[75]

Table 3. Continued

Sample	Analytes	Sample pretreatment	Electrophoretic conditions	Time (t_{anal}, min)	LOD	Ref.
Osmotically treated water	Oxalic, formic, fumaric, pyruvic, malonic, maleic, citric, lactic, succinic, aspartic, gluicoric, acetic, ascorbic, shikimic, propionic, and butyric acid, Cl^-, NO_3^-, F^-, $C_2O_4^{2-}$, HPO_4^{2-}		Fused-silica capillary (56 cm × 75 μm ID) Indirect UV detection at 365 nm −15 kV potential t_{inj} 0.1 T^a = 45°C 5 mM Boric acid, 10 mM sodium chromate, 0.03 mM CTAB, 4% butanol, pH 8 with 0.1 M NaOH	10	0.10–0.50 mg/L with 3:1 signal-to-noise ratio	[76]
Orange pulp-wash and water samples from juice processing plants	Cl^-, NO_3^- and SO_4^{2-}	Dilution and filtration	Fused-silica capillary (60 cm × 75 μm ID) Indirect UV detection at 254 nm −20 kV potential t_{inj} = 30 s 5 mM sodium chromate, 0.4 mM OFM, pH 8.0 with lactic acid	2	0.20 mg/L	[77]
Serum of natural rubber latex	Oxalic, formic, fumaric, aconitic, succinic, malic, glutaric, citric, acetic, propionic, glycolic, and quinic acid, NO_3^-	Coagulation and filtration 0.45 μm	Fused-silica capillary (57 cm × 50 μm ID) Direct UV detection at 200 nm −10 kV potential t_{inj} = 5 s T^a = 25°C 0.5 M H_3PO_4, 0.5 mM CTAB, pH 6.25	15	0.002–1.612 mM	[78, 79]
Waste streams from pulp processing	Oxalic, formic, acetic, propionic, and butyric acid, Cl^-, NO_3^-, SO_4^{2-}, SO_3^-, CO_3^{2-}, $S_2O_3^{2-}$, S^{2-}	Acidification and centrifugation	Fused-silica capillary (24.5 cm × 50 μm ID) Indirect UV detection at 185 nm −30 kV potential 5 mM chromate, 32% ACN, 0.001% HDB, pH 10.8	3	0.5–1 mg/L	[80]
Wine residues	Tartaric acid and Cl^-	Sonication and dilution with water	Fused-silica capillary, 30 cm Indirect UV detection at 260 nm −11 kV potential t_{inj} = 1 s 12 mM benzoic acid, 10 mM His and 1 mM TTAB, pH 5.0 with NaOH	2		[81]
Wafer surfaces	Oxalic, formic acid, Cl^-, ClO_3^-, NO_3^-, SO_4^{2-}, Br^-, NO_2^-, F^-, PO_4^{3-}	Electrokinetic sample injection with transient isotachophoretic preconcentration	Fused-silica capillary (40 cm × 50 μm) Indirect UV detection at 350 nm with reference at 245 nm −30 kV potential t_{inj}, electrokinetic 2.25 mM PMA, 6.5 mM NaOH, 1.6 mM triethanolamine, 0.75 mM HDB, pH 7.7	5	50–500 mM	[82]

DTAB, dodecyltrimethylammonium bromide; HDB, hexadimethrine bromide

Table 4. Determination of short-chain organic acids and inorganic anions in miscellaneous samples by CE

Sample	Analytes	Sample pretreatment	Electrophoretic conditions	Time (t_{anal}, min)	LOD	Ref.
Chinese traditional herbs	Oxalic, malonic, formic, fumaric, tartaric, malic, succinic, glutaric, adipic, citric, acetic, propionic, lactic, butyric, 2-hydroxyvaleric, valeric, chlorovaleric, caproic, glutamic, octanoic, quinic, and glucoric acid, Cl^-, NO_3^-, SO_4^{2-}	Drying, digestion, and filtration 0.2 μm	Fused-silica capillary (45 cm × 75 μm ID) Indirect UV detection at 254 nm −18 kV potential t_{inj} = 25 s 15 mM Tris, 30 mM BTA, 1.5 mM TEPA, 20% methanol, pH 8.4 with 0.1 M LiOH	20	2×10^{-3}–8.5×10^{-3} mM	[83]
Coco oil extract	Fatty acids (C_2–C_{14})		Fused-silica capillary (50 cm × 50 μm ID) Indirect UV detection at 270 nm −14 kV potential t_{inj} = 1 s T^a = 30°C 20 mM Tris, 10 mM *p*-anisate, 1 mM trimethyl-β-CD, 50% methanol, pH 8.2	9	2×10^{-4}–5×10^{-4} mg/L	[84]
Culture media	Succinic, pyruvic, acetic, lactic, propionic, 2-hydroxybutyric, butyric, isovaleric, 2-hydroxyvaleric, isocaproic, and 3-phenilpropionic acid		Fused-silica capillary (75 cm × 75 μm ID) Indirect UV detection at 220 nm −14 kV potential t_{inj} = 2 s T^a = 20°C 10 mM benzoic acid, 10 mM His, 1 M Tris-base, 1 mM TTAB, pH 6.0	8	0.02–2.75 mg/L	[85, 86]
Drinking water and condensate samples from Space Shuttle and Mir Space Station	Oxalic, formic, glycolic, glyoxylic, acetic, lactic, propionic, and butyric acid, Br^-, Cl^-, NO_2^-, NO_3^-, SO_4^{2-}, F^-, HPO_4^{2-}	Dilution	Fused-silica capillary (56 × 50 μm ID) Indirect photometric detection at 350 with reference at 200 nm −30 kV potential t_{inj} = 20 s T^a = 20°C Organic acids buffer solution (Hewlett-Packard), pH 5.56/5 mM KHP, 2 mM TTAB, pH 5.56		10^{-3} mg/L	[87]
Orange juices, slurry, liquors from pulp and paper industry and milk	Oxalic, citric, maleic, fumaric, tartaric, succinic, formic, malic, acetic, propionic, lactic, butyric, and benzoic acid, Cl^-, NO_3^-, SO_3^{2-}, F^-, OH^-, HCO_3^-, HPO_4^{2-}, SO_3^{2-}	On-line dialysis in an FIA arrangement	Fused-silica capillary (45 × 50 μm ID) Indirect UV detection at 372 nm 25 kV potential 6 mM sodium chromate, 3 mM borate and 0.032 mM CTAB, 3 mM boric acid, 5% ACN, pH 8.0	10		[88]

Table 4. Continued

Sample	Analytes	Sample pretreatment	Electrophoretic conditions	Time (t_{anal}, min)	LOD	Ref.
Parental nutrition solutions	Oxalic acid		Fused-silica capillary (60 × 75 μm ID) Indirect UV detection at 254 nm −15 kV potential t_{inj} = 10 s 10 mM chromate, 0.5 mM TTAB, 0.1 mM EDTA, pH 8	7	0.24 mg/L with 3:1 signal-to-noise ratio	[89]
Standards	Mesaconic, pyruvic, glyoxylic, citraconic, mesaconic, citric, glutaconic, itaconic, 2-hydroxyisobutyric, acrylic, glutaric, methacrylic, acetic, crotonic, and butyric acid		Fused-silica capillary (24.5 × 50 μm ID) Direct UV detection at 185 nm −10 kV potential t_{inj} = 2 s (electrokinetic), −2 kV Different BGEs tested			[90]

Table 5. Determination of short-chain organic acids in body fluids by CE: organic acids profiling, chiral analysis, nephrolithiasis, and neuroblastoma markers

Sample	Analytes	Sample pretreatment	Electrophoretic conditions	Time (t_{anal}, min)	LOD	Ref.
Standards and human saliva	Formic, succinic, acetic, lactic, and propionic acid	Dilution	Reversed polarity Uncoated capillary Phthalate and commercial EOF modifier (OFM), pH 5.6	6	UV 254 nm Indirect detection	[91]
Foods and urine	Acetic, lactic, citric, tartaric, malic, and succinic acid	It does not work with fumaric and orotic acid	Filtration, 0.45 μm Reversed polarity Uncoated capillary Phthalate and commercial EOF modifier	15	UV 254 nm Indirect detection	[15]
CSF	Lactate and pyruvate	Deproteinization by centrifugation and ultrafiltration	Reversed polarity Uncoated capillary Tetraborate and TTAB, pH 9.2	10	UV 185 nm Direct detection	[92]
Urine	Oxalic, formic, methylmalonic, fumaric, succinic, 2-ketoglutaric . . . acid (n = 12).	20 min centrifugation and SPE C_{18}	Reversed polarity Uncoated capillary Tetraborate and commercial EOF modifier, pH 10.0	12	UV 185 nm Direct detection	[93]
Standards	Oxalic, formic, propionic, fumaric, and others acids (n = 14)	It does not work with oxalic acid	Reversed polarity Uncoated capillary 4-Hydroxybenzoate, commercial EOF modifier and calcium salt, pH 4.75	12	UV 254 nm Indirect detection	[43]

Table 5. Continued

Sample	Analytes	Sample pretreatment	Electrophoretic conditions	Time (t_{anal}, min)	LOD	Ref.
Urine and serum	Methylmalonic and short-chain dicarboxylic acids (n = 6)	Derivatization with 1-pyrenyldiazomethane and dilution with organic solvents (1:15)	Normal polarity Uncoated capillary Phosphate-acetate, 50% organic modifier,pH 4.8	14	Fluorescence LIF He-Cd	[94]
Uremic and normal serum	Uric, hippuric and others acids	Ultrafiltration	Normal polarity Uncoated capillary Borate, pH 9.0	16	UV diode-array	[95]
Serum	Pyruvic, citric, malic, acetoacetic, and lactic acid	No pretreatment	Reversed polarity Capillary coated with linear PAA ε-Aminocaproic and 2-hydroxyphenylacetic acid, pH 3.8	12	UV 220 nm Indirect detection	[96]
Standards	Organic and inorganic acids (n = 13)		Normal polarity Uncoated capillary Sodium tetraborate and barium borate	13	Suppressed conductivity	[97]
Urine	Methylmalonic, citric, 2-ketoglutaric, and succinic acid	Liquid-liquid extraction	Reversed polarity Uncoated capillary Phthalate, phosphate, CTAB and, 30% v/v ACN	<6	UV 210 nm Indirect detection	[98]
Serum	Methylmalonic acid	Deproteinization Derivatization with 1-pyrenyldiazomethane and dilution with organic solvents (1:80)	Reversed polarity Capillary coated with linear PAA Tris-acetate, pH 6.4 Hydroxypropylmethylcellulose and dimethylformamide	25	Fluorescence detection LIF He-Cd	[99]
a) Serum b) Urine	Short-chain organic acids (n = 14)	a) Deproteinization, centrifugation, evaporation, and redissolution b) Filtration and dilution	Normal polarity Uncoated capillary Carbonate and phthalate, pH 7.0	22	UV 230 nm Indirect detection	[100]
Standards	Aliphatic (formic and tartaric) and aromatic acids		Reversed polarity Capillary coated with poly(acryloylaminoethoxy)-ethyl-β-D-glucopyranose Addition of divalent cations	16	UV 185 nm Direct detection	[101]
Urine	Methylmalonic, glutaric, *N*-acetyl-aspartic, aminoadipic, and propionic acid (n = 10)	Cut-off filtration (M_r 10 000) and centrifugation	Reversed polarity Uncoated capillary Sodium sulfate, calcium chloride and commercial EOF modifier	10	UV 185 nm Direct detection	[102]
Urine	Oxalic, malonic, maleic, succinic, pyruvic, lactic, 3-hydroxybutyric, and hippuric acid	SPE C_{18}	Reversed polarity Uncoated capillary Tetraborate and commercial EOF modifier (TTAB) and calcium salt, pH 10.0	30	UV 196 nm Direct detection	[103]

Table 5. Continued

Sample	Analytes	Sample pretreatment	Electrophoretic conditions	Time (t_{anal}, min)	LOD	Ref.
Urine	Orotic acid	Complex pretreatment: centrifugation, SPE C_{18} and cut-off centrifugation	Normal polarity Capillary coated with polyvinyl alcohol Phosphate, pH 3.0	10	Direct detection UV Diode-array	[104]
Urine	Orotic acid	Cation-exchange resin	Reversed polarity Uncoated capillary Phosphate, pH 2.23	7	Direct detection UV Diode-array	[105]
Urine	Mevalonic, glutaric, glyceric, and methylmalonic acid	No pretreatment	Normal polarity Uncoated capillary Borate, pH 8.5	10–15	Direct detection UV Diode-array	[106]
Serum	Short-chain organic acids	Derivatization with 5-bromomethyl-fluorescein	Normal polarity Uncoated capillary a) Borate, pH 10.0 SDS and urea (MEKC) b) Tris and benzoate	10	a) Fluorescent detection LIF argon 488 nm b) Indirect detection at 220 nm	[107]
Urine	Short-and medium-chain organic acids (n = 9)	Centrifugation with ultrafugue filters (M_r 30 000)	Reversed polarity Capillary coated with linear PAA Phosphate, pH 6.0, +10% v/v methanol	15	UV 200 nm Direct detection	[108]
Urine	Short-and medium-chain organic acids (n = 27)	Centrifugation and dilution (1:3)	Reversed polarity Capillary coated with linear PAA a) Phosphate, pH 6.0, + 10% v/v methanol b) Phosphate and acetate, pH 4.0	15	UV 200 nm Direct detection	[109]
a) Urine from healthy people b) Urine from patients	Orotic acid	ITP preconcentration and preseparation on-line with CZE	Normal polarity Uncoated capillary a) Glutamate and spermine, pH 5.2 b) Phosphate and glycine, pH 2.15	6	a) UV 254 nm b) UV 280 nm	[110]
Standards	Succinic, maleic, malonic, and glutaric acid		Reversed polarity Uncoated capillary Naphthalene disulfonate, pyromellitic acid, methanol, and diethylene triamine	30	CE-MS ESI interface Quadrupole	[111]
Urine	Homogentisic, and pyroglutamic acid, and others compounds	No pretreatment	Normal polarity Uncoated capillary Ammonium bicarbonate, pH 8.5	15	CE-MS-MS ESI interface Ion-trap MS	[112]
Urine	Propionic, benzoic, homogentisic, HVA, VMA, glyceric, orotic acid, and more organic acids	Filtration	Normal polarity Uncoated capillary Ammonium acetate, pH 8.5	15	CE-MS-MS ESI interface Triple-quadrupole MS	[2]
Urine, CSF, amniotic fluid	D- and L-lactic acid	Centrifugation and dilution (1:4)	Reversed polarity Capillary coated with linear PAA Phosphate, pH 6.0, and 2-hydroxypropyl-β-cyclodextrin	40	UV 200 nm Direct detection	[113]

Table 5. Continued

Sample	Analytes	Sample pretreatment	Electrophoretic conditions	Time (t_{anal}, min)	LOD	Ref.
Urine	Oxalic and citric acid	Acidification, heating, centrifugation, and dilution	Reversed polarity Uncoated capillary Chromate and TTAB, pH 8.1	5	UV 254 nm Indirect detection	[114]
Urine	Oxalic, ascorbic, and uric acid	Filtration and dilution with BGE (1:200)	Reversed polarity Uncoated capillary Phosphate and CTAB, pH 5.7	12	Amperometric detection 0.8 V *vs.*Ag/AgCl Cobalt phthalocyanine, modified carbon-paste electrode	[115]
Amniotic fluid and plasma	Oxalic acid	Six equilibration solvents mixed with sample Centrifugation and cation (Ag+-form) resin	Reversed polarity Uncoated capillary Chromate, TTAB, EDTA, pH 8	10	UV 254 nm Indirect detection	[116]
Urine	Oxalic, citric, glyoxylic, and glyceric acid	Acidification, and centrifugation	Reversed polarity Capillary coated with linear PAA a) Phosphate, pH 6.0, +10% v/v methanol b) Phosphate and acetate, pH 4.0	10	UV 200 nm Direct detection	[117]
Urine	VMA and HVA	Acidification, liquid-liquid extraction, evaporation, and redissolution	Normal polarity Uncoated capillary Acetate buffer, pH 4.10	12	UV 214 nm Direct detection	[118]
Urine	VMA, HVA, HIA, and others urinary indole derivatives	Centrifugation and dilution	Normal polarity Uncoated capillary MEKC Phosphate-tetraborate buffer with SDS, pH 9.2	10	a) Absorption at 220 nm Direct detection b) Fluorescence at 340 nm	[119]
Urine	Creatinine, VMA, HVA, and uric acid	Centrifugation and dilution	Normal polarity Uncoated capillary MEKC Phosphate buffer with SDS, pH 7.0	15	UV 245 nm Direct detection	[120]
Urine	VMA and HVA (only detection)	No pretreatment	Normal polarity Uncoated capillary Borate, pH 8.5	10	Direct detection UV Diode-array	[106]
Urine	Biogenic amines and VMA, HVA and HIA	a) Hydrolysis with HCl (basic amines) or with NaOH (acidic metabolites) b) Centrifugation and filtration	Normal polarity Uncoated capillary Ammonium acetate buffer, pH 4.0	35	UV 220 nm Direct detection	[121]
Urine	VMA, HVA and HIA	Centrifugation and dilution	Reversed polarity Capillary coated with linear PAA Phosphate-acetate buffer, pH 4.4 with 10% v/v methanol, or pH 4.3 with 5% v/v methanol	30	UV 192 nm Direct detection	[122]

Table 5. Continued

Sample	Analytes	Sample pretreatment	Electrophoretic conditions	Time (t_{anal}, min)	LOD	Ref.
Urine	VMA, HVA and more organic acids	Filtration	Normal polarity Uncoated capillary Ammonium acetate buffer, pH 8.5	15	CE-MS-MS ESI interface Triple-quadrupole MS	[2]
Urine	VMA and HVA	Acidification, liquid-liquid extraction, evaporation, and redissolution	Normal polarity Uncoated capillary Phosphate buffer, pH 5.2	12	Amperometric detection 1.1 v vs. Hg/Hg_2Cl_2 Carbon-fiber microdisk bundle electrode	[123]