Microextraction techniques combined with capillary electrophoresis in bioanalysis

Kohler, Isabelle; Schappler, Julie; Rudaz, Serge

How to cite

KOHLER, Isabelle, SCHAPPLER, Julie, RUDAZ, Serge. Microextraction techniques combined with capillary electrophoresis in bioanalysis. In: Analytical & bioanalytical chemistry, 2013, vol. 405, n° 1, p. 125–141. doi: 10.1007/s00216-012-6367-y

This publication URL: https://archive-ouverte.unige.ch/unige:28396
Publication DOI: 10.1007/s00216-012-6367-y

Anal Bioanal Chem (2013) 405:125–141
DOI 10.1007/s00216-012-6367-y

REVIEW

Microextraction techniques combined with capillary electrophoresis in bioanalysis

Isabelle Kohler · Julie Schappler · Serge Rudaz

Received: 4 July 2012 / Revised: 14 August 2012 / Accepted: 19 August 2012 / Published online: 11 September 2012

Abstract Over the past two decades, many environmentally sustainable sample-preparation techniques have been proposed, with the objective of reducing the use of toxic organic solvents or substituting these with environmentally friendly alternatives. Microextraction techniques (MEs), in which only a small amount of organic solvent is used, have several advantages, including reduced sample volume, analysis time, and operating costs. Thus, MEs are well adapted in bioanalysis, in which sample preparation is mandatory because of the complexity of a sample that is available in small quantities (mL or even μL only). Capillary electrophoresis (CE) is a powerful and efficient separation technique in which no organic solvents are required for analysis. Combination of CE with MEs is regarded as a very attractive environmentally sustainable analytical tool, and numerous applications have been reported over the last few decades for bioanalysis of low-molecular-weight compounds or for peptide analysis. In this paper we review the use of MEs combined with CE in bioanalysis. The review is divided into two sections: liquid and solid-based MEs. A brief practical and theoretical description of each ME is given, and the techniques are illustrated by relevant applications.

I. Kohler · J. Schappler · S. Rudaz (✉)
School of Pharmaceutical Sciences,
University of Geneva, University of Lausanne,
Bd d'Yvoy 20,
1211 Geneva 4, Switzerland
e-mail: Serge.Rudaz@unige.ch

I. Kohler · J. Schappler · S. Rudaz
Swiss Centre for Applied Human Toxicology,
University of Geneva, CMU,
Rue Michel-Servet 1,
1211 Geneva 4, Switzerland

Keywords Bioanalysis · Capillary electrophoresis · Environmentally sustainable chemistry · Green chemistry · Microextraction · Sample preparation

Abbreviations

μ-SLM	Micro-supported liquid membrane
BGE	Background electrolyte
C^4D	Capacitively-coupled contactless conductivity detector
CB-ICE	Chip-based immunoaffinity capillary electrophoresis
CE	Capillary electrophoresis
CME	Centrifuge microextraction
CM-LPME	Carrier-mediated liquid-phase microextraction
CM-SDME	Carrier-mediated single-drop microextraction
CZE	Capillary zone electrophoresis
DEHP	bis(2-Ethylhexyl) phosphate
DI-SPME	Direct-immersion solid-phase microextraction
DLLME	Dispersive liquid–liquid microextraction
DMD-LPME	Droplet–membrane–droplet liquid-phase microextraction
DSDME	Directly suspended droplet microextraction
EK	Electrokinetic injection
EME	Electro membrane extraction
ENB	1-Ethyl-2-nitrobenzene
ESI	Electrospray ionization
FASI	Field-amplified sample injection
GC	Gas chromatography
HS	Headspace
i-PrOH	Isopropanol
ILBE	In-line back-extraction
IT	Ion trap
LC	Liquid chromatography
LIF	Laser-induced fluorescence

LLE	Liquid–liquid extraction
LLLME	Liquid–liquid–liquid microextraction
LOD	Limit of detection
LVSS	Large-volume sample stacking
MCE	Microchip capillary electrophoresis
ME	Microextraction
MeCN	Acetonitrile
MEKC	Micellar electrokinetic chromatography
MeOH	Methanol
MEPS	Microextraction by packed sorbent
MIP	Molecularly imprinted polymer
NACE	Non-aqueous capillary electrophoresis
NPOE	2-Nitrophenyl octyl ether
NSAIDs	Non-steroidal anti-inflammatory drugs
OLBE	On-line back-extraction
PAH	Polycyclic aromatic hydrocarbons
PF	Preconcentration factor
PP	Protein precipitation
RAM	Restricted-access material
SBSE	Stir-bar-sorptive extraction
SL	Sheath liquid
SPE	Solid-phase extraction
THF	Tetrahydrofuran
tITP	Transient isotachophoresis
TOF	Time-of-flight
UHPLC	Ultra-high-pressure liquid chromatography
USAEME	Ultrasound-assisted emulsification microextraction

Introduction

The overall analytical procedure includes several consecutive steps—sampling, sample storage, sample preparation, separation of target analytes, detection, and data treatment. For analysis of complex samples and matrices, for example biological, environmental, or food analysis, the sample preparation is of utmost importance for obtaining the analytes of interest in a suitable injection solution able to provide reliable and accurate results. Sample preparation has substantial objectives before sample injection, including:

1. reducing or eliminating matrix interferents or undesired endogenous compounds;
2. increasing selectivity for targeted analyte(s);
3. preconcentrating the sample to enhance sensitivity; and
4. stabilizing the sample by reconstituting it in an inert solvent.

Although great improvements have been made in the development of fast separation techniques, sample pretreatment remains the most time-consuming step, accounting for ca two thirds of the entire analytical procedure [1]. In addition, because of the lack of automation of several offline procedures, sample preparation is also regarded as a primary source of analytical errors that can significantly affect the throughput [2].

Sample preparation can be based either on selective methods, e.g., the widely used solid-phase extraction (SPE) and liquid–liquid extraction (LLE), or non-selective methods, e.g., using membrane techniques or protein precipitation (PP). A common feature of all these conventional sample-preparation techniques is the relatively high consumption of solvents that are environmentally hazardous and health risks for humans. The advent of the concept of "green chemistry" at the beginning of the 1990s emphasized the need for non-toxic and environmentally friendly analytical procedures. The concept also promoted the use of environmentally sustainable sample-preparation methods with the development of solvent-free or miniaturized extraction methods [3, 4]. Different approaches can be envisaged when developing environmentally sustainable sample preparation:

1. solventless procedures [5, 6];
2. substitution of organic solvents with less-toxic alternatives, for example use of supercritical-fluid extraction, cloud-point extraction, subcritical water extraction, or extraction with ionic liquids [7, 8]; or
3. use of microextraction techniques (MEs), in which miniaturization of the extraction procedure not only minimizes the use of organic solvents but also the sample volume required.

MEs are defined as non-exhaustive procedures that use very small volumes of the extracting phase and for which the volume of sample is relatively large compared with that of the extracting phase [9]. MEs reduce or eliminate the consumption of solvents while simultaneously reducing sample volume, analysis time, and operating costs [10]. Many techniques have been developed over the last few decades for a variety of applications, i.e., in environmental analysis (pesticides, hormones) [11–13], food analysis [11, 13–15], and bioanalysis [16, 17] for clinical, toxicological and forensic purposes [18, 19] or doping analysis [20]. In bioanalysis, often only small amounts of the sample are available, typically in the mL range for urine and in the μL range for serum or plasma or alternative matrices, for example sweat, saliva, or tears. Because of the complexity of theses matrices and the low concentrations of the target analytes compared with endogenous interferents, sample preparation is mandatory, and MEs are particularly well adapted for this purpose.

A variety of analytical techniques, including separation-based approaches, can be implemented in combination with MEs in bioanalysis. Non-polar and volatile compounds are conveniently analyzed by gas chromatography (GC), whereas liquid chromatography (LC), including ultra-high-

pressure liquid chromatography (UHPLC), is extensively used in bioanalysis for both quantitative and qualitative purposes, because of its wide applicability to a large number of compounds with different physicochemical properties. Capillary electrophoresis (CE) is another powerful separation technique that is often used in bioanalysis, because of its high separation efficiency. As very small amounts of (μL range) or no organic solvents are required for CE analysis, its use in combination with ME techniques is regarded an attractive, environmentally sustainable analytical tool. Extracts can be directly injected for analysis, or evaporated and reconstituted in a very small volume. Because a few nL of sample is injected in CE, very high preconcentration factors (PFs) can be achieved, enhancing the overall sensitivity, which is a disadvantage of the capillary format.

Applications of ME techniques before to CE analysis have been reported over the past few decades in bioanalysis of low-molecular-weight compounds or small peptides. In this paper we review the MEs used in bioanalysis and combined with CE. It is divided into two sections: liquid and solid-based MEs. MEs are classified according to their extraction principle and improvement of extraction performance. A brief description and the theoretical concepts of each ME technique are introduced and discussed, and illustrated by relevant applications.

Liquid-based microextraction techniques

LLE, which involves partition of analytes between an aqueous sample and water-immiscible organic solvent, has been widely used in bioanalysis because of its simplicity and ease of implementation. LLE suffers from major drawbacks, for example emulsion formation at the interface of the immiscible phases, lack of selectivity (co-extraction of endogenous interferents), lack of automation, and use of large sample volumes and large amounts of toxic organic solvents that are environmentally harmful (up to 10 mL per mL of sample) [10, 16, 17, 21].

New methods based on the LLE principle or with original set-ups have been developed during the last two decades to overcome these drawbacks. Miniaturization of LLE has led to several new liquid-based ME techniques in which the total volume of organic solvent required has been reduced to the *sub*-mL level.

In 1996, Cantwell and co-workers [22] and Dasgupta and co-workers [23] were the first to propose the use of a solvent drop in the μL range as extractant, laying the foundation for liquid-phase microextraction (LPME). Cantwell and co-workers used an 8-μL drop of *n*-octane held at the end of a Teflon rod to extract 4-methylacetone from water [22], whereas Dasgupta and co-workers extracted sodium dodecyl sulfate (SDS) from a water sample with only 1.3 μL chloroform [23]. A variety of liquid MEs based on LPME were subsequently developed, leading to a large selection of miniaturized techniques that are still evolving. A schematic diagram of these techniques, based on their principle of extraction, is given in Fig. 1. All of the bioanalytical applications that use LPME-based techniques before CE are listed in Table 1.

Liquid-based ME techniques are derived either from single-drop microextraction (SDME), in which a single drop of water-immiscible solvent suspended from the tip of a syringe is immersed in the aqueous sample, or hollow-fiber liquid-phase microextraction (HF-LPME), in which a hollow polymeric fiber is used as a support for the acceptor (aqueous or organic) phase.

Single-drop microextraction (SDME)

SDME was introduced in 1997 by Jeannot et al. and He et al. [24, 25]. In the first study, a 1-μL drop of *n*-octane was suspended in a stirred aqueous sample from the tip of a microsyringe needle. After a few minutes, the drop was retracted into the needle and injected directly for gas chromatographic (GC) analysis [24]. He and Lee used the same method with a 1-μL drop of toluene that was immersed in the aqueous sample for 15 min before retraction and injection [25]. SDME uses very small amounts of organic extraction solvents, which enables important PFs to be achieved. The main problems with this method are lack of droplet stability at high stirring speeds and the high manual dexterity required. Moreover, SDME is only suitable for relatively non-polar analytes and suffers from low recovery and repeatability. Therefore, SMDE was regarded as be not suitable for biological matrices, in which an extra filtration step is necessary [2, 11, 26]. Many derived techniques based on SDME were thus proposed (Fig. 1), including liquid–liquid–liquid microextraction (LLLME) or dispersive liquid–liquid microextraction (DLLME), and used in combination with CE to obtain sufficient selectivity, sensitivity, and repeatability in bioanalysis.

Liquid-liquid-liquid microextraction (LLLME)

LLLME, also referred to as LPME by back-extraction, was first introduced in 1998 by Ma and Cantwell [27] and is particularly suitable for water-soluble analytes, for example ionizable compounds. In LLLME, the targeted analytes are first extracted from the aqueous sample (donor) into a water-immiscible organic phase (acceptor I) and then back-extracted into a separate aqueous phase (acceptor II). The transfer occurs by manipulating the pH in the donor and acceptor phases. LLLME is particularly suitable for CE analysis, because of the direct injection of the aqueous acceptor phase into the system.

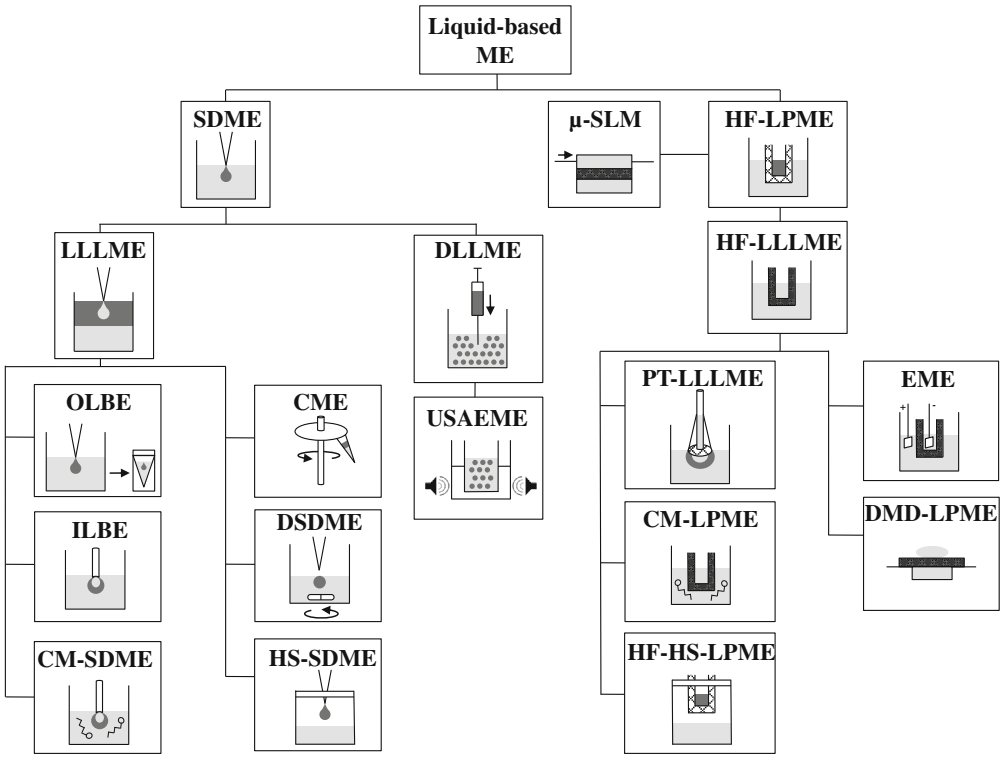

Fig. 1 Classification of liquid-based microextractions used in combination with CE. *Light gray*, aqueous phase; *dark gray*, organic phase; *cross hatched*, membrane or fiber

Extraction improvement

The particular configuration of CE enables on-line or in-line back-extraction to be performed. On-line back-extraction (OLBE) with field-amplified sample injection (FASI) was developed for analysis of cocaine and thebaine in urine samples [28]. Eight milliliters of urine were placed in a vial, and a 2-μL drop of chloroform was generated at the tip of a syringe and immersed in the sample. After extraction for 5 min, with stirring, the chloroform drop was retracted and transferred to another vial that was sealed with 40 μL acidified water for back-extraction. OLBE was performed by carefully immersing the capillary tip in the water plug. During high-voltage application, FASI occurred, and charged analytes moved rapidly from the organic phase to the capillary, stacked at the boundary with the high-conductivity background electrolyte (BGE). MeCN (20 %, *v*/*v*) was also added to the water plug to reduce conductivity, thus substantially enhancing sensitivity.

In-line back-extraction (ILBE) was performed with a water–organic drop hanging at the tip of the capillary [29]. In this case, the capillary was filled with acidic BGE (acceptor phase), and then 13 nL octanol was injected. After injection, the tip of the capillary was immersed in the urine sample, which had previously been made alkaline, and a backpressure was applied from the outlet to the inlet, forming a small drop of the acceptor phase that was covered with a thin organic layer hanging at the tip. After extraction, the acceptor phase was injected into CE. This configuration is well adapted to saline samples, for example urine; however, it is hardly achievable on a commercial CE instrument [26].

To enhance the transfer of analytes between the sample and organic phase, use of carriers with LLLME was envisaged by Choi et al. in 2011, in so-called carrier-mediated single-drop microextraction (CM-SDME) [30]. Amino acids were extracted from urine by use of nonane-1-sulfonic acid as carrier. Addition of this negatively charged carrier at a low pH with positively charged amino acids enabled the formation of a neutral ion pair that could be extracted into the organic phase. Octanol was chosen as the extracting phase because of its capacity to form hydrogen bonds with the ion pair. CM-SDME enabled 120-fold sensitivity improvement compared with CZE without SDME.

Drop stability improvement

Although the above-mentioned LLLME technique [28] has been shown to be fast, simple, inexpensive, and sensitive, it clearly suffers from drop instability. Therefore, in the same year, Fang et al. developed centrifuge microextraction (CME), which combines desalting, preconcentration, and removal of macromolecular contaminants and other interfering components in a single step [31]. After pH adjustment and addition of NaCl for the salting-out effect, 1 mL urine was mixed with 50 μL toluene and centrifuged at 10,000 rpm for 10 min. A lower-density water-immiscible solvent was chosen so the acceptor phase was at the top of the sample. During centrifugation, the centrifugal force applied by the rotor led to sedimentation of

Table 1 Liquid-based microextraction techniques used in combination with CE in bioanalysis

Analyte(s)	Matrix	ME	Sample volume	Organic solvent(s)	Organic solvent(s) volume	Analysis	PF/LOD	Ref.
Cocaine Thebaine	Urine	OLBE	8 mL	$CHCl_3$	4 μL	FASI–CZE–UV	120–340/2–10 $ngmL^{-1}$	[28]
Basic amines	Urine	ILBE	1.8 mL	*n*-octanol	13 nL	CZE–UV	1,000/0.5 $ngmL^{-1}$	[29]
Alkaloids	Urine	LLLME	4 mL	*n*-octanol	350 μL	MEKC–UV	1,583–3,556/0.2–1.5 $ngmL^{-1}$	[128]
Cyanide	Urine, saliva	HS-SDME	4.5 mL	–	–	CZE–UV after derivatization	58/0.08 μmol/L	[37]
Doping agents	Horse urine	HS-SDME	10 mL	$CHCl_3$–MeOH 90:10	5 μL	OT–CEC–UV	38–102/0.9–17.6 $ngmL^{-1}$	[38]
Toxic drugs	Urine	DLLME	4 mL	*i*-PrOH–CH_2Cl_2 70:30	2 mL 20 μL (*i*-PrOH, SL)	CZE–ESI–TOF MS	75–100/0.1–10 $ngmL^{-1}$	[40]
Serotonin Creatinine	Urine	USAEME	5 mL	Ethyl acetate	500 μL	CZE–UV	360/7.9 $nmolL^{-1}$ and 13.3 $\mu molL^{-1}$	[43]
Ephedrine derivatives	Urine, serum	CME	1 mL (urine) 20 μL (serum)	Toluene	50 μL	FASI–CZE–UV	3,800/0.15–0.25 $ngmL^{-1}$	[31]
Steroid hormones	Urine	CME	1.3 mL	Cyclohexane	100 μL	MEKC–UV	500/5–15 $ngmL^{-1}$	[32]
Alkaloids	Urine	DSDME	3.5 mL	*n*-octanol	< 60 μL	CZE–UV	231–524/8.1–14.1 $ngmL^{-1}$	[34]
Amino acids	Urine	CM-SDME	20 mL	*n*-octanol	24 nL	CZE–UV	120/70–500 $nmolL^{-1}$	[30]
NSAIDs	Urine	μ-SLM	4 mL	Dihexyl ether and MeOH	n.d.	MEKC–UV	1.2–1.7 μgmL^{-1}	[129]
Nitroimidazoles	Pig liver tissues	μ-SLM	0.5-5 g	2-Phenylpropane	n.d.	MEKC–UV	0.01–0.99 μgmL^{-1}	[47]
Bambuterol	Plasma	μ-SLM	500 μL	MeOH	n.d.	CZE–UV	n.d.	[130]
Bambuterol	Plasma	μ-SLM	350 μL	MeOH	n.d.	CZE–UV	14	[46]
Amino acids	Serum, plasma	μ-SLM	12.5 μL	ENB–DEHP	2.5 μL	CZE–C^4D	0.75–2.5 $\mu molL^{-1}$	[48]
Methamphetamine	Urine, serum	HF-LLLME	2.5 mL	1-octanol	n.d.	CZE–UV	75/5 $ngmL^{-1}$	[49]
Organomercury	Hair	HF-LLLME	12 mL	Bromobenzene	n.d.	LVSS–CZE–UV	2,610–4,580/0.03–0.14 μgmL^{-1}	[57]
NSAIDs	Urine	HF-LLLME	2.5 mL	Dihexyl ether	25 μL	CZE–UV	75–100/1 $ngmL^{-1}$	[52]
Methamphetamine Naproxen	Urine, plasma	HF-LLLME	4.0 mL (urine) 2.5 mL (plasma)	*n*-octanol	n.d.	CZE–UV	30–125	[50]
Citalopram Desmethylcitalopram	Plasma	HF-LLLME	1 mL	Hexyl ether	n.d.	CZE–UV	25–30/5 and 5.5 $ngmL^{-1}$	[54]
Mianserin	Plasma	HF-LLLME	0.5 mL	Di-*n*-hexyl ether	n.d.	CZE–UV	4 $ngmL^{-1}$	[131]
Citalopram Desmethylcitalopram	Plasma	HF-LLLME	1 mL	Dodecyl acetate	n.d.	CZE–UV	19–31/1.4–3.4 $ngmL^{-1}$	[53]
Methamphetamine Citalopram	Urine, plasma Whole blood	HF-LLLME	2.5 mL	Hexyl ether	n.d.	CZE–UV	95–145/2 $ngmL^{-1}$	[51]
Basic drugs	Plasma	HF-LLLME	250 μL	Dihexyl ether and MeOH	n.d.	CZE–UV	<20 $ngmL^{-1}$	[55]
Antidepressants	Human milk	HF-LLLME	500 μL	Polyphenylmethylsiloxane	n.d.	CZE–UV	14–23/<50 $ngmL^{-1}$	[56]
Rosiglitazone	Plasma, urine	HF-LLLME	5.0 mL (urine) 2.5 mL (plasma)	Dihexyl ether	n.d.	CZE–UV	280/2.83 $ngmL^{-1}$	[132]
Tricyclic antidepressants	Plasma	HF-LLLME	250 μL	Dihexyl ether	10 μL	CZE–UV	0.02–0.03 $\mu molL^{-1}$	[133]

Table 1 (continued)

Analyte(s)	Matrix	ME	Sample volume	Organic solvent(s)	Organic solvent(s) volume	Analysis	PF/LOD	Ref.
Cyanide	Urine, saliva	HF-HS-LPME	4.5 mL	–	–	CZE–UV	0.01 $\mu mol L^{-1}$	[134]
Polar drugs	Plasma, urine	CM-LPME	100 µL	1-octanol	n.d.	CZE–UV	10/< 1 $\mu g mL^{-1}$	[59]
Basic drugs	Plasma	CM-LPME	50 µL	*n*-octanol	n.d.	CZE–UV	<0.5 $\mu g mL^{-1}$	[60]
Basic drugs	Plasma, urine	EME	100 µL	NPOE	~15 µL	CZE–UV	n.d.	[62]
Basic drugs	Plasma, whole blood	EME	500 µL	1-ethyl-2-benzene	n.d.	CZE–UV	<0.5 $\mu g mL^{-1}$	[64]
Angiotensins	Plasma	EME	500 µL	1-octanol–DEHP	n.d.	CZE–UV	<2.5 $ng mL^{-1}$	[66]
Basic drugs	Plasma, urine Human milk	EME	1 mL	1-isopropyl-4-nitrobenzene	n.d.	CZE–UV	Up to 22	[65]
Amino acids	Serum, plasma Whole blood,	EME	36 µL	ENB–DEHP	n.d.	CZE–C^4D	0.15–10 $\mu mol L^{-1}$	[67]
Lithium	Urine Serum, plasma Whole blood	EME	35 µL	1-octanol	n.d.	CZE–C^4D	9 $nmol L^{-1}$	[68]
Basic drugs Amino acids	Urine, serum	EME	875 µL (urine) 145 µL (serum)	ENB/DEHP	n.d.	CZE–UV	1–4 $ng mL^{-1}$ (bases) 0.6–3 $\mu mol L^{-1}$ (acids)	[135]
Amlodipine	Plasma, urine	EME	1 mL (serum) 1.5 mL (urine)	NPOE	n.d.	CZE–UV	Up to 150/3 $ng mL^{-1}$	[69]
Trimipramine	Plasma, urine	EME	1.25 mL (serum) 2.5 mL (urine)	NPOE	n.d.	CZE–UV	120–149/8–10 $ng mL^{-1}$	[70]
Basic drugs	Urine	EME	10 µL	NPOE	1 µL	CZE–UV	< 1 $\mu g mL^{-1}$	[71]
Inorganic and organic mercury	Hair	PT-LLLME	0.25 g	Toluene MeCN Acetone	200 µL (Toluene) 225 µL (MeOH)	LVSS–CZE–UV	12,138	[58]
Basic and acidic drugs	Urine	DMD-LPME	15 µL	1-octanol	1 µL	MCE–LIF	2	[72]
Tramadol, paracetamol and metabolites	Urine	DMD-LPME	4 µL	1-octanol	~1 µL	MCE–ESI–MS	2/9.3 $nmol L^{-1}$ for model compound	[73]

n.d., not defined

LOD is determined at a signal-to-noise ratio of 3

macromolecules whereas diffusion enabled transfer of the targeted compounds to the acceptor phase. The supernatant was directly injected in CE–UV with FASI, leading to a limit of detection (LOD) of 0.15 $ngmL^{-1}$. CME has also been used for analysis of steroids (e.g., testosterone and progesterone) in urine by micellar electrokinetic chromatography (MEKC) [32].

In 2006, Yangcheng et al. proposed directly suspended droplet microextraction (DSDME) to further improve drop stability. Here, the microdroplet of solvent is suspended at the top in the center of the aqueous sample before sampling [33]. A symmetrical rotated flow field is created by a stirring bar that is placed on the bottom of the cylindrical sample cell to ensure the droplet suspension. This rotation also intensifies transfer of analytes to the inside of the droplet. DSDME has been combined with single-drop back-extraction and CE for analysis of alkaloids in urine samples [34]. After the first extraction in a large microdrop (approx. 60 μL) of *n*-octanol, alkaloids were back-extracted in a 1-μL aqueous drop that was immersed in the organic phase droplet. PFs of greater than 500 were achieved with lower solvent consumption and shorter extraction time than those of LLLME.

Introduced by Theis et al. [35] in 2001, headspace single-drop microextraction (HS-SDME) has excellent extraction and preconcentration performance for volatile compounds. With a suspended drop in the gaseous phase (headspace), this method enables rapid stirring of an aqueous sample, for a shorter analysis time, without affecting drop stability. Moreover, non-volatile matrix interferences are reduced or eliminated [19, 26, 36]. HS-SDME has also been used in combination with CE analysis with in-drop derivatization. Free cyanide was solventlessly extracted from smoker and non-smoker urine and saliva [37], using water to extract volatile and water-soluble compounds. An aqueous 5-μL drop containing Ni(II)–NH_3 as derivatization agent for CE analysis was used for the extraction. In the basic acceptor phase, cyanide reacted with Ni^{2+} to form a stable $Ni(CN_4)^{2-}$ complex analyzed by CE–UV at 257 nm. Water-based HS-SDME was very selective, despite the rather universal detection wavelength, because the non-volatile interferents remained unaffected in the sample. HS-SDME with a chloroform–MeOH mixture as extracting drop has also been used to extract seven toxic compounds from horse urine samples at room temperature, before analysis by open tubular capillary electrochromatography (OT-CEC) [38].

Dispersive liquid–liquid microextraction (DLLME)

In DLLME, which was first introduced by Rezaee et al. in 2006, the extracting solvent is mixed with a dispersing solvent that is miscible both with the former and with the aqueous sample [39]. The mixture is rapidly injected into the sample with a syringe, producing high turbulence that leads to the formation of tiny droplets. Because of the large surface area between the extracting droplets and sample, the extraction time is drastically reduced. After centrifugation, the sedimented phase at the bottom of the tube is collected and either injected directly or evaporated to dryness before reconstitution and injection.

DLLME combined with CE and time-of-flight mass spectrometry (TOF/MS) was used for qualitative toxicological screening of urine samples [40]. An experimental design strategy was used to increase the extraction efficiency. CH_2Cl_2 and *i*-PrOH were selected as extracting and dispersing solvents, respectively, with a total volume of 2 mL. Because of a high PF (more than 130) and the high sensitivity and selectivity of CE–TOF/MS, LODs down to the *sub*-$ngmL^{-1}$ range were obtained for more than 30 toxic basic compounds and their main metabolites and confirmed by real case analysis.

Extraction improvement

One of the main disadvantages of DLLME is the need to use a dispersing solvent to create an emulsion, which can reduce the partition coefficient of the analytes in the extracting phase and increase total solvent consumption. The dispersing solvent can be substituted by using ultrasound to achieve ultrasound-assisted emulsification-microextraction (USAEME) [41]. Based on previous work on ultrasound-assisted sample preparation [42], USAEME is beneficial for promoting emulsion formation, extending the contact surface between both phases, and reversing the potential coalescence effect. Increasing the temperature also enables efficient and fast extraction [41]. A serial USAEME procedure was developed for analysis of creatinine and serotonin in urine samples [43]. Five hundred microliters of ethyl acetate was added to 5 mL urine and the sample was immersed in an ultrasonic bath for 5 min at 40 Hz. The emulsion was centrifuged, and the organic supernatant mixed with 25 μL 0.1 $molL^{-1}$ HCl. Back-extraction was performed by 3-min ultrasonication at 40 Hz. After centrifugation, the sedimented acceptor phase was collected and injected by use of a pH-mediated stacking procedure. With serial USAEME and sample stacking, a PF of 360 was obtained for serotonin.

Hollow-fiber-based liquid-phase microextraction (HF-LPME)

The chemical principle of HF-LPME is derived from supported-liquid membrane (SLM) extraction, which was previously developed by Jönsson and coworkers [44]. In SLM, analytes are extracted through a flat porous polymeric membrane sheet with continuous sample pumping. SLM was first miniaturized ("μ-SLM") in 1996 by Jönsson and coworkers [45] and applied to the analysis of bambuterol in plasma samples that were continuously pumped to on-line

SLM–CZE [46]. An in-line SLM approach with a Teflon micromembrane unit glued to a plastic microtube integrated in the CE vial was developed by Nozal et al. for analysis of nitroimidazoles in pig liver tissues homogenized in water [47]. Very recently, Kuban and Bocek proposed on-line μ-SLM-CE with a planar SLM screwed between two PTFE blocks to determine amino acids in plasma or serum [48]. This home-built set-up did not require additional pumps.

In contrast to μ-SLM, HF-LPME is performed without any pumping device. It was introduced in 1999 by Pedersen-Bjergaard and Rasmussen [49]. In HF-LPME, the extracting phase is placed inside the lumen of a porous polypropylene fiber (pore size 0.2 μm) of minimal dimensions used in different configurations, e.g., U-shaped, rod-like, 96-well, or directly connected to a microsyringe [21, 44]. The polymeric fiber, which is compatible with a broad range of organic solvents, enables use of a larger extraction volume compared with SDME and acts as a physical barrier between phases, avoiding undesirable emulsions and enhancing cleanup efficiency [19]. HF-LPME can be performed in either two or three-phase systems. In three-phase systems, referred to as hollow-fiber-based liquid–liquid–liquid microextraction (HF-LLLME), supported liquid membrane microextraction (SLMME), or, rather improperly, LPME, the analytes are extracted from the aqueous sample through the organic film (a few microliters) that is present in the pores of the aqueous acceptor phase in the lumen of the hollow fiber. HF-LLLME is well suited to extraction of polar or ionizable compounds and particularly suitable for CE analysis.

Hollow-fiber-based liquid–liquid–liquid microextraction (HF-LLLME)

Pedersen-Bjergaard and Rasmussen with co-workers have developed many applications of HF-LLLME in combination with CE. Methamphetamine [49–51], non-steroidal anti-inflammatory drugs (NSAIDs) [52], naproxen [50], citalopram and metabolites [51, 53, 54], and a variety of basic drugs [55] have been successfully extracted from urine and serum or plasma. HF-LLLME has also been used to extract antidepressants from human milk [56]. Human milk is characterized by high protein, fat, and carbohydrate content, which can affect the recovery and repeatability of the extraction procedure. Because of interaction of antidepressants with fat and proteins, recovery from milk was lower than from water. Thus, PP was implemented, with addition of hydrochloric acid to the sample before centrifugation and extraction to remove the fat-rich layer and release unbound drugs, leading to recovery of 50–70 %. Li et al. used HF-LLLME for extraction of organomercury from human hair, with the fiber pores impregnated with bromobenzene [57]. Hair samples were first rinsed with detergent and acetone, and air-dried before cutting and leaching. The leached solution was centrifuged, and the supernatant collected for HF-LLLME. An aqueous acceptor phase containing L-cysteine for organomercury complexation was injected with large volume sample stacking (LVSS), enabling enrichment of more than 4,000.

Extraction improvement

A new LPME-based technique referred to as phase-transfer-based liquid–liquid–liquid microextraction (PT-LLLME) was developed in 2011 by Li et al. for extraction of organic and inorganic mercury from hair [58]. In this homemade set-up, a porous, hydrophilic, nylon-membrane-supported extraction tip was built and used with 15 μL aqueous acceptor phase. MeCN and dodecylamine were added to the sample before extraction as intermediate solvent and complexing reagent, respectively. MeCN improved the dispersion of water-immiscible dodecylamine in the aqueous sample to ensure maximum contact with the mercury. Compared with mercury extraction by HF-LLLME, PT-LLLME provided the potential for simultaneous speciation of inorganic and organic mercury and improved the sensitivity with enhanced extraction efficiency.

Use of carriers, also used in SDME (section “Liquid–liquid–liquid microextraction (LLLME)”, subsection “Extraction improvement”), was first introduced in 2003 by Ho et al. in the so-called carrier-mediated liquid-phase microextraction (CM-LPME) to enhance extraction recovery of polar or ionic analytes [59]. The carriers form lipophilic complexes with the target analytes, promoting the transport of the analytes through the organic membrane. Polar basic compounds could be extracted from plasma and urine samples, through the 1-octanol layer into the aqueous acceptor phase, with good recovery, after addition of sodium octanoate (ion-pair reagent) to the sample. The pH of the sample had to be adjusted so the analytes and carrier were ionized in such a way to enable the formation of ion-pair complexes that could diffuse through the membrane. Numerous carriers, including organic borates, phosphates, sulfates, and carboxylic acids, were investigated at different concentrations with a special emphasis on their compatibility with plasma samples [60]. Bromothymol blue (sulfate carrier) resulted in the best recovery from the plasma samples. Interestingly, recovery was enhanced when sodium sulfate was added to the sample to reduce matrix effects.

In 2005, Lee and coworkers developed hollow-fiber-protected headspace liquid-phase microextraction (HF-HS-LPME), in which the hollow fiber protected and held the extractant droplet in the headspace [61]. The surface area between the organic and acceptor phases was dramatically enhanced compared with HS-SDME (section “Liquid–liquid–liquid microextraction (LLLME)”, subsection “Drop

stability improvement"), increasing the extraction efficiency. HS-HF-LPME was used to extract free cyanide from urine and saliva with a simultaneous in-fiber derivatization to form a stable $Ni(CN)_4^{2-}$ complex. Lower LODs (0.01 $\mu molL^{-1}$ versus 0.08 $\mu molL^{-1}$) and similar recovery (90–105 %) were obtained compared with HS-SDME [37]. HF-HS-LPME is an effective alternative to HS-SDME for quantitative analysis of volatile compounds.

Throughput improvement

In 2006, Pedersen-Bjergaard and Rasmussen proposed use of an electrically-driven force to aid extraction of charged compounds and to speed HF-LLLME [62, 63]. This technique was first referred to as "electro membrane isolation" (EMI) and was later termed electro membrane extraction (EME). Two platinum electrodes are placed in the sample solution and in the aqueous acceptor phase in the lumen of the fiber. A potential (typically 300 V) is applied, and charged analytes migrate through the membrane toward the oppositely charged electrode in the acceptor solution in less than 5 min. Interesting clean-up, enrichment, and isolation of basic compounds with 2-nitrophenyl octyl ether (NPOE) as organic solvent were observed with high extraction recovery (>70 %) from plasma and urine.

Pedersen-Bjergaard, Rasmussen, and co-workers showed the benefits of EME for analysis of basic drugs (e.g., analgesics, antidepressants, and antiepileptics) in plasma and whole blood [64], or urine and human milk [65]. They also evaluated the potential of EME as a fast and effective extraction technique for peptides (angiotensin as the model peptide) in plasma [66].

Other groups evaluated EME for a variety of applications, for example extraction of amino acids [67], lithium [68], amlodipine enantiomers [69], and trimipramine enantiomers [70] from urine, plasma or serum, or whole blood.

A miniaturized form of EME, termed drop-to-drop LPME, has been proposed for extraction of basic drugs from urine and plasma [71]. A small well with a volume of 15 μL was pressed into 5-cm^2 aluminium foil connected to the power supply's positive outlet. The well, containing 10 μL sample, was covered with the membrane and a 10-μL acceptor droplet. Recovery of 33–47 % was obtained with excellent clean-up, short extraction time, and very low solvent and sample consumption.

In 2010, this miniaturization was built upon with the development of on-line droplet–membrane–droplet LPME (DMD-LPME) [72]. The extraction set-up was the same as in Ref. [71] and was combined on-line with microchip capillary electrophoresis (MCE) with fluorescence detection. DMD-LPME was directly compatible with MCE because of the very low acceptor phase volume. After 5 min, analysis of two model analytes spiked in blank urine led to recovery of 15 and 25 %, which was lower than from aqueous standards. However, DMD-LPME was found to be competitive for high-throughput analysis, because of the high extraction speed and its feasibility for coupling with rapid microfluidic analysis. DMD-LPME has also been combined with MCE for drug metabolism studies with ESI-triple quadrupole MS detection [73]. Compared with SPE, DMD-LPME enabled faster analysis and higher selectivity for phase I metabolites.

Solid-based microextraction techniques

SPE is the most widely used technique for clean-up, preconcentration, and selective extraction. Over the last few decades, a large variety of commercial silica-based or polymeric sorbents (e.g., normal-phase, reversed-phase, ion-exchange mode, mixed-mode, and, more recently, molecularly imprinted polymers (MIP), monoliths, and restricted-access media, RAM) have been developed to enable extraction of a variety of analytes with divergent chemical structure and polarity, with careful attention to higher loading capacity and efficiency. SPE can be automated easily, furnishes high recovery, and is claimed to be highly selective in relation to matrix interferences [2, 17]. However, conventional SPE has some limitations, for example relatively high solvent consumption and batch-to-batch variability [16, 17]. A significant amount of progress has been made with SPE to substantially reduce solvent consumption and increase sample throughput, for example the advent of column-switching systems with on-line extraction, or the multi-well plate format. Another substantial step was achieved with SPE miniaturization and the development of new microextraction techniques (solid-based MEs), for example solid-phase microextraction (SPME), microextraction by packed sorbent (MEPS), and stir-bar-sorptive extraction (SBSE), all of which have several advantages and result in significantly improved sample preparation. The solid-based MEs used in combination with CE are presented in Fig. 2, and all the bioanalytical applications are listed in Table 2.

Solid-phase microextraction (SPME)

SPME was introduced in 1990 by Arthur and Pawliszyn [74]. A small amount of sorptive, homogenous, non-porous extracting phase dispersed on the surface of or inside a solid support is exposed to the sample for a specific period of time until equilibrium is reached [75, 76]. The main commercially used sorbents are polydimethylsiloxane (PDMS) for rather non-polar or volatile compounds and polyacrylate (PA), PDMS–divinylbenzene (PDMS–DVB), or Carbowax–divinylbenzene (CW–DVB) for polar compounds. Extraction can be performed in two main formats: fiber SPME and in-tube SPME.

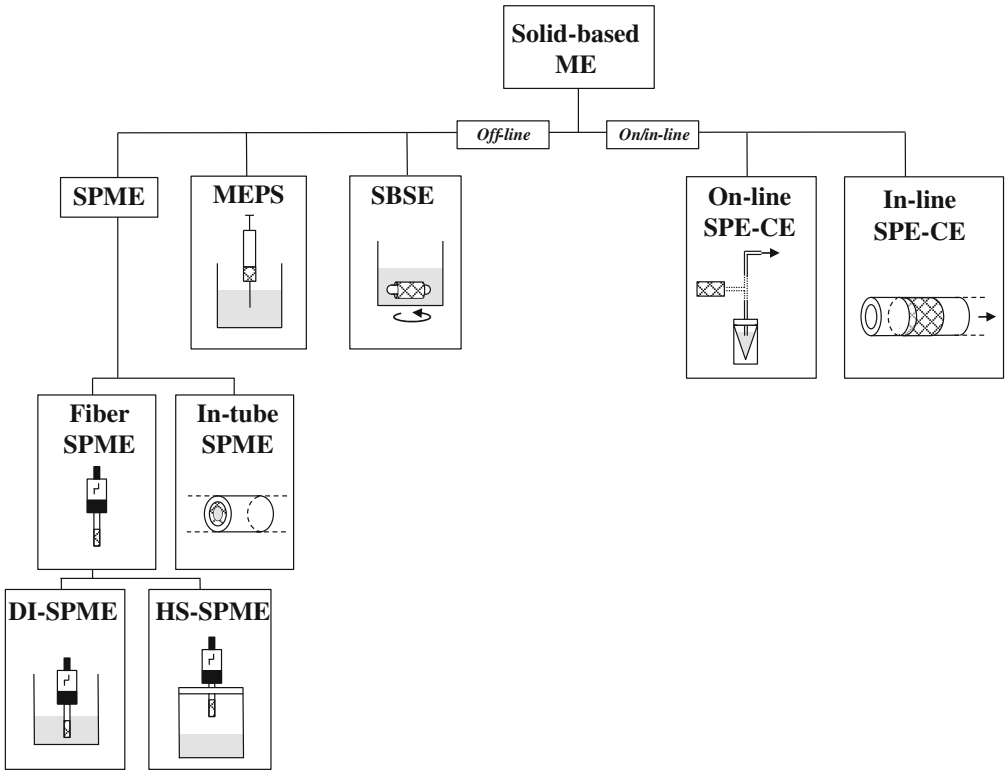

Fig. 2 Classification of solid-based microextractions used in combination with CE. *Light gray*, aqueous phase; *cross hatched*, solid support

Fiber solid-phase microextraction (fiber SPME)

In fiber SPME, the sorbent (variable film thickness) is coated on the external surface of a fused-silica fiber tip as an appropriate polymeric stationary phase. The device, a modified syringe, consists of a fiber assembly with the built-in fiber inside the needle and an assembly holder. A plunger is used to move the coated fiber inside or outside the needle [19, 77]. Two extraction modes can be used with fiber SPME: direct immersion of the fiber in the aqueous sample (DI-SPME) or headspace extraction (HS-SPME), which was first described in 1993 [78].

Direct-immersion fiber solid-phase microextraction (DI-SPME)

DI-SPME entails direct immersion of the fiber into the aqueous sample with consequent stirring, enabling transfer of non-volatile analytes into the coating [76]. Barbiturates and benzodiazepines have been extracted by use of a PA-coated fiber that was immersed in 10 mL urine for 2 h at 60 °C. After extraction, the targeted drugs were desorbed into 20 μL MeCN for 30 min and analyzed by MEKC on neutral polyacrylamide-coated capillaries [79, 80].

Headspace fiber solid-phase microextraction (HS-SPME)

HS-SPME has been shown to be advantageous, mainly for volatile compounds, because of its higher speed, higher recovery, greater selectivity, longer fiber lifetime, and lower fiber contamination than for DI-SPME, but it is only suitable for highly volatile compounds [76, 81]. Instead of using a conventional PDMS, PA, or poly(vinyl chloride) fiber, Zeng and coworkers developed HS-SPME with a calix{4}arene fiber [82], for propranolol determination, and co-poly (butyl methacrylate–hydroxy-terminated silicone oil), using a sol–gel coating, for extraction of ephedrine derivatives in urine [83]. After a second back-extraction step in a MeCN–water solution (less than 20 μL MeCN), the analytes were injected with the FASI stacking method, leading to important PFs.

In-tube SPME

In-tube SPME, which was introduced in 1997, was primarily developed to overcome the inherent problems of fiber SPME, i.e., fiber fragility, low sorption capacity, and bleeding of fiber coatings, and to provide an automation option [84]. In this method, targeted compounds are directly extracted into the internally coated stationary phase of a fused-silica capillary, enabling on-line coupling with CE [85]. In-tube SPME is a type of so-called capillary MEs, which also include open-tubular trapping, wire-in-tube SPME, fiber in-tube SPME, sorbent-packed capillary in-tube SPME, and monolithic capillary in-tube SPME [75, 84, 86]. Capillary MEs are distinguished from the composition of the extraction stationary phase (fiber, polymer, sorbent) and its packing [86] and can be used on-line with CE.

Table 2 Solid-based microextraction techniques used in combination with CE in bioanalysis

Analyte(s)	Matrix	ME	Sample volume	Organic solvent(s)	Organic solvent(s) volume	Analysis	PF/LOD	Ref.
Ephedrine Pseudoephedrine	Urine, serum	MIP-SPME	5 mL	Toluene Ethanol MeOH	5 mL 7 mL n.d.	CZE–UV	0.96 and 1.1 $ngmL^{-1}$ (water sample)	[136]
Ephedrine derivatives	Urine	HS-SPME	5 mL	MeCN	16 µL	FASI–CZE–UV	3–5 $ngmL^{-1}$	[83]
Propranolol enantiomers	Urine	HS-SPME	5 mL	MeCN	10 µL	FASI–CZE–UV	8–10 $ngmL^{-1}$	[82]
Barbiturates	Human urine, bovine serum	DI-SPME	3.5 mL	–	–	CZE–UV	Up to 60/0.1–0.3 μgmL^{-1} (urine) and 1 μgmL^{-1} (serum)	[137]
Barbiturates, benzodiazepines	Urine	DI-SPME	10 mL	MeCN	20 µL	MEKC–UV	<1 μgmL^{-1}	[79, 80]
Amphetamines	Urine	Monolithic in-tube SPME	40 µL	MeOH	n.d.	EK–CZE–UV	25–34 μgmL^{-1}	[88]
Opiates	Urine	Monolithic in-tube SPME	1 mL	MeOH	325 µL	EK–CZE–UV	6.6–19.5 $ngmL^{-1}$	[89]
Ephedrine Pseudoephedrine	Urine, plasma	Monolithic in-tube SPME	1 mL	MeOH MeCN	300 µL (MeOH) ~100 µL (MeCN)	LVSS–CZE–UV	5.3–8.4 $ngmL^{-1}$	[90]
Angiotensin II receptor antagonists	Urine	Monolithic in-tube SPME	2 mL	MeCN	500 µL+BGE	CZE–UV	15–20 $ngmL^{-1}$	[91]
Propranolol enantiomers	Urine	In-tube SPME	1 mL	MeOH MeCN	35 µL (MeOH) 100 µL (MeCN)	CEC–UV	4 and 7 $ngmL^{-1}$	[138]
Tricyclic antidepressants	Urine	Fiber-in-tube SPME	1 mL	MeCN	1.8-2.2 µL+BGE	CZE–UV	>100/44–153 $ngmL^{-1}$	[87]
Caffeine, paracetamol, acetylsalicylic acid	Bovine plasma	RAM capillary in-tube SPME	<10 µL	MeOH	n.d.	CZE–UV	0.3–1.9 $ngmL^{-1}$	[92]
PAHs	Fish bile	SBSE	0.3 g	MeCN	150 µL	MEKC–UV	2–11 μgmL^{-1}	[99]
Fluoroquinolones	Urine	MEPS	48 µL	MeCN MeOH	n.d.	NACE–ESI–MS	6.3–10.6 μgmL^{-1}	[96]
Anesthetic drugs	Plasma	MEPS	200 µL	MeCN MeOH	n.d.	NACE–ESI–MS	10.4–15.2 μgL^{-1} (free) 0.6–1.6 $ngmL^{-1}$ (total)	[95]
Opioids	Urine	In-line SPE	~60 µL	MeOH *i*-PrOH	~30 nL (MeOH) ~45 µL (*i*-PrOH)	CZE–ESI–MS	0.013–0.210 $ngmL^{-1}$	[139]
Methionine encephalin	Cerebrospinal fluid	In-line SPE	3.2 µL	MeCN MeOH	40 nL+conditioning (MeCN) ~90 µL (MeOH, SL)	CZE–ESI–MS	40/1 $ngmL^{-1}$	[117]
Enkephalin peptides	Cerebrospinal fluid	On-line SPE	100 µL	MeCN	1.7 µL (elution and rinse) ~20 µL (SL)	CZE–ESI–IT/MS	1,000/1.5–3 $ngmL^{-1}$	[103]
Cephalosporins	Cow plasma	On-line SPE	50 µL	MeCN	1.8 µL	CZE–UV	100 $ngmL^{-1}$	[104]
Opioid peptides	Plasma	In-line SPE	200 µL	MeOH *i*-PrOH MeCN	~40 nL (MeOH, elution) ~30 µL (*i*-PrOH, SL) 1200 µL (MeCN, PP)	CZE–ESI–IT/MS	100–10,000/0.1–10 $ngmL^{-1}$	[112]
Neuropeptides	Plasma	In-line SPE	200 µL	MeOH *i*-PrOH MeCN	~40 nL (MeOH, elution) ~50 µL (*i*-PrOH, SL) 1400 µL (MeCN, PP)	CZE–ESI–IT/MS	100–10,000/0.1–10 $ngmL^{-1}$	[111]
Opioid peptides	Plasma	In-line SPE	200 µL	MeOH *i*-PrOH	~40 nL (MeOH, elution) ~10 µL (*i*-PrOH, SL)	tITP–CZE–TOF/MS	Up to 5,000/0.1 $ngmL^{-1}$	[113]

Table 2 (continued)

Analyte(s)	Matrix	ME	Sample volume	Organic solvent(s)	Organic solvent(s) volume	Analysis	PF/LOD	Ref.
				MeCN	1400 µL (MeCN, PP)			
Opioid peptides	Plasma	In-line SPE	200 µL	MeOH *i*-PrOH	<100 nL (MeOH, elution) <70 µL (*i*-PrOH, SL)	CZE–ESI–IT/MS	10–100/0.1–1 ngmL^{-1}	[110]
				MeCN	1400 µL (MeCN, PP)			
Endormorphins	Plasma	In-line SPE	200 µL	*i*-PrOH MeCN	~20 µL (*i*-PrOH, SL) 1400 µL (MeCN, PP)	CZE–ESI–TOF/MS	100/1 ngmL^{-1}	[114]
Neurotransmitters	Urine	In-line SPE	<1 µL	-	-	CZE–UV	Up to 462/3.7–4.3 ngmL^{-1}	[118]
Caffeine	Urine	In-line SPE	<1 µL	-	-	CZE–UV	1,500–1,900/0.5–0.7 ngmL^{-1}	[119]
Escitalopram	Urine	In-line SPE	<2.25 µL	MeCN MeOH	n.d. (MeCN, BGE and elution) ~25 µL (MeOH, SL)	CZE–ESI–TOF/MS	10 ngmL^{-1}	[140]
Sulfonamides	Urine, serum	On-line SPE	1 mL	THF MeCN	600 µL (THF, elution) 200 µL (MeCN, PP)	CZE–UV	0.05–0.1 µgmL^{-1} (urine) 0.05–0.3 µgmL^{-1} (serum)	[100]
NSAIDs	Urine, serum	On-line SPE	1 mL	MeCN	<1 mL (PP and elution)	CZE–UV	0.05–0.1 µgmL^{-1} (urine) 0.1–1 µgmL^{-1} (serum)	[101]
Tricyclic antidepressants	Urine, serum	On-line SPE	0.5 mL	MeOH MeCN	<1 mL (elution, wash, BGE composition)	NACE–UV	40–80 ngmL^{-1} (urine) 60–100 ngmL^{-1} (serum)	[102]
Endogenous biomarkers	Urine	On-line SPE	2.5 mL	MeCN MeOH	<1 mL (elution, wash, BGE composition)	CZE–UV	0.14–4.50 µgmL^{-1}	[141]
3-Nitrotyrosine	Rat urine	In-line SPE	<200 µL	MeCN MeOH	n.d.	CZE–UV	100/4.4 µmolL^{-1}	[142]
Triazine herbicides	Urine	In-line SPE	1 mL	MeCN MeOH	~30 nL (MeCN, elution) ~15 µL (MeOH, rinse)	CZE–UV	0.2–0.6 µgmL^{-1}	[115]

n.d., not defined

LOD is determined at a signal-to-noise ratio of 3

Fiber in-tube SPME has been used for analysis of four tricyclic antidepressant drugs (TCAs) in urine [87]. A 10-mm-long Zylon fiber filling a capillary placed inside a 0.25-mm i.d. Teflon tube was connected on-line to the CE system. After continuous pumping of the sample, TCAs were desorbed with a few microliters of MeCN, directly transferred to a cross connector, and separated by CE, leading to 100-fold greater sensitivity.

Feng and co-workers used monolith capillary in-tube SPME with poly(methacrylic acid–ethylene glycol dimethacrylate) for extraction of amphetamines [88], opiates [89], ephedrine and pseudoephedrine [90], and angiotensin II receptor antagonists [91] from urine and plasma samples. Some of these applications were performed with an adapted device composed of a regular plastic syringe and a monolithic capillary connected by a pinhead (polymer monolith microextraction, PMME) [89, 91]. In monolith capillary in-tube SPME, a single piece of monolith with a double-pore structure enables use of high flow rates with a low generated pressure through the capillary, leading to high throughput [84, 86]. An alternative approach is the use of continuous bed RAM in-tube SPME, which enabled simultaneous protein separation from the matrix while directly extracting target analytes [92].

Microextraction by packed sorbent (MEPS)

MEPS was developed in 1993 and consists of a 100 to 250-μL syringe containing 1 to 4 mg of packed sorbent (inserted into the barrel of the syringe as a plug or between the barrel and needle as a cartridge). The sorbents are miniaturized to work with microliter bed volumes, enabling use of sample and elution volumes as low as 10 μL. All the commercially available SPE sorbents, including RAM and MIP, can be used in MEPS [17, 77, 93, 94].

Recently, Morales-Cid et al. used at-line and on-line coupled MEPS with CE–MS for determination of anesthetic drugs in plasma [95] and fluoroquinolones in urine [96]. In the first study, MEPS was performed with a 200-μL syringe containing 4 mg C_{18} packing. A microdialysis probe was connected to the needle of the MEPS syringe and the method was fully automated. Using 200 μL plasma and non-aqueous CE (NACE) analysis coupled with MS, LODs as low as 10 $ng mL^{-1}$ were reported for the free anesthetic drugs [95].

In the second study, the extraction step was directly integrated into a commercial CE system. The barrel insert and needle containing 4 mg C_{18} packing were fitted to the outlet position of the CE–MS cartridge and connected to a Teflon tube inside the cartridge working as a reservoir (300 μL) for conditioning, preconcentration, and elution. Using CE equipment pressures, samples were preconcentrated and extracted on-line before separation. Only 48 μL urine and 140 μL MeOH were required for conditioning and elution. The eluates were analyzed by NACE-MS to increase resolution and sensitivity. With this configuration, absolute recovery from urine ranged from 70 to 109 % with LODs of less than 10 $ng mL^{-1}$ [96].

Stir-bar-sorptive extraction (SBSE)

Based on the same extraction principle as SPME, SBSE was first developed in 1999 by Baltussen et al. to overcome the limited amount of extraction sorbent used in SPME [97, 98]. In SBSE, the extraction sorptive phase is coated (0.5 to 1-mm layer) on to magnetic stir bars (1 to 4 cm in length) composed of a magnetic rod surrounded by a glass jacket. During stirring of the aqueous sample (typically 30 to 240 min), analytes are extracted in accordance with their partition coefficients. Desorption can be performed thermally or by liquid desorption by organic solvent back-extraction.

There have been few applications of SBSE in combination with CE. Do Rosario et al. developed an SBSE–MEKC method for determination of polynuclear aromatic hydrocarbons (PAHs) in fish bile [99], but no applications in human bioanalysis have been found. Nonetheless, SBSE could be used for extraction of urine samples, because of the relatively large volumes of urine available and the long detection times required to achieve very low LODs of metabolites. However, commercial coated stir bars (Twisters; Gerstel, Mühlheim, Germany) are still limited to PDMS and PDMS–ethylene glycol phases, which are better suited to extraction of non-polar compounds.

SPE-CE

Off-line SPE is largely used in combination with CE, because of its ease of implementation. Over the past two decades, new setups have been developed to automate this process, increase sample throughput, and reduce solvent consumption. At-line coupling of SPE with CE is performed with a robotic arm interface or a modification of the replenishment system. Despite increased throughput, the same solvent quantities are used for sample preparation. More advantageous techniques are on-line and in-line SPE-CE, in which the liquid stream is shared between SPE and CE and analysis can therefore be achieved with relatively small volumes of organic solvents.

On-line SPE-CE

In on-line SPE-CE, an interface (vial, valve, or T-piece type) is used to directly connect the stream from the SPE part and the CE capillary. Because the SPE process is performed independently of CE analysis, no adsorption of the matrix components on to the capillary wall is observed, nor any perturbation of the electrophoretic process. However, peak broadening can be

observed because the desorption volume generally larger than the CE injection volume.

Veraart et al. developed on-line dialysis SPE–CE for analysis of sulfonamides [100] and NSAIDs [101] in urine and serum. The system comprised a dialysis unit, four switching valves, four high-pressure pumps, and a polymer-based SPE column. When dialysis SPE had been performed, a signal was sent to the CE system to transfer the analytes that could be analyzed. A THF–water mixture could be used for analyte desorption to avoid bubble formation [100] or a MeCN–water mixture could be used to ensure a good stacking effect during injection [101]. For serum analysis, a PP with MeCN and decanoic acid was first performed to disrupt drug–protein bonding. LODs in the ng mL^{-1} range were reported for urine samples analyzed by CE–UV. This dialysis set-up, with NACE, was also used for analysis of tricyclic antidepressants in urine and serum [102].

More recently, de Jong and co-workers proposed use of on-line SPE-CE with ion-trap (IT) MS detection for analysis of peptides in cerebrospinal fluid [103]. Enkephalin peptides were extracted on C_{18} sorbent from diluted cerebrospinal fluid and introduced into the CE system via a valve interface. Less than 2 μL MeCN was necessary for analyte desorption. The sensitivity was 1,000-fold better than that obtained by conventional CE–MS. This technique was applied to a tryptic digest of cytochrome *c*, and LODs were as low as 20 nmol L^{-1}, indicating the potential for proteomics. An alternative on-line SPE–CE–UV procedure was proposed for analysis of antibiotics (cephalosporins) in cow plasma with a T-split interface [104]. Part of the SPE eluate was injected and the rest of the sample was flushed to waste (split ratio 1:40). Before SPE–CE, PP with 10 % perchloric acid was performed for plasma samples, avoiding the use of organic solvent, which would reduce the breakthrough volume on the C_{18} SPE column or increase the total analysis time, because of evaporation and reconstitution. With these conditions, LODs were in the 50–100-ng mL^{-1} range, similar to those reported for other LC–UV methods.

In-line SPE-CE

In in-line SPE-CE, the SPE material is part of the CE capillary, and the potential is applied on the entire system during separation using either an open tubular capillary coated with SPE sorbent, a packed-bed sorbent retained with frits, silica- or polymer-based monoliths, or an impregnated membrane. Recently, carbon nanotubes, magnetic particles, or antibodies for immunoaffinity recognition have been successfully investigated. The overall SPE eluate is analyzed by CE, resulting in good recovery. Nevertheless, the latter greatly depends on the nature and volume of the elution solvent. Furthermore, because of direct transfer of the extraction eluate, adsorption of matrix components on to the capillary wall can affect the separation or clog the capillary [105–109].

Sanz-Nebot, Barbosa, and co-workers developed several applications of CE–ESI–MS with in-line SPE microcartridges [110–114]. In their homemade set-up, a CE capillary is cut into two pieces to enable insertion of an SPE microcartridge. Its body is coupled to both parts with a 0.5-cm polyethylene sleeve and equipped with 0.1-cm polyethylene frits after sorbent filling. The tight junction obtained means no adhesive sealing is necessary, and the modified capillary is fitted into commercial CE cartridges. This approach has been successfully applied to the analysis of opioid peptides [110, 112, 113] and neuropeptides [111] in plasma samples with C_{18} or other sorbents [110]. In a recent study, use of an immunoaffinity sorbent for the analysis of endormorphins in plasma by in-line SPE–CE–ESI-MS was also evaluated [114]. In this case, the previously developed microcartridge contained the immunoaffinity sorbent consisting of anti-endorphin antibodies that were covalently attached to activated hydrazide silica particles via carbohydrate groups. Immunoaffinity sorbents resulted in improved selectivity and extraction efficiency with a larger introduced sample volume. LODs as low as 1 ng mL^{-1} in standard solutions were achieved with a 100-fold PF compared with CE–MS, and LODs as low as 100 ng mL^{-1} were achieved for plasma after PP and filtration. However, some cross-reactivity against dynorphin, because of non-specific binding, was also observed.

MIPs, also, are regarded as highly selective synthetic materials with recognition sites that can specifically bind target analytes. Molecularly imprinted solid-phase extraction (MISPE) has been evaluated as an in-line SPE-CE technique for monitoring of triazine herbicides in urine, and compared with use of HLB sorbent [115]. MIPs have several advantages, for example physical robustness, rigidity, resistance to elevated temperature or pressure, and inertness toward organic solvents. The concentrator was constructed from a 2-mm capillary filled with MIP sorbent (particle size 55 μm) by use of a vacuum pump and then introduced into a 1.5-cm piece of PTFE tubing that fitted the outer diameter of the capillary. No frits were necessary to retain the sorbent. The results obtained for MIPs were superior to those for HLB sorbent.

Finally, during the last five years, increasing attention has been paid to the use of monoliths as sorbent in in-line SPE-CE. Monoliths are rapidly synthesized in one step and are characterized by low backpressure and chemical stability over a wide range of pH. Silica-based (prepared by use of sol–gel technology) and polymer-based (prepared by in-situ polymerization of monomers and cross-linkers) monoliths can be easily fixed at the end of a capillary by chemical modification [105, 116]. Several in-line SPE–CE applications with a variety of monolith materials have recently been proposed for analysis of, for example, methionine enkephalin in deproteinated cerebrospinal fluid [117], neurotransmitters (e.g., dopamine, adrenaline, histamine, and serotonin) in urine [118], or caffeine in urine [119].

Conclusions and future trends

Sample preparation is recognized as the most critical step in bioanalysis if good accuracy, selectivity, sensitivity, and robustness are to be achieved. Over the past few decades significant efforts have been devoted to reducing time, cost, manual handling, and consumption of solvents and samples. MEs have been shown to be very attractive compared with conventional LLE or SPE, and numerous innovative developments in respect of the liquid phase or miniaturized solid devices have been proposed. The combination of miniaturized sample preparation with CE has significant potential in bioanalysis, with only a few microliters of solvent required for the entire analytical process. Several MEs with CE analysis have been emphasized in this review, with a variety of bioapplications. A suitable approach should be selected considering the physicochemical properties of the analyte, the nature and volume of the biological matrix, the concentration range of the targeted analyte(s), the selectivity and sensitivity required, and the possibility of at-line, on-line and in-line automation.

Future developments will, hopefully, enable CE analysis to be used with the most recent sample pretreatments, which have already attracted attention in combination with LC or GC. As examples, disposable pipette extraction (DPX), in which a loose SPE sorbent is placed inside a pipette tip [120], was first proposed in 2008 and has already been successfully applied to a variety of applications with LC or GC analysis and is also fully adapted to CE analysis. In vivo SPME, in which sample preparation encompasses less-invasive sampling with direct exposure to human or animal living systems, could be of great interest in combination with CE for pre-clinical studies or clinical purposes [121]. Dried-blood spot sampling (DBS) has been shown to be not only a biofluid support but also a sample pretreatment with use of a small amount of solvents, inducing "on support" PP and selective desorption [122–124]. Moreover, recent microextraction techniques (e.g., SDME, HS-SDME, DLLME, and HF-LPME) substituting organic extraction solvents with non-toxic ionic liquids [125, 126] or natural oils [127] combined with CE could lead to powerful and solvent-free analytical procedures.

Surprisingly, especially in combination with liquid-based MEs, very few applications have revealed the potential of CE hyphenation with MS detection to substantially increase both sensitivity and selectivity. However, CE–MS is now easily implemented with dedicated interfaces, either with addition of a sheath liquid or in the sheathless configuration, and should undoubtedly be considered in combination with MEs for all bioanalytical applications to achieve the desired sensitivity (sub-ngmL^{-1} range) and provide the possibility of compound identification.

References

1. Hyotylainen T (2009) Anal Bioanal Chem 394(3):743–758
2. Ramos L (2012) J Chromatogr A 1221:84–98
3. Anastas P, Eghbali N (2010) Chem Soc Rev 39(1):301–312
4. Anastas PT, Kirchhoff MM (2002) Acc Chem Res 35(9):686–694
5. Urbanowicz M, Zabiegala B, Namiesnik J (2011) Anal Bioanal Chem 399(1):277–300
6. Nerin C, Salafranca J, Aznar M, Batlle R (2009) Anal Bioanal Chem 393(3):809–833
7. Tobiszewski M, Mechlinska A, Zygmunt B, Namiesnik J (2009) Trends Anal Chem 28(8):943
8. Tobiszewski M, Mechlinska A, Namiesnik J (2010) Chem Soc Rev 39(8):2869–2878
9. Pawliszyn J, Pedersen-Bjergaard S (2006) J Chromatogr Sci 44 (6):291–307
10. Novakova L, Vlckova H (2009) Anal Chim Acta 656(1–2):8–35
11. Lambropoulou DA, Albanis TA (2007) J Biochem Biophys Methods 70(2):195–228
12. Aufartova J, Mahugo-Santana C, Sosa-Ferrera Z, Santana-Rodriguez JJ, Novakova L, Solich P (2011) Anal Chim Acta 704(1–2):33–46
13. Pico Y, Fernandez M, Ruiz MJ, Font G (2007) J Biochem Biophys Methods 70(2):117–131
14. Ridgway K, Lalljie SP, Smith RM (2007) J Chromatogr A 1153(1–2):36–53
15. Asensio-Ramos M, Ravelo-Perez LM, Gonzalez-Curbelo MA, Hernandez-Borges J (2011) J Chromatogr A 1218(42):7415–7437
16. Kole PL, Venkatesh G, Kotecha J, Sheshala R (2011) Biomed Chromatogr 25(1–2):199–217
17. Ashri NY, Abdel-Rehim M (2011) Bioanalysis 3(17):2003–2018
18. Samanidou V, Kovatsi L, Fragou D, Rentifis K (2011) Bioanalysis 3(17):2019–2046
19. Kataoka H (2010) Anal Bioanal Chem 396(1):339–364
20. Badoud F, Guillarme D, Boccard J, Grata E, Saugy M, Rudaz S, Veuthey JL (2011) Forensic Sci Int 213(1–3):49–61
21. Lucena R, Cruz-Vera M, Cardenas S, Valcarcel M (2009) Bioanalysis 1(1):135–149
22. Jeannot MA, Cantwell FF (1996) Anal Chem 68(13):2236–2240
23. Liu H, Dasgupta PK (1996) Anal Chem 68(11):1817–1821
24. Jeannot MA, Cantwell FF (1997) Anal Chem 69:235–239
25. He Y, Lee HK (1997) Anal Chem 69:4634–4640
26. Xu L, Basheer C, Lee HK (2007) J Chromatogr A 1152(1–2):184–192
27. Ma M, Cantwell FF (1998) Anal Chem 70(3912–3919):3912
28. Fang H, Zeng Z, Liu L, Pang D (2006) Anal Chem 78(4):1257–1263
29. Choi K, Kim J, Jang YO, Chung DS (2009) Electrophoresis 30 (16):2905–2911
30. Choi J, Choi K, Kim J, Ahmed AY, Al-Othman ZA, Chung DS (2011) J Chromatogr A 1218(41):7227–7233
31. Fang H, Zeng Z, Liu L (2006) Anal Chem 78(17):6043–6049
32. Fang H, Yang F, Sun J, Tian Y, Zeng Z, Xu Y (2011) Talanta 85 (4):2148–2153
33. Yangcheng L, Quan L, Guangsheng L, Youyuan D (2006) Ana Chim Acta 566:25–264
34. Gao W, Chen G, Chen T, Zhang X, Chen Y, Hu Z (2011) Talanta 83(5):1673–1679
35. Theis AL, Waldack AJ, Hansen SM, Jeannot MA (2001) Anal Chem 73(23):5651–5654
36. Pena-Pereira F, Lavilla I, Bendicho C (2010) Trends Anal Chem 29(7):617–628
37. Jermak S, Pranaityte B, Padarauskas A (2006) Electrophoresis 27 (22):4538–4544

38. Stege PW, Lapierre AV, Martinez LD, Messina GA, Sombra LL (2011) Talanta 86:278–283
39. Rezaee M, Assadi Y, Milani Hosseini MR, Aghaee E, Ahmadi F, Berijani S (2006) J Chromatogr A 1116(1–2):1–9
40. Kohler I, Schappler J, Sierro T, Rudaz S (2012) J Pharm Biomed Anal. doi:10.1016/j.jpba.2012.03.036
41. Regueiro J, Llompart M, Garcia-Jares C, Garcia-Monteagudo JC, Cela R (2008) J Chromatogr A 1190(1–2):27–38
42. Luque de Castro MD, Priego-Capote F (2007) Talanta 72(2):321–334
43. Huang H, Chen Z, Yan X (2012) J Sep Sci 35(3):436–444
44. Pedersen-Bjergaard S, Rasmussen KE (2008) J Chromatogr A 1184(1–2):132–142
45. Thordarson E, Palmarsdottir S, Mathiasson L, Jonsson JA (1996) Anal Chem 68(15):2559–2563
46. Palmarsdottir S, Thordarson E, Edholm LE, Jonsson JA, Mathiasson L (1997) Anal Chem 69(9):1732–1737
47. Nozal L, Arce L, Simonet BM, Rios A, Valcarcel M (2006) Electrophoresis 27(15):3075–3085
48. Kuban P, Bocek P (2012) J Chromatogr A 1234:2–8
49. Pedersen-Bjergaard S, Rasmussen KE (1999) Anal Chem 71 (14):2650–2656
50. Rasmussen KE, Pedersen-Bjergaard S, Krogh M, Ugland HG, Gronhaug T (2000) J Chromatogr A 873(1):3–11
51. Halvorsen TG, Pedersen-Bjergaard S, Rasmussen KE (2001) J Chromatogr B 760:219–226
52. Pedersen-Bjergaard S, Rasmussen KE (2000) Electrophoresis 21 (3):579–585
53. Andersen S, Halvorsen TG, Pedersen-Bjergaard S, Rasmussen KE, Tanum L, Refsum H (2003) J Pharm Biomed Anal 33(2):263–273
54. Halvorsen TG, Pedersen-Bjergaard S, Rasmussen KE (2001) J Chromatogr A 909(1):87–93
55. Ho TS, Pedersen-Bjergaard S, Rasmussen KE (2002) Analyst 127(5):608–613
56. Bjorhovde A, Halvorsen TG, Rasmussen KE, Pedersen-Bjergaard S (2003) Anal Chim Acta 491:155–161
57. Li P, Duan J, Hu B (2008) Electrophoresis 29(14):3081–3089
58. Li P, Zhang X, Hu B (2011) J Chromatogr A 1218(52):9414–9421
59. Ho TS, Halvorsen TG, Pedersen-Bjergaard S, Rasmussen KE (2003) J Chromatogr A 998(1–2):61–72
60. Ho TS, Pedersen-Bjergaard S, Rasmussen KE (2006) J Chromatogr Sci 44(6):308–316
61. Jiang X, Basheer C, Zhang J, Lee HK (2005) J Chromatogr A 1087(1–2):289–294
62. Pedersen-Bjergaard S, Rasmussen KE (2006) J Chromatogr A 1109(2):183–190
63. Petersen NJ, Rasmussen KE, Pedersen-Bjergaard S, Gjelstad A (2011) Anal Sci 27(10):965–972
64. Gjelstad A, Rasmussen KE, Pedersen-Bjergaard S (2009) Anal Bioanal Chem 393(3):921–928
65. Kjelsen IJ, Gjelstad A, Rasmussen KE, Pedersen-Bjergaard S (2008) J Chromatogr A 1180(1–2):1–9
66. Balchen M, Halvorsen TG, Reubsaet L, Pedersen-Bjergaard S (2009) J Chromatogr A 1216(41):6900–6905
67. Strieglerova L, Kuban P, Bocek P (2011) J Chromatogr A 1218 (37):6248–6255
68. Strieglerova L, Kuban P, Bocek P (2011) Electrophoresis 32 (10):1182–1189
69. Nojavan S, Fakhari AR (2010) J Sep Sci 33(20):3231–3238
70. Fakhari AR, Tabani H, Nojavan S, Abedi H (2012) Electrophoresis 33(3):506–515
71. Petersen NJ, Jensen H, Hansen SH, Rasmussen KE, Pedersen-Bjergaard S (2009) J Chromatogr A 1216(9):1496–1502
72. Sikanen T, Pedersen-Bjergaard S, Jensen H, Kostiainen R, Rasmussen KE, Kotiaho T (2010) Anal Chim Acta 658(2):133–140
73. Nordman N, Sikanen T, Moilanen ME, Aura S, Kotiaho T, Franssila S, Kostiainen R (2011) J Chromatogr A 1218(5):739–745
74. Arthur CL, Pawliszyn J (1990) Anal Chem 62(19):2145–2148
75. Baltussen E, Cramers CA, Sandra PJ (2002) Anal Bioanal Chem 373(1–2):3–22
76. Theodoridis G, Koster EH, de Jong GJ (2000) J Chromatogr B: Biomed Sci Appl 745(1):49–82
77. Kataoka H (2005) Curr Pharm Anal 1:65–84
78. Zhang Z, Pawliszyn J (1993) Anal Chem 65:1843–1852
79. Jinno K, Han Y, Sawada H, Taniguchi M (1997) Chromatographia 46(5/6):309–314
80. Jinno K, Sawada H, Han Y (1998) Biomed Chromatogr 12 (3):126–127
81. Snow NH (2000) J Chromatogr A 885(1–2):445–455
82. Zhou X, Li X, Zeng Z (2006) J Chromatogr A 1104(1–2):359–365
83. Fang H, Liu M, Zeng Z (2006) Talanta 68(3):979–986
84. Kataoka H, Saito K (2011) J Pharm Biomed Anal 54(5):926–950
85. Eisert R, Pawliszyn J (1997) Anal Chem 69:3140–3147
86. Kataoka H, Ishizaki A, Nonaka Y, Saito K (2009) Anal Chim Acta 655(1–2):8–29
87. Jinno K, Kawazoe M, Saito Y, Takeichi T, Hayashida M (2001) Electrophoresis 22(17):3785–3790
88. Wei F, Fan Y, Zhang M, Feng YQ (2005) Electrophoresis 26 (16):3141–3150
89. Wei F, Zhang M, Feng YQ (2006) Electrophoresis 27(10):1939–1948
90. Wei F, Zhang M, Feng YQ (2007) J Chromatogr B 850(1–2):38–44
91. Zhang M, Wei F, Zhang YF, Nie J, Feng YQ (2006) J Chromatogr A 1102(1–2):294–301
92. Jarmalaviciene R, Szumski M, Kornysova O, Klodzinska E, Westerlund D, Krawczyk S, Mickevicius D, Buszewski B, Maruska A (2008) Electrophoresis 29(8):1753–1760
93. Abdel-Rehim M (2010) J Chromatogr A 1217(16):2569–2580
94. Abdel-Rehim M (2011) Anal Chim Acta 701(2):119–128
95. Morales-Cid G, Cardenas S, Simonet BM, Valcarcel M (2009) Electrophoresis 30(10):1684–1691
96. Morales-Cid G, Cardenas S, Simonet BM, Valcarcel M (2009) Anal Chem 81(8):3188–3193
97. Baltussen E, Sandra P, David F, Cramers CA (1999) J Microcol Sep 11(10):737–747
98. David F, Sandra P (2007) J Chromatogr A 1152(1–2):54–69
99. do Rosario PM, Nogueira JM (2006) Electrophoresis 27 (23):4694–4702
100. Veraart JR, van Hekezen J, Groot MC, Gooijer C, Lingeman H, Velthorst NH, Brinkman UA (1998) Electrophoresis 19(16–17):2944–2949
101. Veraart JR, Groot MC, Gooijer C, Lingeman H, Velthorst NH, Brinkman UA (1999) Analyst 124(2):115–118
102. Veraart JR, Brinkman UA (2001) J Chromatogr A 922(1–2):339–346
103. Tempels FW, Underberg WJ, Somsen GW, de Jong GJ (2007) Electrophoresis 28(9):1319–1326
104. Puig P, Tempels FW, Borrull F, Calull M, Aguilar C, Somsen GW, de Jong GJ (2007) J Chromatogr B 856(1–2):365–370
105. Puig P, Borrull F, Calull M, Aguilar C (2008) Anal Chim Acta 616(1):1–18
106. Ramautar R, Jong GJ, Somsen GW (2012) Electrophoresis 33 (1):243–250
107. Ramautar R, Somsen GW, de Jong GJ (2010) Electrophoresis 31 (1):44–54
108. Tempels FW, Underberg WJ, Somsen GW, de Jong GJ (2008) Electrophoresis 29(1):108–128
109. Saavedra L, Barbas C (2007) J Biochem Biophys Methods 70 (2):289–297
110. Benavente F, Medina-Casanellas S, Barbosa J, Sanz-Nebot V (2010) J Sep Sci 33(9):1294–1304

111. Hernandez E, Benavente F, Sanz-Nebot V, Barbosa J (2008) Electrophoresis 29(16):3366–3376
112. Hernandez E, Benavente F, Sanz-Nebot V, Barbosa J (2007) Electrophoresis 28(21):3957–3965
113. Medina-Casanellas S, Benavente F, Barbosa J, Sanz-Nebot V (2011) Electrophoresis 32(13):1750–1759
114. Medina-Casanellas S, Benavente F, Barbosa J, Sanz-Nebot V (2012) Anal Chim Acta 717:134–142
115. Lara FJ, Lynen F, Sandra P, Garcia-Campana AM, Ales-Barrero F (2008) Electrophoresis 29(18):3834–3841
116. Namera A, Nakamoto A, Saito T, Miyazaki S (2011) J Sep Sci 34 (8):901–924
117. Ramautar R, Ratnayake CK, Somsen GW, de Jong GJ (2009) Talanta 78(2):638–642
118. Thabano JR, Breadmore MC, Hutchinson JP, Johns C, Haddad PR (2007) J Chromatogr A 1175(1):117–126
119. Thabano JR, Breadmore MC, Hutchinson JP, Johns C, Haddad PR (2009) J Chromatogr A 1216(25):4933–4940
120. Ellison ST, Brewer WE, Morgan SL (2009) J Anal Toxicol 33 (7):356–365
121. Ouyang G, Vuckovic D, Pawliszyn J (2011) Chem Rev 111 (4):2784–2814
122. Deglon J, Thomas A, Daali Y, Lauer E, Samer C, Desmeules J, Dayer P, Mangin P, Staub C (2011) J Pharm Biomed Anal 54 (2):359–367
123. Deglon J, Thomas A, Mangin P, Staub C (2012) Anal Bioanal Chem 402(8):2485–2498
124. Thomas A, Deglon J, Steimer T, Mangin P, Daali Y, Staub C (2010) J Sep Sci 33(6–7):873–879
125. Poole CF, Poole SK (2010) J Chromatogr A 1217(16):2268–2286
126. Liu R, Liu JF, Yin YG, Hu XL, Jiang GB (2009) Anal Bioanal Chem 393(3):871–883
127. Pedersen-Bjergaard S, Rasmussen KE (2004) J Sep Sci 27(17–18):1511–1516
128. Gao W, Chen G, Chen Y, Li N, Chen T, Hu Z (2011) J Chromatogr A 1218(33):5712–5717
129. Nozal L, Arce L, Simonet BM, Rios A, Valcarcel M (2007) Electrophoresis 28(18):3284–3289
130. Palmarsdottir S, Mathiasson L, Jonsson JA, Edholm LE (1997) J Chromatogr B: Biomed Sci Appl 688(1):127–134
131. Andersen S, Halvorsen TG, Pedersen-Bjergaard S, Rasmussen KE (2002) J Chromatogr A 963(1–2):303–312
132. Al Azzam KM, Makahleah A, Saad B, Mansor SM (2010) J Chromatogr A 1217(23):3654–3659
133. Lin SC, Whang CW (2008) J Sep Sci 31(22):3921–3929
134. Meng L, Liu X, Wang B, Shen G, Wang Z, Guo M (2009) J Chromatogr B 877(29):3645–3651
135. Slampova A, Kuban P, Bocek P (2012) J Chromatogr A 1234:32–37
136. Deng DL, Zhang JY, Chen C, Hou XL, Su YY, Wu L (2012) J Chromatogr A 1219:195–200
137. Li S, Weber SG (1997) Anal Chem 69:1217–1222
138. Lin B, Zheng MM, Ng SC, Feng YQ (2007) Electrophoresis 28 (15):2771–2780
139. Botello I, Borrull F, Calull M, Aguilar C, Somsen GW, de Jong GJ (2012) Anal Bioanal Chem 403(3):777–784
140. Johannesson N, Bergquist J (2007) J Pharm Biomed Anal 43 (3):1045–1048
141. Ruiz-Jimenez J, Mata-Granados JM, Luque de Castro MD (2007) Electrophoresis 28(5):789–798
142. Saavedra L, Maeso N, Cifuentes A, Barbas C (2007) J Pharm Biomed Anal 44(2):471–476