- 1
- 2
- 3
- 4
- 5
- 6
- 7
- 8
- 9
- 10
- 11
- 12
- 13
- 14
- 15
- 16
- 17
- 18
- 19
- 20

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Abstract

This review highlights recent developments and applications of on-line sample preconcentration techniques in capillary electrophoresis (CE) from 2010 to April 2013. Various preconcentration techniques based on the analyte velocity change in two or three discontinuous solutions system including field-amplified stacking, transient isotachophoresis, pH-mediated stacking, sweeping, and their modified and combined techniques have been employed to enrich and separate biological, environmental, food, toxicological, forensic and nanoparticle samples in CE. More than 170 published research articles collected from Scopus databases from the year 2010 described the on-line sample preconcentration techniques. This review provides comprehensive tables listing the applications of the on-line sample preconcentration techniques with categorizing by the fundamental preconcentration mechanism and application area.

32	Contents
33	1. Introduction
34	2. Field-amplified stacking
35	2.1. FASS
36	2.2. FASI
37	2.3. LVSS
38	2.4. Stacking in MEKC and other techniques
39	3. Transient isotachophoresis
40	3.1. tITP
41	3.2. EKS
42	4. pH-mediated stacking
43	5. Sweeping and related techniques
44	5.1. Sweeping
45	5.2. transient-trapping, AFMC and MSS
46	5.3. Coupling with FASI and other hyphenated techniques
47	6. Conclusion
48	References
49	

1. Introduction

Capillary electrophoresis (CE) is one of the candidates for the separation of a small amount of analytes in complex samples since it has much merits relative to conventional HPLC in terms of high separation efficiency, short analysis time, simple operation, small consumption of reagents, buffers and samples. Additionally, several separation modes and detection schemes are introduced in CE, so that the application area are extended to various analytes including ionic, neutral, small and large molecules. In spite of such versatilities, poor concentration sensitivity is still problematic due to a low detection volume in CE. To detect a low amount of analytes, three approaches are mainly selected, *i.e.*, highly sensitive detector, on-line and off-line sample preconcentration techniques. Among various detectors, laser-induced fluorescence (LIF), mass spectrometry (MS) and electrochemical (EC) detection are often employed for sensitive CE analysis. However, recent progresses of proteomics, metabolomics and glycomics need further high detection sensitivity to analyze a trace amount of biomolecules. Hence, a coupling of these sensitive detectors and on-line and off-line sample preconcentration techniques is desired. Especially for biosamples, off-line sample preconcentration techniques with sample pretreatment are necessary to extract, isolate and enrich the analytes of interest from complex sample matrices. In recent years, several off-line extraction techniques including solid phase microextraction (SPME), liquid-phase microextraction (LPME), hollow-fiber liquid-phase microextraction (HF-LPME), liquid-liquid-liquid microextraction (LLLME), dispersive LLME (DLLME), single-drop microextraction (SDME), ultrasound-assisted emulsification microextraction (USAEME), solvent-bar microextraction (SBME), electrokinetic membrane extraction (EME), and so on, are combined with CE [1–4]. These methods are effective to enrich and purify target analytes from crude sample matrices prior to the CE separation. To achieve high-throughput and automated analysis, however, these off-line preconcentration and pretreatment techniques are not favorable since they are often tedious due to increased processing steps and time.

Compared with off-line pretreatments, on-line sample preconcentration techniques, where most of the enrichment processes are performed in the separation capillary, can easily improve

the detection sensitivity. The on-line sample preconcentration techniques applied to CE are classified to three categories: preconcentration by analyte velocity change in two or three discontinuous solutions system, focusing, and solid phase extraction (SPE). Focusing techniques such as isoelectric focusing (IEF) are used for the preconcentration and pre-separation [5]. SPE techniques concentrate analytes at the solid-phase immobilized capillary. Although the SPE approaches can give high preconcentration efficiency and/or highly-selective separation performance, they often requires labor-intensive preparation processes and complicated experimental procedures. On the other hand, the on-line sample preconcentration based on the changes in the analyte migration velocity at the boundary of discontinuous solution systems can be carried out with easy experimental procedure and relatively short enrichment time. For the stacking at discontinuous solutions boundary, target analytes are dissolved in a sample matrix whose components differ from those in a background solution (BGS). At the boundary between the sample and BGS zones, the changes in the migration velocity of the analytes should occur according to the difference in the electric field, pH and the retention factor, resulting in the preconcentration of the analytes in the long sample zone. These techniques can be combined with several separation modes such as capillary zone electrophoresis (CZE), cyclodextrin modified-CZE (CDCZE), electrokinetic chromatography (EKC), micellar EKC (MEKC), cyclodextrin EKC (CDEKC), cyclodextrin modified-MEKC (CDMEKC), microemulsion EKC (MEEKC), capillary gel electrophoresis (CGE), non-aqueous CE (NACE) and isotachopheresis (ITP).

In this review, recent developments and applications of the on-line sample preconcentration techniques mainly based on the migration velocity change in “capillary”-based electrophoresis from the year 2010 are briefly reviewed because various review papers on the on-line sample preconcentration have been published [6–22]. For detailed mechanism and discussion, these papers and references therein should be referred. Principal on-line sample preconcentration techniques include field-amplified stacking, transient isotachopheresis, pH-mediated stacking, and sweeping techniques. Please refer to Tables 1~6 for an overview of applications of the on-line sample preconcentration techniques in CE.

2. Field-amplified stacking

2.1. Field-amplified sample stacking (FASS)

Among several on-line sample preconcentration techniques developed in CE, field-amplified sample stacking (FASS) is the simplest and most common technique. In FASS, a low-conductivity sample solution is hydrodynamically injected into the capillary filled with a high-conductivity BGS. Because the electric field strength is inversely proportional to the electrical conductivity, a higher field is applied to the sample zone relative to the BGS zone. Hence, the electrophoretic migration of ionic analytes in the sample zone is faster than that in the BGS, which causes the “stacking” of the analytes around the sample/BGS boundary. Although the conductivity of the sample is limited to be low in FASS, up to 100-fold increase in the detection sensitivity can be obtained by injecting a long sample plug (typically, < 5 cm).

FASS and modified techniques were employed to many applications including biological, environmental, food, and pharmaceutical areas as summarized in Table 1 [23–44]. As a typical biological application, Liu *et al.* reported FASS-CZE-LIF analysis of fluorescein isothiocyanate (FITC)-labeled catecholamines in rat brain, which provided the enrichment factor of 118~132 and the limit of detection (LOD) of 22 pg/L [25]. For the determination of cationic and anionic neurochemicals, a PEO-based stacking technique was applied to CZE [32]. In the modified technique, in addition to the FASS effect, ionic neurochemicals can be stacked through the viscosity difference between a sample and BGS zones, giving 116~281-fold sensitivity improvements for biogenic amines and their metabolites. In recent years, FASS has been often combined with on-line or off-line extraction techniques, such as SPE and LLME, to purify crude samples and improve the detection sensitivity. For example, a coupling of FASS with dispersive LLME (DLLME) resulted in the 814-fold sensitivity improvement for an antidepressant drug, sertraline, which belongs to the class of selective serotonin reuptake inhibitors [38]. Khan and co-workers reported a novel quantification of microRNA based on off-line coupling of SPE using RNA binding protein-coated magnetic

beads with FASS in a protein facilitated affinity capillary electrophoresis (ProFACE) assay for the detection of ultralow amounts of microRNA [41]. As a typical result, LOD was 0.5 fM or 30000 microRNA molecules in 1 mL of serum. Such selective pre-extraction technique is quite effective for desalting of the bio-samples, which is requisite for the field-amplified stacking.

2.2. Field-amplified sample injection (FASI)

To obtain higher enrichment efficiency, a field-amplified sample injection (FASI) technique is available [45–70]. In FASI, ionic analytes are electrokinetically injected from the inlet vial filled with a sample solution to the capillary according to the FASS concentration mechanism, which can provide the introduction of a larger amount of the analytes compared to FASS. Jin *et al.* reported the sensitivity enhancement factors (SEFs) between 62000 and 200000-fold for the FASI-CDCZE analysis of β -blockers with the sample injection for 5 min at +10 kV after introducing a short plug of water for 10 s at 0.5 psi to improve the SEF and reproducibility [62]. In place of pure water plug, Lau *et al.* effectively used a solvent plug containing 45% acetonitrile. The solvent plug was introduced hydrodynamically into the capillary for 40 s before injecting four toxic metal ions electrokinetically at +10 kV for 60 s in the FASI step, yielding the SEF of up to 91800-fold with the LODs ranging from 0.005 to 2.32 mg/L [53].

As an alternative sensitivity enhancement approach in FASI, a pressure-assisted electrokinetic injection (PAEKI) method was applied. PAEKI utilizes the principle of FASI to achieve a theoretically unlimited enrichment power by applying an external pressure to balance the EOF in opposite for a potentially unlimited injection time. The balance creates a stationary boundary at the inlet of the capillary where the target analytes accumulate in a narrow sample zone according to principles of FASS. Feng and co-workers proposed a counter-ion layer theory to describe the working mechanism of PAEKI [71]. Compared to the diffusion behavior of hydrodynamically injected plugs, interestingly, it was found that PAEKI plugs experienced less band broadening overall, attributing to higher concentration efficiencies. This method was successfully applied to the enrichment of inorganic ions [71], haloacetic acids [72] and perchlorate [73] with up to 20000-fold increase in the sensitivity.

As a unique approach, Zhang *et al.* combined FASI with two-end sample injection, namely two-end FASI, for the simultaneous stacking and separation of positive and negative compounds under a suppressed EOF condition [74]. Over 1000-fold enhancement in the sensitivity was gained for both cationic matrine and andoxymatrine, and anionic 5-sulfosalicylic acid.

2.3. Large volume sample stacking (LVSS)

In the large volume sample stacking (LVSS) technique, a large volume (up to the entire capillary volume) of a low-conductivity sample is injected hydrodynamically into the capillary. Since the stacking occurs at the sample/BGS boundary, the effective separation length is reduced as the sample injection length is increased. In addition, the electric field strength in the BGS zone is decreased by the remained sample matrix. Hence, the remained matrix should be removed immediately after the field-amplified sample stacking. The sample matrix removal is carried out with or without polarity switching. In LVSS with polarity switching, the sample matrix is removed via EOF, while the stacked zone gradually moves back towards the inlet. Before the stacked analyte zone exits the inlet of the capillary, the electric polarity is reversed for the separation. In recent years, LVSS with polarity switching are employed to the CZE analysis of parabens [75], barbiturates [76], polyphenolic compounds [77], flavonoids [78], mercury ions [79], herbicides [80], and so on [81, 82]. However, LVSS with polarity switching requires the current monitoring to determine the timing of the switching, which sometimes causes irreproducibility. On the other hand, LVSS without polarity switching can be performed by suppressing or direction-inversion of the EOF during the matrix removal [83]. In the case of using the EOF as a pump to remove the sample matrix in LVSS, namely a large-volume sample stacking with an EOF pump (LVSEP), both the sample enrichment and the matrix removal occurs simultaneously under a constant voltage [84–92]. Recently, Kawai *et al.* used polymer coated-capillaries to suppress the EOF, which was applied to the LVSEP analysis of various biogenic compounds including oligosaccharides, cationic amines, enantiomers, and proteins [85–87]. Wang *et al.* reported a combination of LVSEP with anion-selective exhaustive injection (ASEI), where a large amount of anionic analytes are

electrokinetically injected, to achieve highly sensitive enantioseparation of trace fenoprofen and amino acid derivatives by using vancomycin as the chiral selector [89]. Since successful LVSEP-ASEI needs a very low EOF, poly(dimethylacrylamide) coated capillary was employed. Under an optimized condition, more than 1000-fold enhancement in detection sensitivity compared with the normal injection was achieved by LVSEP-ASEI. Very recently, LVSEP was coupled with PAEKI to inject an extremely large volume of a sample (up to 200 μ L) due to a counter pressure-flow against the EOF [90]. As a result, the sensitivity increase of 1100000 and 2800-10000 was achieved for fluorescein and glucose ladder, respectively. From the first report on LVSEP [93], a high-pH sample and a low-pH BGS has been employed to control the EOF rate for the matrix removal and separation steps, respectively. On the other hand, in the recent LVSEP applications, the polymer-coated capillaries were utilized to suppress both the EOF and the analytes adsorption onto the inner surface of the capillary, which is effective for the on-line preconcentration of biogenic compounds.

2.4. Stacking in MEKC and other techniques

Stacking technique was first introduced in CZE to concentrate ionic analytes. Normal stacking technique cannot enrich neutral analytes due to no electrophoretic mobility. In the application of the field-amplified stacking to neutral analytes, hence, charged micellar pseudostationary phase (PSP), *e.g.*, sodium dodecyl sulfate (SDS), should be employed. In the recent three years, the combination of the field-amplified stacking with MEKC was applied to the analysis of a wide range of analytes (Table 2) [94–101]. Oita *et al.* reported that up to 14-fold signal enhancement can be successfully achieved for the suspension of poliovirus (PV) particles by normal FASS in MEKC [94]. In the analysis of the PV particles, the reduction of the conductivity of the sample matrix was essential to obtain not only the signal enhancement but reproducible results. Liu *et al.* employed the reversed electrode polarity stacking mode (REPSM) in MEKC for the concentration and size-separation of gold nanoparticles (GNPs) [99, 100]. REPSM is a modified technique of the FASS with polarity switching, *i.e.*, a low-conductivity sample is injected for a long time into the capillary filled with a high-conductivity micellar BGS, and then a negative polarity is applied [102]. After the

removal of the vacant sample matrix, the polarity is switched to start the MEKC separation. REPSM-MEKC was quite effective for separating GNPs, providing high performance in terms of the resolution and the plate numbers, both of which were improved by greater than five-fold relative to previous reports. Notably, the SEFs for the 5.3- and 40.1-nm-diameter GNPs were improved to 20- and 500-fold, respectively, compared to those obtained using conventional CE separation. Strictly speaking, since these nanoparticles are not incorporated into the micelles, the separation mode may be categorized not to MEKC, but surfactant-modified CZE. In MEKC, stacking with reverse migrating micelles (SRMM) or pseudostationary phase (SRMP) is also useful technique, which is similar to LVSEP under the acidic condition. Hefnawy *et al.* investigated the combination of microemulsion electrokinetic capillary chromatography (MEEKC) with a modified SRMP with polarity switching for analyzing the ultra-short hypnotic HIE-124, which is a novel intravenous anaesthetic agent [101]. The proposed method gave a selective preconcentration with over 25-fold improvement of sensitivity and good quantification of HIE-124 in mice serum.

As a novel on-line preconcentration technique using the field-amplified stacking technique, successful coupling of electrochemical preconcentration (EPC) based on stripping voltammetry with FASS-CZE was reported by Lopes *et al.* for the first time (Table 3) [103, 104]. The EPC-CE interface comprises a dual glassy carbon electrode block, a spacer and an upper block with flow inlet and outlet, pseudo-reference electrode and a fitting for the CE silica capillary (Fig. 1). For the EPC, traces of heavy metal ions were extracted from a flowing sample and accumulated on a glassy carbon working electrode by electro-reduction for some minutes. Anodic stripping of the accumulated metals was synchronized with hydrodynamic injection into the capillary. After the EPC, the injected analytes were concentrated by FASS, separated by CZE and detected by contactless conductivity detection (C⁴D) with enhancement factors close to 50 for thallium, lead, cadmium and copper ions, and about 16 for zinc ion. Besides preconcentration, easy matrix exchange using a low conductivity buffer between accumulation and stripping/injection can be carried out in EPC, which is advantageous to promote FASS and improve separation of highly saline samples.

Recently, Wu *et al.* reported a novel electrokinetic stacking technique at an HF etched porous junction on a fused-silica capillary [105]. The HF etched porous junction actually behaved as a nanofluidic interface, with which an ion concentration polarization effect-based electrokinetic stacking (ES) inside the capillary can be carried out simply by manipulation of the voltage. It should be emphasized that, in the preparation of etched capillary, over-etching was avoided with the autostop etching principle (Fig. 2a), and an extremely high stacking rate at the junction was observed. It was demonstrated with the same junction that all charged species, whatever the size (big or small) and charge type (negative or positive), can be effectively stacked inside the capillary and over 9 orders of magnitude concentration can be achieved (Fig. 2b). Considering the widespread use of the capillary, this electrokinetic stacking technique will find more applications in CE for the analysis of low abundance biomolecules.

3. Transient isotachophoresis (tITP)

3.1. tITP

In employing the isotachophoresis (ITP) phenomenon as the preconcentration technique, another separation mode *e.g.* is often coupled with ITP to attain both effective preconcentration and separation. Such technique is termed transient ITP (tITP) [106, 107], in which the preconcentration by ITP and the separation by CZE, MEKC, capillary gel electrophoresis (CGE) or non-aqueous CE (NACE) are subsequently carried out. In recent applications [108–123], two types of tITP modified arrangements are mainly used to stack low levels of analytes; (i) injection of a sample solution into the capillary filled with a BGS containing leading electrolytes (LE), and then a suitable terminating electrolytes (TE) injection behind the sample plug, (ii) injection of a sample solution containing LE, *e.g.*, ionic analytes dissolved in a high salt concentration matrix, into the capillary filled with a BGS containing TE.

Recently, coupling of tITP-CZE with MS detection has attracted much attention since ammonium and acetate ions work well as LE and TE, respectively, for tITP [109], which also fulfill the requirement of mandatory volatility for electrospray ionization (ESI) -MS detection.

By employing tITP in the CZE-MS, biological samples including tryptic peptides [109, 110], phosphopeptides [111] and metabolites [112] are successfully analyzed as summarized in Table 4. Busnel *et al.* reported that t-ITP using ammonium and acetate ions as LE and TE, respectively, was integrated with a porous sheathless CZE-ESI-MS to increase the mass loading of the system while maintaining peak efficiency and resolution with sample plugs as long as one-third of the separation capillary (>200 nL) [109]. Peptides concentration limits of detection in the subnanomolar or nanomolar range can be achieved with or without t-ITP, respectively. Resembled approach was applied to the analysis of phosphopeptides, demonstrating that comparable resolution to conventional CZE and unprecedented sensitivities could be achieved by tITP-CZE [111]. In the analysis of metabolite mixture in human urine, the porous sheathless tITP-CZE-ESI-MS approach was also effective, resulting in the LODs ranging from 0.6 to 24 nM, which were at least 5-fold lower than the LODs obtained without tITP [112]. Wang *et al.* employed ammonium acetate as LE in the tITP-CZE-MS analysis of proteins and peptides [110]. Since proteomic samples were pretreated by solid-phase microextraction (SPME) with organic elution, pseudo-tITP effect [124] can also be obtained in the stacking process. Thus, the peptides are primarily stacked by the elution plug, then mobility-ordered between the ammonium acetate LE plug and the organic solvent plug, acting as TE. The LE zone is gradually disrupted as higher mobility protons migrate through the zone and the tITP process is stopped, finally the peptides begin to be separated in the normal CZE. In the report, the feasibility and sensitivity gains were achieved through coupling SPME and tITP with CZE-MS. Such pseudo-tITP approaches were applied to the determination of NSAIDs [118], tricyclic antidepressants [119], uremic toxin indoxyl sulfate in serum [120], and warfarin enantiomers [121].

As a unique application, Saito *et al.* employed a polymer-enhanced tITP-CZE approach to the analysis of gram-positive bacteria [117]. In addition to the ability of polyethyleneoxide (PEO) to stack microorganisms, the combination with tITP allowed to focus the bacteria to a single peak when glycine was added to the sample as LE while a BGS contained PEO and carbonate ions as TE (Fig. 3). By coupling tITP-CZE with amperometric detection (AD),

highly sensitive detection of four doping substances was attained with the LOD of 10^{-14} M level [115]. In the tITP method, the sample zone was injected between Tris-HCl LE and glycine TE zones, respectively, which allowed the determination of human urine samples only with pretreatment by centrifugation to remove precipitated proteins and other particulate substances.

3.2. Electrokinetic supercharging (EKS)

Electrokinetic supercharging (EKS), which is a combination of electrokinetic injection with tITP, is one of the most powerful on-line sample preconcentration techniques to obtain extremely high enrichment factors. EKS is also used in several separation modes including CZE, CGE and NACE as summarized in Table 4 [125–135]. Hirokawa *et al.* demonstrated that it was possible to substantially improve analyte loading and hence the detectability by modifying sample introduction configuration [125]. As shown in Fig. 4, the volume of sample vial was increased, the common wire electrode was replaced by a ring electrode, and the sample solution was stirred. With these alterations, more analyte ions were accumulated within the effective electric field during electrokinetic injection and then maintained as focused zones due to tITP. The EKS-CZE approach was applied to a mixture of seven rare-earth metal ions with an enrichment factor of 500000 giving the LODs at or below 1 ng/L. These LODs were over 100000 times better than those obtained by normal hydrodynamic injection. The modified electrode configuration was applied for greatly loading of DNA fragments that was followed with EKS preconcentration and final CGE separation [129]. An ultralow-viscosity BGS consisting of TBE, 2% low-molecular-weight hydroxypropyl methyl cellulose and 6% mannitol was employed as a sieving media, in which the boric acid complexing with mannitol acted as LE for tITP. The LOD of CGE-UV analysis of DNA is always at ppm level, which could be improved to ppt level by the modified EKS-CGE method.

As another sensitivity enhancement approach in EKS, EKS with a positive pressure (pressure-assisted EKS, or PA-EKS) during injection to improve stacking of NSAIDs was reported by Meighan *et al* [133]. Balancing the EOF which was in the direction of the inlet, with a hydrodynamic pressure which was in the direction of the outlet, allowed long

electrokinetic injection. When the LE was injected hydrodynamically and the potential was switched, the injected analytes were restacked and separated by CZE. By optimization of several parameters, 50000-fold sensitivity enhancement was obtained for seven NSAIDs. Tian *et al.* developed an incorporated approach of the sweeping by dendritic polyamidoamine and EKS for on-line preconcentration of DNA fragments [135]. During the run, a plug of polyamidoamine was hydrodynamically injected first, followed by FASI of DNA fragments, which were concurrently swept by polyamidoamine via complexation with DNA. Then, a segment of releasing agent, SDS, was hydrodynamically introduced and subsequently electrophoretically driven to interact with the DNA-polyamidoamine complexes, forming more stable supramolecular SDS-polyamidoamine complexes and releasing the initially bound DNA fragments. The excess dodecyl sulfate anion also acted as TE in the separation, thereby the DNA fragments were enriched by the joint effects of FASI, sweeping and tITP. Due to the extremely low mobility of the dendriplexes, the polyamidoamine sweeping-EKS strategy allowed long FASI time, providing enrichment factors more than 3500-folds relative to the conventional hydrodynamic injection.

4. pH-mediated stacking

By manipulating electrophoretic mobilities with a pH change between sample and BGS zones, weakly acidic and basic analytes can be stacked, which is generally referred to dynamic pH junction [136–138], pH-mediated stacking [139], neutralization reaction boundary (NRB), moving reaction boundary (MRB) [140, 141], and transient moving chemical reaction boundary (tMCRB) [142, 143]. In the recent reports, a wide range of applications, *e.g.*, amino acids, peptides, proteins, hormones, food samples, forensic, and pharmaceuticals, have appeared as summarized in Table 5 [144–164]. Hasan *et al.* reported a combination of the dynamic pH junction with CZE-MS for analyzing the intracellular angiotensin II within the peptides extracted from HL1 cells [147]. It was demonstrated that the preparation of the sample in a water-miscible organic solvent yielded better stacking than the use of aqueous buffers. When borate buffer with 50% v/v acetonitrile was used as the sample matrix,

approximately 4000~11000-fold sensitivity enhancement was achieved. Bachmann *et al.* demonstrated an off-line hyphenation of pH-mediated stacking-CZE with subsequent deposition of the separated analytes onto a target for MALDI-TOF-MS in the analysis of low-abundant tryptic phosphorylated peptides from phosphoproteins [149]. The on-line preconcentration method allowed loading of a large sample volume (~150 nL), which was introduced into the capillary after the hydrodynamic injection of a short plug of ammonium hydroxide and was sandwiched between two plugs of the acidic BGS, giving the separation and identification of multiphosphorylated peptides at femtomole level. Kamentsev *et al.* reported a unique application of the pH-mediated stacking to the analysis of metal ions [150]. For the on-line preconcentration of metal ions, shifting the equilibrium between the metal aquation and its anionic complex with a chromophoric reagent, Xylenol Orange, on the pH-boundary was utilized. In the analysis of zinc(II) and cadmium(II) ions, high efficiencies of about $1\sim2 \times 10^6$ theoretical plates was achieved even for the sample matrix containing high-concentration of salt up to 0.1 M, giving significantly better sensitivity than conventional CE procedures with direct and indirect photometric detection in the UV region. Anouti *et al.* reported on-line preconcentration of amino acid enantiomers by tMCRB using ammonium formate as a sample matrix and acetic acid as a BGS in heart-cutting 2D-CE with C⁴D detection [161]. This on-line tMCRB preconcentration was applied to the chiral separation of D,L-phenylalanine and D,L-threonine in a mixture of 22 native amino acids. When a volume corresponding to 10% of the total capillary volume was injected, LOD of 2 μ M was determined for L-threonine.

In the recent years, several theoretical and simulation studies on the pH-mediated stacking techniques have appeared [156–158, 163, 165–167]. Cao's group reported theoretical and comparative studies on the stacking of zwitterions on the pH boundary [165]. The theoretical results brought several important findings, *e.g.*, (1) a much long front plug of a strong base is needed for the complete pH-mediated stacking of zwitterions, if high-concentration of alkali is contained in a sample matrix stacking; (2) the existence of salts in a sample matrix holds almost no obvious effect on the stacking of zwitterions if the pH-boundary is prepared with a

weak alkaline buffer, but a dramatic effect on the stacking is present if with a strong base. The theoretical predictions were quantitatively confirmed by the experiments employing amino acids as model analytes [166]. Yeung and co-workers reported a computer simulation study on the mechanism of the pH-boundary formation and the electrophoretic migration of various ions [167] and the effect of the BGS concentration on the protein enrichment [158]. The obtained results were well employed to the removal of unwanted buffering electrolytes from the concentrated sample. The key was to use either the acid or the base of the discontinuous buffers to adjust the pH of the sample, such that the net charge of the unwanted buffering ions was near-zero. The successful isolation and enrichment of myoglobin from TRIS, MES and phosphate was demonstrated with the enrichment factors from 130-470. Since buffer ions present in the samples can significantly alter the local pH of the discontinuous buffers and thus potentially disrupt protein enrichment, the isolation of buffering ions from the enriched zones should be an important step towards the handling of biological samples.

Quirino and Breadmore investigated the pH-mediated acid or base stacking technique experimentally and with computer simulation [156, 157]. In the acid stacking, a plug of acid is injected after the sample of basic analytes. In the proposed model, protons from the acid plug penetrated the alkaline sample zone, causing the formation of a dynamic pH junction that swept through the sample zone. The neutralization of the protons in the sample zone also caused tITP stacking of basic or cationic analytes with ammonium and hydronium ions as LE and TE, respectively. Computer simulation clearly showed that the stacking was an ITP mechanism. The acid stacking strategy afforded an order of magnitude sensitivity improvement for model tricyclic antidepressant and β -blocker drugs [157]. In the base stacking technique, analytes were stacked at the junction where the mechanism of focusing was tITP with the acetate and hydroxide ions as LE and TE, respectively. The base stacking strategy was tested using hypolipidemic, nonsteroidal anti-inflammatory, and diuretic drugs, and afforded 19~37 improvements in the peak height [156]. The authors concluded that an advantage of the acid or base stacking technique compared with conventional tITP is the

simplification of the selection of TE.

Ševčík and co-workers studied on the effect of the electrokinetic injection at the pH boundary formed by borate and phosphate electrolytes on the on-line preconcentration of weak acids. They verified that the differences in mobilities of adjoining co-ions, borate and phosphate, created the tITP state and the heterogeneity in the electric field strength across the capillary caused FASI [163]. This technique was named on-line electrokinetic accumulation on the pH boundary/mobilization preconcentration. In this technique, the addition of organic solvent to the injection electrolyte could enhance the preconcentration performance. Subnanomolar LODs of hydroxybenzoic acids were achieved with 80% MeOH in the injection electrolyte which represented approximately 70000-fold preconcentration. This technique was applied to the preconcentration of ketoprofen enantiomers followed by the enantioselective separation by CD-modified MEKC (CDMEKC) [164]. As demonstrated above, the significance of the contribution of the tITP state and the addition of organic solvent was proved by the recent pH-mediated stacking studies.

5. Sweeping and related techniques

5.1. Sweeping

Sweeping is one of the most popular technique, which utilizes the migration velocity changes of analytes due to the interaction with pseudostationary phase (PSP) in electrokinetic chromatography (EKC). In the conventional sweeping, SDS is employed as the PSP to enrich neutral and/or hydrophobic analytes since a higher retention factor to the SDS micelle provides an efficient preconcentration. In the recent three years, the SDS-sweeping technique is applied to the MEKC, MEEKC or CD-modified MEKC (CDMEKC) analysis of steroids [168], hydrophobic pharmaceuticals [169], tricyclic antidepressant, β -blockers [170], enantiomers [169, 171], and nanocrystals [172] as summarized in Table 6 [168–177]. Xia *et al.* combined an open tubular capillary microextraction with sweeping for the analysis of hydrophobic compounds [169]. In the developed method, a sample solution was fully

injected into a stationary phase-coated capillary for the extraction. Followed by an eluent injection, a plug of microemulsion solution was aspirated into the capillary from the outlet. By applying a high voltage, the sweeping was carried out, providing a high enrichment factor of 1355 for a hydrophobic pharmaceutical in aqueous solutions. In the recent works, the application of EDTA [173] and latex nanoparticles [174] as the PSP have appeared for the sweeping of metal ions and hydrophobic compounds, respectively. Palmer *et al.* developed novel latex nanoparticles as the PSP for the sweeping-EKC-UV and MS analysis [174]. The nanoparticles with a hydrophobic core, and an ionic shell had a minor effect on the noise with UV detection and minor effects on analyte ionization efficiency during ESI. The nanoparticles did not cause fouling or degradation of the ESI-MS interface. The combination of sweeping and selective MS yielded LODs of 10-16 ppb for hydrophobic dialkyl phthalates. Such a development of new PSP for EKC should be useful for the sweeping of a variety of analytes before MS detection.

5.2. tr-trapping, AFMC and MSS

Sueyoshi *et al.* proposed a novel preconcentration mechanism, transient trapping (tr-trapping), which was the combination of a partial filling (PF) with the sweeping, for a high-speed separation of analytes with very-high retention factors in a short separation length [178]. In the tr-trapping, a short plug of the SDS micelle was partially injected into the separation column filled with a BGS containing no SDS. Immediately after injecting the micellar solution, a sample solution void of SDS was injected as a long plug. The hydrophobic analytes were initially swept and then trapped for a short time by the SDS micelles around the sample/micellar boundary. The concentration of the micelle at the boundary was decreased during electrophoresis, causing trapped analytes to be released from the boundary and then separated by difference in the timing of the release, which called “trap and release mechanism”. The tr-trapping technique was also effective for the enrichment and separation of enantiomers by employing both SDS and CDs as the PSP [179]. Breadmore *et al.* investigated the mechanism of transient trapping by the use of computer simulations and compare it to sweeping in MEKC [180]. The simulation results confirmed the trap and

release mechanism originally proposed. Importantly, tr-trapping can achieve comparable resolution to the conventional sweeping in about 1/10 of the time and 1/4 of the separation length, while also providing a modest increase in sensitivity. They concluded that the tr-trapping is superior than sweeping in configurations with a short separation column, that is, sweeping is better for conventional CE arrangements with long capillaries.

Micellar PSPs were also employed in the preconcentration techniques of analyte focusing by micelle collapse (AFMC) [181] and micelle to solvent stacking (MSS) [182], which have distinctions of being used for neutral and charged analytes, respectively. In AFMC and MSS, a sample containing micelles was introduced into the capillary filled with an organic solvent-modified BGS devoid of micelles. Application of voltage caused the enrichment due to the focusing of analytes effectively incorporating in the micelle around the high-conductivity BGS/low-conductivity sample zone. After the enrichment, the neutral analytes were released from the boundary due to micelle collapse in AFMC. In MSS, the direction of the charged analytes' effective electrophoretic mobilities was reversed due to the decrease in the retention factor between the analytes and oppositely charged micelles, leading the focusing of ionic analytes and the separation by CZE or NACE (Fig. 5). These AFMC and MSS techniques have been applied to drug components [183–187], herbicides [183, 186, 188], and alkaloids [189–191]. As a typical result, the use of SDS micelle collapse using a methanol–water BGS for MSS provided 300 and 1036-fold sensitivity enhancements for cationic ephedrine and berberine, respectively [191]. In MSS, anionic SDS micelle is effective for concentrating cationic analytes. Cationic surfactants including cetyltrimethylammonium bromide (CTAB) [192] and ionic liquid (IL) type surfactant [188] were also introduced for the MSS concentration of anions. Two-step stacking of cations [193] and anions [194] by sweeping and MSS was also investigated. In the two-step stacking, a micellar solution is injected before the sample that is prepared without the micelles. The charged micelles sweep and transport the oppositely charged analytes to the boundary zone between the sample and BGS. The presence of organic solvent in the BGS induces the second stacking step of MSS. A 17-46 times sensitivity increase compared to typical

injection was obtained for beta blockers, two tricyclic antidepressant drugs, hypolipidaemic drugs, non-steroidal anti-inflammatory drugs, and herbicides. Recently, a similar experimental configuration technique, which was named transient micellar phase extractor, using CTAB was studied for the enrichment of anions [183]. In the modified method, the micellar solution was partially injected in front of a sample solution prepared in a matrix with organic solvent. Anionic analytes and CTAB micelles interacted where the anions were extracted by the micelles due to hydrophobic and electrostatic interactions. The micelle-bound anions were then transported to the boundary that contained a solvent where analyte focusing by MSS occurred. In the present stage, the SEFs was only 13-28 for drug components and 30 for herbicides. However, the application of MSS to CE-MS is expected because the preconcentration technique is based on a partially injected micellar phase [195].

5.3. Coupling with FASI and other hyphenated techniques

As well as other stacking techniques, a combination of sweeping and related techniques with the electrokinetic injection of a large volume sample is significantly effective for obtaining higher enrichment factors [196–216]. Cation or anion selective exhaustive injection (CSEI or ASEI)-sweeping technique is a combination of FASI and sweeping, which can provide more than 100000-fold increases in detection sensitivity [196, 197]. Recently, Jin *et al.* reported that ASEI and CSEI were employed to sweeping-MEKC for the determination of melamine and its derivatives [198]. Compared to LVSS, the detection limits in CSEI were improved around 8000-9200 fold for melamine and ammeline. Wei *et al.* combined a successive CSEI-ASEI with EOF suppressants and sweeping to analyze both acidic and basic components in a single run [199]. The developed technique was successfully applied to the quantification of acidic and basic drugs in commercial tablets and in plasma samples. Zhang *et al.* developed a modified sweeping technique under a controlled EOF [200]. The EOF was used to balance the electrophoretic migration of SDS micelle to generate an immobile sweeping boundary. Sample solution was continually injected into capillary and sweeping by SDS micelles, *e.g.*, injected plug length of 51-fold the effective length for 70 min, resulted in 21000 and 23000-fold increase in the sensitivity for phlorizin

and quercitrin, respectively. Coupling of FASI with MSS was also effective for enhancing the enrichment factors of alkaloids [201], antipsychotic drugs [202], and cationic drug components [203], typically to several hundreds to thousands.

Chemometric approach and simulations were employed to elucidate the impacts of parameters influencing CSEI-sweeping [204, 205]. In the case of using phosphate buffer as the BGS, higher sensitivity enhancements can be obtained to perform injection under constant current rather than constant voltage in CSEI because a lower analyte amount is injected due to the decrease in the current intensity during the electrokinetic injection under constant voltage [205].

Since the electrokinetic injection of ionic analytes differs the injected sample composition from original one, there are cases when the hydrodynamic injection of a large sample volume is a better selection in the on-line sample preconcentration in CE. In the recent years, a few reports on coupling of LVSS with sweeping have appeared for determining angiotensins [206], vitamins [207], p-hydroxybenzoic acid esters [208] and carbamazepine metabolites [209] with up to 300-fold sensitivity enhancement. Wang *et al.* reported a unique stacking method of repetitive large volume sample injection (rLVSI) and sweeping-MEKC for determination of androgenic steroids in urine [210]. In the developed method, a sample solution devoid of micelles was injected at 10 psi for 20 s, and then stacked at -10 kV for 1 min around SDS-sweeping boundary. These injecting and stacking steps were repeated several times. The sensitivity enhancements for steroids were about 2500, which was higher than that in the electrokinetic injection mode.

Sequential preconcentration technique based on the dynamic pH junction and sweeping was applied to the analysis of methotrexate metabolites in cerebrospinal fluid [211] and dipeptides in human serum [212]. When the sample has a different buffer pH and is devoid of micelles relative to the BGS, the preconcentration by dynamic pH junction-sweeping is expected. In the MEKC-LIF analysis, this hyphenated focusing provided a highly sensitive detection of four dipeptides with the LOD ranging from 1.0 to 5.0 pmol/L [212]. Zhang *et al.* reported a sequential preconcentration/separation approach involving on-line combination of

the modified sweeping under a controlled EOF [200] and AFMC with heart-cutting 2D-CE in a single capillary for the analysis of Herba Leonuri and mouse blood samples [213]. After injecting a large amount of analytes by the modified sweeping, the preconcentration scheme was incorporated into the heart-cutting 2D-MEKC/CZE separation. To release analytes from the SDS micelle and to counteract the primarily preconcentrated and separated analyte band diffusion during the hydrodynamic mobilization process, AFMC method was utilized between the two dimensions. The dual concentration methods resulted in the increased detection factors of 6000-fold relative to the traditional pressure injection method.

Sequential preconcentration/multi-dimensional separation will become an attractive method for on-line sample concentration and separation of complex samples. Among several sequential preconcentration techniques, a coupling of field-amplified stacking or tITP with the electrokinetic injection is considerably effective for extremely higher sensitivity, *e.g.*, SEFs of 110000 for LVSEP-PAEKI [90] and 500000 for EKS [125]. However, these FASI-based techniques can enrich only low-ionic strength samples despite especially ITP is well known as a useful technique for concentrating highly saline sample. Furthermore, the electrokinetic injection causes the changes in the sample composition in the capillary because the injection amounts of analytes are proportional to their electrophoretic mobility. Hence, alternative approaches without the electrokinetic injection are still desired. For example, in-line coupling of microextraction techniques such as single drop microextraction and on-capillary SPE with FASS/LVSEP-NACE are quite effective for cleanup of crude matrices, desalting highly saline sample for the field-amplified stacking, and subsequent concentrating minor components since enriched extracts eluted from a SPE cartridge with organic solvents or from a liquid-liquid extraction can be injected directly into the FASS/LVSEP-NACE system. Although several applications on sequential coupling of the microextraction with field-amplified stacking techniques are found in recent years, in-line or on-line combination with other sample preconcentration including tITP, pH-mediated stacking and sweeping techniques are still limited. Since tITP, pH-mediated stacking and sweeping are useful for concentrating saline samples, amphoteric and hydrophobic compounds, respectively, further

developments on these integrated sample preconcentration systems is expected in CE.

6. Conclusion

In this article, recent applications of the on-line sample preconcentration techniques including field-amplified stacking, transient isotachopheresis, pH-mediated stacking and sweeping, and their modified and combined techniques are reviewed. Many applications have appeared in the fields of biological, environmental, food, toxicological, forensic and nano-sciences. It should be noted that, in recent years, the preconcentration technique was applied to the CE separation of nanoparticles, such as PV particles, GNPs, CdSe nanocrystals and gram-positive bacteria, to improve not only sensitivity but theoretical plates, resolution, size-separation performance and reproducibility. This demonstrated that the stacking effect minimized the band broadening of the nanoparticles due to the heterogeneity of size, shape and surface charges, resulting in the reproducible data.

Especially for biological applications, the importance of MS detection will be growing continuously, so that considerable developments for combining on-line preconcentration and CE separation with MS detection are desired. In the recent works, a combination of tITP or dynamic pH junction with CZE-MS has attracted much attention since electrolytes used in these techniques fulfill the requirement of volatility for ESI. Novel preconcentration techniques have been also developed in the last three years, *i.e.*, coupling of electrochemical preconcentration based on stripping voltammetry with FASS-CZE, electrokinetic stacking at an etched porous junction, transient trapping, analyte focusing by micelle collapse, micelle to solvent stacking, and so on. Although there are several issues in these approaches, such as a long preconcentration time, poor enrichment efficiency and repeatability, they can be essentially compatible with MS detection. Therefore, the application of these new stacking methods and sophisticated-tITP to CE-MS will be expected for the analysis of trace amount of biological samples.

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Figure Legends

Fig. 1. EPC–CE interface. (A) Cross section cut through the capillary and the working electrode, with flow channel perpendicular to the plane; (B) lateral schematic view of the upper and lower blocks of the interface, tightly screwed together (screws not shown). Relevant parts: a, acrylic body; b, bushing (Tygon capillary tube); c, polyimide-coated fused-silica capillary; d, glassy carbon electrode; e, spacing gasket; f, Kel-F block; g, Ag wire (pseudoreference electrode); h, auxiliary electrode and CE ground electrode (switchable between the potentiostat and the HV supply); i, flow inlet; j, flow outlet. (C) Evaluation of the stacking effect in EPC–CE–C⁴D of metal ion mixtures. Sample, 1.0 mg L⁻¹ Tl⁺, Pb²⁺ and Cd²⁺ and 0.5 mg L⁻¹ Cu²⁺ and Zn²⁺ stripped in (a) 30 mmol L⁻¹ MES + 30 mmol L⁻¹ HIS and 5 mmol L⁻¹ HIBA) and (b) 6 mmol L⁻¹ MES + 6 mmol L⁻¹ HIS. [Reprinted with permission from *Electrochem. Commun.* 12 (2010) 1387. Copyright 2010 Elsevier B.V.]

Fig. 2. (a) Schematic of the porous junction preparation. Insert indicates the process of HF etching of the naked capillary outside wall. The HF etchant (red) should be cleaned immediately after the ion conduction of the interface as indicated by an abrupt increase of the current or by the sound alarm. (b) Stacking time vs peak intensity in ES-CZE-LIF of fluorescein: Probe solution, 1 pM fluorescein in 1 mM pH 9.2 Tris-HCl; stacking voltage, 50 V; CZE, 1000 V; diode laser, 487 nm; PMT, 700 V. The dotted reference lines refer to the respective signal levels of standard probe solutions. [Reprinted with permission from *Anal. Chem.* 84 (2012) 7085. Copyright 2012 American Chemical Society.]

Fig. 3. Polymer-enhanced tITP of *Bacillus (B.) globigii* and *Saccharomyces (S.) cerevisiae* spores. Separation buffer consists of 10 mM carbonate (pH 10.0) with 0.0125% PEO and 1.0 μM labeling probe with (a) 0 mM, (b) 10 mM, and (c) 1 mM added borate. Sample consists of 3.4×10^7 cfu/mL *B.globigii* and 200 μg/mL *S.cerevisiae* prepared in 3.2–50 mM Tris-glycine

buffer (pH 8.9) with 0.0125% PEO. [Reprinted with permission from Anal. Chem. 84 (2012) 2452. Copyright 2012 American Chemical Society.]

Fig. 4. Schematic of sequential alterations of the injection setup implemented to improve the sensitivity of EKS-CE: a wire electrode in (a) a standard and (b) a large-volume sample vial; a ring electrode in a large-volume sample vial (c) without and (d) with stirring. The red zone depicts the actual part of the sample solution subjected to injection (not to scale). C and E stand for capillary and electrode, respectively. (e) Electropherograms of (upper trace) blank and (lower trace) 25 pM sample analyzed at the optimized EKS conditions as shown in (d). Sample: 100 000-fold diluted at 25 pM. Injection: 20 kV for 300 s. Separation voltage: 20 kV. [Reprinted with permission from Anal. Chem. 83 (2011) 398. Copyright 2011 American Chemical Society.]

Fig. 5. Schematics of MSS of cations in CZE. (a) Injection of sample solution (S) (gray area) prepared in a micellar matrix into the capillary filled with BGS that is modified by an organic solvent. The MSSB is the micelle to solvent stacking boundary. (b) Transport of micelle-bound analytes to the MSSB by the anodically (electrophoretic) moving micelles. At the MSSB, the effective electrophoretic mobility of the cations changes from $\mu_{\text{eff}}(\text{S})$ in the S zone to $\mu_{\text{eff}}(\text{MSSB})$ at the MSSB zone. The reversal in direction of the effective electrophoretic mobility causes the focusing of transported cations at the MSSB zone (dark zone). (c) Continued transport of analytes, causing all micelle-bound cations to be focused at the MSSB zone. All the micelles from the S traverse the MSSB zone. The mobility of the cations changes from $\mu_{\text{eff}}(\text{MSSB})$ to $\mu(\text{BGS})$, which is dependent only on the electrophoretic mobility of the cations (no micelle interaction). (d) The cations are separate by virtue of CZE. [Reprinted with permission from J. Chromatogr. A 1216 (2009) 294. Copyright 2009 Elsevier B.V.]

Figure1

Figure2

Figure3

Figure4

Figure5

Table 1. Applications of field-amplified stacking in CZE, CGE and ACE.^a

Target analytes	Sample matrix	Preconcentration and separation modes, and detection scheme	LOD	Enrichment factor	Application	Comments	Ref. #
Endogenous compounds (homovanillic acid, vanillylmandelic acid, indoxyl sulfate, tryptophan, ascorbic acid, uric acid)	Urine	FASS-CZE-EC	75 nM	4	Pharmaceuticals	GNPs coated capillary	23
Aminobutyric acid, alanine	Tea	FASS-CZE-LIF	0.7-0.8 nM		Food analysis		24
Catecholamines (dopamine, epinephrine, and norepinephrine)	Rat brain	FASS-CZE-LIF	22-34 pg/L	118-132	Biological applications		25
Danshensu, salvianolic acid B, caffeic acid and lithospermic acid	Danshen sample	FASS-CZE-MS		30-50	Pharmaceuticals		26
Nine (fluoro)quinolone antibiotics	Chicken	FASS-CZE-UV	2.4-34 ng/g	5-252	Food analysis	Combined with monolith-extraction	27
Alkylpyrazines	Leaf-cutting ants' venom glands	FASS-CZE-UV	50 ng/venom glands		Biological applications		28
Benzimidazole drugs	Animal tissue samples	FASS-CZE-UV	1.05–12.61 ng/g	15.6-57.0	Food analysis	Combined with magnetic solid-phase extraction	29
Doripenem and related substances	Medicinal product	FASS-CZE-UV	3.0 µg/mL		Pharmaceuticals		30
Ephedrine, pseudoephedrine	Urine	FASS-CZE-UV	0.21-0.23 µg/mL	50	Pharmaceuticals	Coupled with cross-H-channel interface for flow injection	31
Ionic neurochemicals	Urine	FASS-CZE-UV	14.7-313.4 nM	116-281	Biological applications	PEO-based stacking	32
NSAIDs (etodolac, naproxen, ketoprofen, flurbiprofen and diclofenac)	Milk, dairy products	FASS-CZE-UV	3.0-13.1 µg/kg	46-229	Pharmaceuticals	Combined with DLLME	33
Phenazine-1-carboxylic acid	Soil sample	FASS-CZE-UV	0.02 mg/kg		Environmental		34
Posaconazole	Plasma	FASS-CZE-UV	10 ng/mL		Pharmaceuticals		35
Pyoluteorin	Soil sample	FASS-CZE-UV	0.107-0.36 µg/mL		Environmental		36
Se(IV), Se(VI) ions	River water	FASS-CZE-UV	57-71 ng/L		Environmental	Combined with SPE	37
Sertraline	Urine	FASS-CZE-UV	2.5 nM	814	Pharmaceuticals	Combined with DLLME	38
β-Blockers (atenolol, esmolol, and bisoprolol)	Urine, serum	FASS-CZE-UV	2.2-3.5 nM	26-38	Pharmaceuticals		39
(±)-Threo-methylphenidate	Plasma	FASS-CDCZE-UV	600 pg/mL		Pharmaceuticals		40
MicroRNAs	Serum	FASS-ProFACE-LIF	0.5 fM (30000 molecules/mL)	1000	Biological applications	Coupled with SPE using RNA binding protein-coated magnetic beads	41
Arachidonic acid metabolites	Cell extracts	HC-FASS-CDMEKC-	30-50 ng/mL	80-350	Biological		42

Cocaine, benzoylecgonine, ecgonine methyl ester, norcocaine, cocaethylene	Urine	UV FASI-CZE-MS	1.5-10 ng/mL	5-10	applications Forensic		45
Alkaloids (cytisine, sophocarpine, matrine, sophoridine and oxymatrine)	Medicinal plants and <i>Sophora</i> instant granule	FASI-CZE-UV	1.3-4 ng/mL		Pharmaceuticals	Combined with subcritical water extraction	46
Brain-gut peptides	Human cerebrospinal fluids	FASI-CZE-UV	0.02-0.06µM	45-102	Biological applications		47
β ₂ -agonists (cimbuterol, clenbuterol, mabuterol, and mapenterol)	Urine	FASI-CZE-UV	1.8-37 µg/L	41-1046	Pharmaceuticals	Combined with DLLME	48
3,4-Methylenedioxymethamphetamine, lysergic acid diethylamide, and phencyclidine	Urine	FASI-CZE-UV	1.0-4.5 ng/mL		Pharmaceuticals	Combined with DLLME	49
Arginine and dimethylated arginines	Plasma	FASI-CZE-UV		10-20 nM	Biological applications		50
Aripiprazole and its active metabolite (dehydroaripiprazole)	Plasma	FASI-CZE-UV	2.0-2.5 ng/mL		Pharmaceuticals		51
Aromatic amines (aniline, o-tolidine, o-toluidine, and m-aminophenol)	River water	FASI-CZE-UV	0.29-0.44 ng/mL		Environmental		52
Cr(III), Pb(II), Hg(II), Ni(II) ions	Tap, drain, and reservoir waters	FASI-CZE-UV	0.005-2.32 µg/L	91800	Environmental		53
Ephedrine, norephedrine, pseudoephedrine	Urine	FASI-CZE-UV	1.0-2.8 ng/mL	1750	Pharmaceuticals	Combined with USAEME	54
Imatinib mesylate and related compounds	Synthetic samples	FASI-CZE-UV	1.5-5.6 ng/mL		Pharmaceuticals		55
Inorganic anions	Post-blast residues, detonator extracts, soil	FASI-CZE-UV	0.31-0.64 mg/L	15-23	Forensic		56
Malondialdehyde	Plasma	FASI-CZE-UV	3 nM	100	Biological applications		57
Meurotransmitters (dopamine, 3-methoxytyramine and serotonin)	Urine	FASI-CZE-UV	5.9-10 nM	500	Biological applications	Combined with MIP-SPE	58
Selenomethionine, selenomethionine selenoxide		FASI-CZE-UV	0.2-0.5 µM	800	Biological applications		59
Three aconitine alkaloids	<i>Aconitum</i> plants	FASI-CZE-UV	21-560 ng/mL	5-142	Toxicological		60
Toxic alkaloids (strychnine, brucine)	Urine	FASI-CZE-UV	2.0-2.5 ng/mL	350-550	Biological applications		61
β-Blockers (pindolol, oxprenolol, propranolol enantiomers)	Urine	FASI-CDCZE-UV	0.10-0.65 nM	62000-200000	Pharmaceuticals		62
DNA fragments (PCR products from pathogenic bacteria)		FASI-CGE-UV	2.2-5.8 ng/mL	25-52	Biological applications	Ion pair reagent-assisted CGE	63
Proteins (cytochrome c, lysozyme,		FASI-CZE-UV/MS	5-10 nM	1344-4869	Biological		64

ribonuclease A, S-ribonuclease, lactalbumin, lactoglobulin A and B, lactoferrin, bovine serum albumin, aldolase and myoglobin)					applications	
Arginine and methylated metabolites	Human plasma	FASI-CZE-MS	10-20 nM	25	Biological applications	65
Sorbic and benzoic acids	Soy sauce	FASI-CZE-C ⁴ D	0.05-0.08 µM		Food analysis	66
Neurotransmitters (tyramine, serotonin, octopamine, dopamine)	<i>Drosophila</i> brains	FASI-CZE-FSCV	1-4 nM		Biological applications	67
Imatinib mesylate and related substances	Synthetic samples	FASI-NACE-UV	9.13-22.5 ng/mL		Pharmaceuticals	68
Perfluorooctanoic acid and perfluorooctanesulfonic acid	River water	FASI-NACE-UV	31-40 nM	624-806	Environmental	69
As(III), As(V), Se(IV), Se(VI) and bromate ions	Drinking water	PAEKI-CZE-MS	1-3 ng/mL		Environmental	71
Haloacetic acids (Monobromoacetic, dibromoacetic, tribromoacetic, monochloroacetic, dichloroacetic, trichloroacetic, bromochloroacetic, moniodoacetic, chlorodibromoacetic, bromodichloroacetic, and carbon-13 labelled monochloroacetic acids)	Drinking and tap water	PAEKI-CZE-MS	0.013–0.12 µg/L	20000	Environmental	72
Perchlorate	Water and soil samples	PAEKI-CZE-MS	18.7 ng/L (water), 3.3 ng/g (soil)		Environmental	73
Phenylarsenic compounds (phenylarsonic, 4-aminophenylarsonic, 4-hydroxyphenylarsonic, 4-nitrophenylarsonic and 3-nitro-4-hydroxyphenylarsonic acids)	Pig feed, pig litter, soil and lake water	ASEI-CZE-UV	0.68-6.90 µg/L	155-1780	Environmental	70
Matrine, andoxymatrine, 5-sulfosalicylic acid	Extractant from <i>Sophora flavescens</i> Ait	two-end FASI-CZE-UV	0.06-0.2 ng/mL	1003-1380	Food analysis	74
Six parabens (methylparaben, ethylparaben, propylparaben, isopropylparaben, butylparaben, and benzylparaben)		LVSS with PS /FASI-CZE-UV	18–27 (sweep), 3–4 (LVSS), 2 (FASI) ng/mL,	29 (sweep), 77 (LVSS), 120 (FASI)	Food analysis	Compared with sweep-MEKC-UV 75
Barbiturates (secobarbital, amobarbital, barbital, and phenobarbital)	Urine, whole blood	LVSS with PS-CZE-UV	15-57 ng/mL	169-203	Forensic	76
Flavonoids (kaempferol and quercetin)	Broccoli extract	LVSS with PS-CZE-UV	0.6-0.9 mg/kg		Food analysis	77
Eight polyphenolic compounds (carnosic acid, cinnamic acid, caffeic acid, rosmarinic acid, quercetin, apigenin, luteolin, rutin)	Medicinal plant (<i>Orthosiphon stamineus</i> Benth.)	LVSS with PS-CZE-UV	9-16 ng/mL	90	Pharmaceuticals	78
Mercury, methylmercury, ethylmercury	Fish tissue,	LVSS with	42-370 pg/mL	2250-12138	Toxicological	Combined with 79

and phenylmercury ions	human hair, lake water	PS-CZE-UV				PT-LLLME	
Sulfonylurea herbicides	Environmental water	LVSS with PS -CZE-UV	0.04-0.12 µg/L (water sample), 0.97-8.30 µg/kg (grape sample)		Environmental		80
Cephalosporins (cephalexin, cephoperazone, ceftiofur, cephapirin and cephazolin)	Spring, ground and river water	LVSS with PS-CZE-UV	0.1-0.3 µg/L		Environmental		81
Alkyl alkylphosphonic acids (ethyl methylphosphonic acid, pinacolyl methylphosphonate and methylphosphonic acid)	River and lake water	LVSS with PS-CZE-indirect UV	1.2-3.1 µg/L	742-1583	Environmental	Combined with Zirconia coated stir bar sorptive extraction	82
Herbicides (glyphosate, glufosinate, aminophosphonic acid)	Drinking water	LVSS without PS /FASI-CZE-C ⁴ D	0.5-20 nM	48-53 (LVSS), 245-1002 (FASI)	Environmental		83
Three chlorophenols	Red wine	LVSEP-CZE-UV	0.3-0.9 nM	3000-3400	Food analysis	Combined with HS-SDME	84
Glucose ladder, glycoprotein-derived oligosaccharides		LVSEP-CZE-LIF	2 pM	400-770	Biological applications		85
Cationic amines, enantiomers, proteins		LVSEP-CZE/CDCZE- UV		100-550	Biological applications		86
warfarin, ibuprofen, abscisic acid, Arg, Met, Leu enantiomers	Urine	LVSEP-CDCZE/CDE KC/CDMEKC-LIF/U V	3.7-4.7 ppb	500-1300	Pharmaceuticals		87
Glycoprotein-derived oligosaccharides		LVSEP-PFACE-LIF	0.3 nM	900	Biological applications		88
Fenoprofen and amino acid enantiomers	River water	LVSEP-ASEI-CZE-U V	0.38-2.1 ng/mL	1000	Environmental		89
Fluorescein, oligosaccharides		PAEKI-LVSEP-CZE- LIF		110000 (fluorescein), 2800-10000 (oligosacchar ides)	Biological applications		90
Herbicides (2,4-dichlorophenoxy acetic acid, 2,4-dichlorobenzoic acid, 4-amino-3,5,6-trichloropicolinic acid, 3,5-dichlorobenzoic acid, 2-(2,4,5-trichlorophenoxy) propionic acid, 2-(2,4-dichlorophenoxy) propionic acid)	River water	FAEP-NACE-UV	0.08-0.14 ng/mL		Environmental	Combined with SBME	91
sinapaldehyde, syringaldehyde, coniferaldehyde, vanillin	Whiskey	SWMR-CZE-UV	22-73 mg/L		Food analysis		92
Sixteen phenolic compounds	Wine	REPSM-CZE-UV	0.06-0.41 mg/L	20	Food analysis		43
Organic acids (linolenic acid, lauric acid, <i>p</i> -coumaric acid, ascorbic acid, benzoic	Chinese medicine	REPSM-CZE-UV	0.4-60 ng/mL	16-73	Pharmaceuticals		44

acid, caffeic acid, succinic acid and samples
fumaric acid)

^a GNPs, gold nanoparticles; NSAIDs, non-steroidal anti-inflammatory drugs; DLLME, dispersive liquid–liquid microextraction; ProFACE, protein facilitated affinity capillary electrophoresis; HC-FASS, head-column FASS; USAEME, ultrasound-assisted emulsification microextraction; C⁴D, capacitively coupled contactless conductivity detection; FSCV, fast-scan cyclic voltammetry; PAEKI, pressure-assisted electrokinetic injection; PS, polarity switching; PT-LLLME, phase transfer based liquid–liquid–liquid microextraction; HS-SDME, headspace single-drop microextraction; PFACE, partial filling affinity capillary electrophoresis; FAEP, field-amplified sample injection and pre-introduced organic solvent plug removal with the EOF as a pump; SBME, solvent-bar microextraction; SWMR, sample stacking with matrix removal; REPSM, reversed electrode polarity stacking mode.

Table 2. Applications of field-amplified stacking in MEKC.^a

Target analytes	Sample matrix	Preconcentration and separation modes, and detection scheme	LOD	Enrichment factor	Application	Comments	Ref. #
Poliovirus particles	Cell extracts	FASS-MEKC-UV	19.8 µg/mL		Toxicological		94
2',3'-Dideoxyadenosine-5'-triphosphate (ddATP)	Peripheral blood mononuclear cells	LVSS without PS-CZE-UV/MEKC-Flu	10 fmol/sample	160	Biological applications		95
Adenine	Green tea	LVSS without PS-MEKC-UV	2 ng/mL	1030	Food analysis	Combined with SD-LLLME	96
Chlorophenols (2,4-dichlorophenol, 2,4,6-trichlorophenol, 2,4,5-trichlorophenol)	Waste and tap waters	FASI-RMM-MEKC-UV	3.5-4 ng/mL	233-797	Environmental	Combined with HSD-DLLME	97
Adenosine, adenine, inosine, Hypoxanthine and xanthine	Melanoma cells	REPSM-MEKC-UV	90-170 nM	10	Biological applications		98
Gold nanoparticles		REPSM-MEKC-UV		5.2-42.1	Nano-science		99
Gold nanoparticles		REPSM-MEKC-UV		20-500	Nano-science		100
Ethyl 8-oxo-5,6,7,8-tetrahydro-thiazolo [3,2-a][1,3]diazepin-3-carboxylate (HIE-124)	Mice serum	REPSM-MEEKC-UV	21.6 ng/mL	25	Biological applications		101

^a Flu, fluorescence detection; SD-LLLME, single-drop liquid–liquid–liquid microextraction; HSD-DLLME, high-density solvent-based solvent de-emulsification dispersive liquid–liquid microextraction; RMM, reverse migrating micelles; REPSM, reversed electrode polarity stacking mode.

Table 3. Applications of other stacking-based techniques in CE.^a

Target analytes	Sample matrix	Preconcentration and separation modes, and detection scheme	LOD	Enrichment factor	Application	Comments	Ref. #
Tl(I), Pb(II), Cd(II), Zn(II), Cu(II) ions		EPC-CZE-C ⁴ D	7.7-100 nM	~100	Environmental		103
Tl(I), Pb(II), Cd(II), Zn(II), Cu(II) ions		EPC-CZE-C ⁴ D	8.8-58 nM	16-55	Environmental		104
Fluorescent dyes, BSA, λ-DNA, DNA marker, plasma protein		ES-CZE/CGE-LIF		>10 ⁹	Biological applications		105

^a EPC, electrochemical preconcentration; C⁴D, capacitively coupled contactless conductivity detection; ES, electrokinetic stacking.

Table 4. Applications of transient isotachophoresis in CE.^a

Target analytes	Sample matrix	Preconcentration and separation modes, and detection scheme	LOD	Enrichment factor	Application	Comments	Ref. #
2'-Deoxycytidine-5'-diphosphate, 2'-deoxycytidine-5'-triphosphate	Cell extracts	tITP-CZE-UV	73-85 nM	150	Biological applications		108
BSA and <i>E. coli</i> tryptic digests		tITP-CZE-MS	0.10-0.21 nM		Biological applications		109
Tryptic peptides		tITP-CZE-MS		10-86	Biological applications	Coupled with SPME and pseudo-tITP	110
Phosphopeptides	Tryptic digests from milk powder	tITP-CZE-MS			Biological applications		111
Twenty-one metabolites	Urine	tITP-CZE-MS	0.6-24 nM	>40	Biological applications		112
Opioid peptides (Dynorphin A, Endomorphin 1 and Methionine enkephalin)	Human plasma	tITP-CZE-MS	0.005-0.05 µg/mL	10	Biological applications		113
Oligosaccharides (dextran ladder)		tITP-CZE-C ⁴ D	10 nM		Biological applications		114
Methylephedrine, celiprolol, sotalol and indapamide	Urine	tITP-CZE-AD	42-950 fM	5500	Pharmaceuticals		115
3-Nitrotyrosine	Urine	tITP-MEKC-UV	0.07 µM	81	Biological applications		116
Gram-positive bacterial spore (<i>Bacillus globigii</i> , <i>S. cerevisiae</i>)		polymer-enhanced tITP-CZE-LIF	32000 cfu/mL		Biological applications		117
NSAIDs	Human plasma	pseudo-tITP-CZE-UV ^b	45	0.07 mg/L	Pharmaceuticals		118
Tricyclic antidepressants (imipramine, desipramine, amitriptyline, nortriptyline, clomipramine, norclomipramine, doxepine and nordoxepine)	Human breast milk	pseudo-tITP-CZE-UV ^b	50 ppb		Biological applications		119
Uremic toxin indoxyl sulfate	Serum	pseudo-tITP-CZE-UV ^b		50	Biological applications		120
Warfarin enantiomers	Urine	pseudo-tITP-CDCZE-UV ^b		10-15	Pharmaceuticals		121
Pyromellitic acid, trimellitic acid, phthalic acid, naphthalenedisulfonic acid	Urine	EOF counter-balanced ITP stacking boundary-CZE-UV		1100-1300	Biological applications		122
Estrogens (estrone, 17β-estradiol, estriol)	Water samples (waterworks, tap water, river, and fishpond)	FASS-tITP-CZE-EC	67-110 nM		Environmental		123
Rare-earth metal ions (La, Nd, Sm, Gd, Y,		EKS-CZE-UV	3.0-7.0 pM	500000	Environmental		125

Er, Yb)							
Four flavonoids (naringenin, hesperetin, naringin and hesperidin)	Extraction from <i>Clematis hexapetala pall</i>	EKS-CZE-UV	2.0-6.8 ng/mL	824-1515	Pharmaceuticals		126
Four phenolic acids		EKS-CZE-UV	1.0-2.5 ng/mL	300-440	Food analysis		127
Hypolipidaemic drugs (atorvastatin, fluvastatin, gemfibrozil, pravastatin and rosuvastatin)	Drinking and waste water	EKS-CZE-MS	180-560 ng/L	1000	Environmental	SEF = vs. FASS	128
DNA fragments		EKS-CGE-UV	7.7 pg/mL	170000	Biological applications		129
Bacteria		EKS-ITP-UV	3000 cells/mL		Biological applications		130
Aromatic acids	River water	EKS-NACE-UV	0.08-0.30 ng/mL	3868-6480	Environmental		131
2,5-Dihydroxybenzoic acid, 2,4-dihydroxybenzoic acid, p-toluic acid, 4-propylbenzoic acid, 3,5-dihydroxybenzoic acid and 4-heptylbenzoic acid	River water	EKS-NACE-UV	0.91-3.56 ng/mL (LVSEP/ASEI) 0.22-0.51 (EKS)	168-534(LV SEP/ASEI) 1333-3440 (EKS)	Environmental	Compared with LVSEP-ASEI	132
NSAIDs (diclofenac, diflunisal, fenoprofen, ibuprofen, indomethacin, ketoprofen, naproxen)		PA-EKS-CZE-UV	6.7-18.7 ng/L	7800-47000	Pharmaceuticals		133
Bacteria	River water	SEHI-FASI-ITP-UV	20000 cells/mL	500	Biological applications		134
DNA fragments		sweep-EKS-CGE-UV	0.49 ng/mL	3500	Biological applications	PSP = polyamidoamine	135

^a SPME, solid phase micro extraction; C4D, capacitively coupled contactless conductivity detection; AD, amperometric detection; SEHI, simultaneous electrokinetic and hydrodynamic injection. ^b Pseudo-tITP was named “acetonitrile stacking”.

Table 5. Applications of pH-mediated stacking in CE.^a

Target analytes	Sample matrix	Preconcentration and separation modes, and detection scheme	LOD	Enrichment factor	Application	Comments	Ref. #
Benzoic acid and sorbic acid	Beverage, vinegar and fruit jam	dynamic pH junction-CZE-UV	0.03-0.02 mg/L		Food analysis		144
Phenylalanine and tyrosine	Urine	dynamic pH junction-CZE-UV	36-49 nM		Biological applications		145
Mono- and di-saccharides		dynamic pH junction-CZE-LIF	0.13 nM	10	Biological applications		146
Peptides (leucine enkephalin, methionine enkephalin, dynorphin A, β -endorphin and angiotensin II)	Extractation from HL1 cells	dynamic pH junction-CZE-MS		4000-11000	Biological applications		147
Small peptides	Urine	dynamic pH junction-CZE-MS	0.2-2.0 nM		Biological applications		148
Tryptic phosphorylated peptides		pH-mediated stacking-CZE-MS			Biological applications	MALDI-MS	149
Zn(II), Cd(II) ions		pH-mediated stacking-CZE-UV	1-2 μ g/L		Environmental		150
Opiates (morphine, codeine and 6-monoacetylmorphine)	Saliva	pH-mediated stacking-CZE-UV	≤ 7 ng/mL	1000	Forensic		151
Serotonin, creatinine	Urine	pH-mediated stacking-CZE-UV	1.4-1500 ng/mL	0.5-360	Pharmaceuticals	Combined with USAEME	152
2'-Deoxyribonucleoside 5'-monophosphates		pH-mediated stacking-MEKC-UV		6	Biological applications		153
Steroid hormones (progesterone, 17 α ,20 β -dihydroxypregn-4-en-3-one, 17 α -hydroxyprogesterone, testosterone, estrone, 11-ketotestosterone, ethynyl estradiol, and 17 β -estradiol)	Fish plasma	pH-mediated stacking-MEKC-UV	4-11 nM		Biological applications		154
Disease-related specific DNA	Extracted from <i>feline panleukopenia</i> virus	base-stacking-CGE-UV		10	Biological applications	Combined with programmed field strength gradients	155
Hypolipidemic drugs (gemfibrozil, fluvastatin, and atorvastatin), NSAIDs (flufenamic acid, fenbufen, and sulindac) and diuretic drug (furosemide)		base-induced tITP-CZE-UV	0.1-0.4 μ g/mL	19-37	Pharmaceuticals		156
Tricyclic antidepressant (nortriptyline, imipramine, amitriptyline, and doxepine) and β -blocker (propranolol, pindolol, and nadolol) drugs		acid-induced tITP-CZE-UV	0.1-0.5 μ g/mL	10-12	Pharmaceuticals		157
Myoglobin		NRB-CZE-UV		130-470	Biological		158

applications						
Plant growth promoter (phenazine-1-carboxylic acid)	Soil sample	MRB-CZE-UV	17 ng/g	214	Environmental	159
Fumaric and maleic acids	Apple juice	tMCRB-CZE-UV	53.4-192 nM	80-100	Food analysis	160
D,L-Phenylalanine, D,L-threonine		tMCRB-heart cut 2D-CE-C ⁴ D	2 µM		Biological applications	161
<i>p</i> -Nitrophenol, <i>o</i> -nitrophenol, 2,4-dinitrophenol	Tap water	sandwich-type spontaneous injection-CZE-UV	1.7-5.0 µM	15-28	Environmental	162
Hydroxybenzoic acids (benzoic acid, ferulic acid and cinnamic acid)		electrokinetic accumulation/mobilization-MEKC-UV	0.3-1.0 nM	70000	Biological applications	Simulations and experimental studies 163
Ketoprofen enantiomers	Waste water	electrokinetic accumulation/mobilization-CDMEKC-UV	2.5-3.4 nM	8529-9921	Environmental	164

^a MALDI, matrix assisted laser desorption ionization; USAEME, ultrasound-assisted emulsification microextraction; NRB, neutralization reaction boundary; MRB, moving reaction boundary; tMCRB, transient moving chemical reaction boundary; C⁴D, capacitively coupled contactless conductivity detection.

Table 6. Applications of sweeping in CE.^a

Target analytes	Sample matrix	Preconcentration and separation modes, and detection scheme	LOD	Enrichment factor	Application	Comments	Ref. #
Anabolic androgenic steroids (androstenedione, testosterone, epitestosterone, boldenone, and clostebol)	Urine	sweep-MEKC-UV		108-175	Pharmaceuticals	Whole capillary sample filling	168
Tricyclic antidepressant (amitriptyline, clomipramine, doxepin and nortriptyline) and β -blocker (alprenolol, labetalol and propranolol) drugs	Waste water	sweep-MEKC-UV	7-27 ng/mL	213-305	Environmental		170
Digoxin and digoxigenin	Urine	sweep-MEKC-UV	0.30 mg/L	30	Pharmaceuticals		175
Pharmaceuticals (loratadine, indomethacin, ibuprofen and doxazosin)	Rabbit blood	sweep-MEEKC-UV		318-1355	Pharmaceuticals	Combined with in-tube SPME	176
Chiral polycyclic musks (galaxolide, tonalide, traseolide and phantolide)	Perfume samples	sweep-CDMEKC-UV	8-12 mg/L	12	Toxicological		169
Aspartic acid enantiomers	Cerebrospinal fluid, soymilk, beer	sweep-CDMEKC-LE DIF	0.24-0.25 nM	100-110	Food analysis		171
Dialkyl phthalates		sweep-EKC-MS	10-16 ppb	30-90	Environmental	PSP = Latex nanoparticles	174
CdSe nanocrystals		sweep-MEKC-CZE-UV	20		Nano-science		172
Ca(II), Mn(II), Zn(II) ions		sweep-CZE-MS	<0.15 mg/L		Environmental	PSP = EDTA	173
Haptoglobins (Hp 1-1 and Hp 2-2)	Serum	protein stacking by SDS-CZE-UV	31.3-40.0 μ g/mL	10	Biological applications	Stacked by complexation of proteins with SDS	177
Pharmaceutical racemates (alprenolol, 1-aminoindane, chlorpheniramine, isoproterenol, bromopheniramine)		tr-trapping-CDEKC-UV	25 ppb	230-240	Pharmaceuticals		179
Dialkylphthalates (diethyl-, dipropyl, and dibutyl-phthalates)		sweep-PF-MEKC-MS	0.02-0.1 μ g/mL	18-135	Environmental		195
Drugs (tolfenamic acid, diclofenac, indoprofen, and indomethacin) and herbicides (mecoprop and fenoprop)	Drinking water	transient micellar phase extractor-CZE-UV	0.1-1 μ g/mL	13-30	Food analysis	CTAB	183
Antipsychotic drugs (olanzapine, chlorpromazine, quetiapine fumarate, droperidol, risperidone, and aripiprazole)	Waste water	MSS-CZE-UV	0.03-0.09 μ g/mL	41-68	Pharmaceuticals		184
Tricyclic psychiatric drugs (promazine, chlorpromazine, prochlorperazine dimaleate, olanzapine, opipramol)	Urine	MSS-CZE-UV	6 ng/mL		Pharmaceuticals		185
Herbicides and profens		MSS-CZE-UV	0.06-0.12 μ g/mL	59-110	Environmental	IL surfactant	188
cationic alkaloids (berberine, palmatine and jatrorrhizine)	Plasma	MSS-CZE-UV	0.01-0.02 μ g/mL	47-53	Pharmaceuticals		189

Berberine, ephedrine	Urine	MSS-CZE-UV	0.5-1.1 ng/mL	300-1036	Biological applications		191
Gemfibrozil, fluvastatin, atorvastatin herbicides (difenzoquat, deiquat and paraquat), tricyclic antidepressants (amitriptyline, clomipramine, doxepin and nortriptyline), β -blockers (alprenolol, labetalol and propranolol) and hydrocortisone	Waste water	MSS-CZE-UV		10	Environmental	CTAB	192
	Urine	MSS-MEKC-UV		101-345	Pharmaceuticals		186
Alkaloids (berberine and jatrorrhizine)	Urine	MSS-NACE-UV	2-3 ng/mL	128-153	Biological applications		190
Atorvastatin, fluvastatin, gemfibrozil, lovastatin, mevastatin, pravastatin, and rosuvastatin	Waste water	sweep/AFMC/(FASS-sweep)-MEKC-UV	36-50 μ g/L	60-83	Environmental		187
β -Blocker (alprenolol, propranolol, nadolol, and labetalol) and tricyclic antidepressant (nortriptyline and clomipramine) drugs		sweep-MSS-CZE-UV	20 ng/mL	20-46	Pharmaceuticals		193
Hypolipidaemic drugs (gemfibrozil, fluvastatin and atorvastatin), NSAIDs (diflunisal, naproxen, ketoprofen, indoprofen and indomethacin), and herbicides (mecoprop and fenoprop)	Waste water	sweep-MSS-CZE-UV	0.05-0.55 μ g/mL	17-33	Environmental		194
Melamine, ammeline, ammelide, cyanuric acid	Liquid milk	CSEI/ASEI-sweep-MEKC-UV	0.01 ng/mL	3000-9200	Food analysis	SEF = vs. LVSS	198
Quinine, propranolol, strychnine, atropine, nicotine and codeine		CSEI-sweep-MEKC-UV	0.005–2 μ g/mL		Pharmaceuticals	Chemometric approach	204
Methadone, 2-ethylidene-1,5-dimethyl-3,3-diphenylpyrrolidine, 2-ethyl-5-Methyl-3,3-diphenyl-1-pyrrolidine	Serum	CSEI-sweep-MEKC-UV	200-400 pg/mL	1826-2352	Forensic		214
Amlodipine, donepezil, atorvastatin, irbesartan, rosuvastatin, and diclofenac	Plasma	successive CSEI-ASEI-sweep-MEKC-UV	10-50 ng/mL		Pharmaceuticals		199
phlorizin and quercitrin	Urine	EKI-sweep-MEKC-EC	6.7-10.4 ng/L	21000-23000	Pharmaceuticals	Swept by immobile SDS micelle under controlled EOF	200
Flavonoids (phlorizin, kaempferol, hesperetin, apigenin, rutin, hyperoside, quercitrin, quercetin)	<i>Herba Leonuri</i> extracts, mouse blood	EKI-sweep-AFMC-heart cut 2D MEKC/CZE-EC	7.3-36.4 ng/L	6000	Biological applications		213
Flavonoids (kaempferol, hesperetin, apigenin, rutin, hyperoside and quercetin)	<i>Leonurus cardiaca</i> sample	heart cut 2D MEKC-AFMC/CZE-EC	15-55 ng/mL		Food analysis		215
Alkaloids (theophyllin, berberine)	Urine	FASI-MSS-CZE-UV	0.3-0.4 ng/mL	640-6300	Pharmaceuticals		201
Antipsychotic drugs (olanzapine, chlorpromazine, quetiapine fumarate, droperidol, risperidone, and aripiprazole)	Urine	FASI-MSS-CZE-UV	1.1-1.8 ng/mL	717-2479	Pharmaceuticals		202

Imipramine, clomipramine, doxepine, nortriptyline, alprenolol, labetalol and propranolol	Waste water	SEHI-MSS-MEKC-UV	0.6–4.2 ng/mL	688-3000	Pharmaceuticals		203
Angiotensins		sweep-FASS-NACE-UV	7-11 nM	54-196	Biological applications		206
Vitamins	<i>Yerba mate</i> leaves, bacterial growth medium	FASS-sweep-MEKC-UV	0.05-0.10 µg/mL		Food analysis		207
<i>p</i> -Hydroxybenzoic acid esters	Cosmetic samples	LVSS-sweep-MEKC-UV	0.005–2 µg/mL	13-144	Food analysis	Chemometric approach	208
Carbamazepine and its five metabolites	Serum	LVSS-sweep-MEKC-UV	0.01 µg/mL	300	Pharmaceuticals		209
Sport doping drugs (testosterone, epitestosterone, epitestosterone glucuronide)		rLVSI-sweep-MEKC-UV	0.2-5.0 ng/mL	2500	Pharmaceuticals		210
Methotrexate and its eight metabolites	Cerebrospinal fluid	stacking-dynamic pH junction-sweep-MEKC-UV	0.1-0.3 µM		Biological applications		211
Dipeptides (Tyr-Phe, Tyr-Leu, Trp-Gly, and Ala-Gln)	Serum	dynamic pH junction-sweep-MEKC-LIF	1.0-5.0 pmol/L		Biological applications		212
Riboflavin	Cereal grains	SRMM-sweep-MEKC-LIF	0.29 ng/mL		Food analysis		216

^a MEEKC, microemulsion EKC; LEDIF, light emitting diode-induced fluorescence; SEHI, simultaneous electrokinetic and hydrodynamic injection; rLVSI, repetitive large volume sample injection.