POLYMER INCLUSION MEMBRANE SAMPLING PROBE FOR ELECTRIC FIELD DRIVEN EXTRACTION OF DOXORUBICIN

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DEDICATION

Specially dedicated to my beloved family members for all support and encouragement in completing this study.

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ABSTRACT

Sample preparation for the analysis of biological fluid samples posed significant challenges to the clinical laboratory. The current sample preparation practice involves multi-steps procedure that are time consuming and may lead to analytes lost. In this study, a low-cost, single-use sampling probe was introduced for electric field driven extraction. The sampling probe consists of a non-conductive glass capillary substrate, and a polymer inclusion membrane (PIM) dip-coated on the glass capillary substrate. The probe features a user-friendly design that can electrically extract targeted analytes from biological fluid sample. The glass capillary, closed at one end, was dipped in a homogeneous membrane solution that consists of an optimum composition of cellulose triacetate (CTA), 2-nitrophenyl octyl ether (2-NPOE) and 1-ethyl-3-methylimidazolium bis(trifluoromethylsulfonyl)imide ([EMIM][NTf_2]), yielded a PIM with thickness of 30 µm on the sampling probe. The effects of dipping cycles (thickness), voltage applied, and sample pH on the extraction efficiency of the sampling probe were thoroughly investigated. The concept of electrokinetic extraction using the PIM sampling probe was first demonstrated on the cationic model analyte, Rhodamine 6G at 500 V for 60 mins. The proven concept was then successfully applied to the extraction of the anticancer drug doxorubicin from samples in liquid or dried form, including dried blood spot (DBS), human plasma and human serum samples. The practicability and reliability of the electrokinetic extraction process were evaluated using liquid chromatography with tandem mass spectrometry (LC-MS/MS) to quantify the desorption of extracted doxorubicin from the PIM sampling probe. Under optimised conditions, the new method provided good linearity over a concentration range of 0.2 to 20 ng/mL; additionally, quantification limits of 0.2 to 2 ng/mL were achieved for the three biological samples and the relative recoveries ranged from 82.7 to 113.8% for DBS, human plasma and human serum samples; good method reproducibility was also achieved, with relative standard deviations (RSDs) ranging from 0.9 and 4.6%. The PIM sampling probe was further integrated into a portable battery-device for safe, lower-voltage (36 V) electrokinetic extraction. The correlation coefficients, r, for the two pairs of data were determined using the laboratory setup and the battery-powered device; values in the range of 0.9926 to 0.9996 were found, indicating an acceptable agreement. This new electrokinetic extraction approach represents a new opportunity for processing samples during sampling and transportation, saving time and reducing manual handling to produce more reliable test results efficiently.

ABSTRAK

Penyediaan sampel untuk analisis sampel biologi menimbulkan cabaran besar kepada makmal klinikal. Amalan penyediaan sampel semasa melibatkan prosedur berbilang langkah yang memakan masa dan boleh menyebabkan kehilangan analit. Dalam kajian ini, *probe* persampelan sekali guna, kos rendah telah diperkenalkan untuk pengekstrakan dipacu medan elektrik. *Probe* persampelan terdiri daripada substrat kapilari kaca bukan konduktif, dan membran rangkum polimer (PIM) bersalut celup pada substrat kapilari kaca. *Probe* ini mempunyai reka bentuk mesra pengguna yang boleh mengekstrak analit disasarkan daripada sampel biologi di bawah aplikasi medan elektrik. Kapilari kaca yang ditutup pada satu hujung telah dicelup dalam larutan membran homogen mengandungi komposisi optimum yang terdiri daripada selulosa triasetat (CTA), 2-nitrofenil oktil eter (2-NPOE) dan 1-etil-3-metilimidazolium bis(trifluorometilsulfonil)imida ([EMIM][NTf_2]) untuk menghasilkan PIM dengan ketebalan 30 µm pada *probe* persampelan. Kesan kitaran pencelupan (ketebalan), voltan yang dikenakan dan pH sampel ke atas kecekapan pengekstrakan *probe* persampelan telah dikaji dengan teliti. Konsep pengekstrakan elektrokinetik menggunakan *probe* persampelan PIM adalah kali pertama ditunjukkan pada analit model kationik, Rhodamine 6G pada 500 V selama 60 minit. Konsep kemudiannya terbukti berjaya digunakan untuk pengekstrakan drug antikanser doksorubisin daripada sampel dalam bentuk cecair atau kering termasuk tompok darah kering (DBS), sampel plasma manusia dan sampel serum manusia. Kebolehamalian dan kebolehpercayaan proses pengekstrakan elektrokinetik telah dinilai menggunakan kromatografi cecair dengan spektrometri jisim tandem (LC-MS/MS) untuk mengukur penyaherapan doksorubisin yang diekstrak daripada *probe* pensampelan PIM. Di bawah keadaan yang dioptimumkan, kaedah baharu memberikan kelinearan yang baik pada julat kepekatan 0.2 hingga 20 ng/mL; di samping itu, had kuantifikasi 0.2 hingga 2 ng/mL telah dicapai untuk tiga sampel biologi dan pemulihan relatif adalah antara 82.7 hingga 113.8% untuk sampel DBS, plasma manusia dan serum manusia; kebolehulangan kaedah yang baik juga dicapai, dengan sisihan piawai relatif (RSD) di antara 0.9 dan 4.6%. *Probe* persampelan PIM disepadukan lagi ke dalam peranti bateri mudah alih untuk pengekstrakan elektrokinetik voltan rendah (36 V) yang selamat. Pekali korelasi, r, untuk dua pasangan data telah ditentukan dengan menggunakan persediaan makmal dan peranti berkuasa bateri; nilai dalam julat 0.9926 hingga 0.9996 didapati, menunjukkan persetujuan yang boleh diterima. Pendekatan pengekstrakan elektrokinetik ini mewakili peluang baharu untuk memproses sampel semasa pensampelan dan pengangkutan, menjimatkan masa dan mengurangkan pengendalian manual untuk menghasilkan keputusan ujian yang lebih dipercayai dengan cekap.

TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

LIST OF ABBREVIATIONS

[EMIM][NTf_2]	-	1-ethyl-3-methylimidazolium bis(trifluoromethyllsulfonyl)imide
2-NPOE	-	2-nitrophenyl octyl ether
2-NPPE	-	2-nitrophenyl pentyl ether
6-mMP	-	6-methylmercaptopurine
6-TG	-	6-thioguanine
ACN	-	Acetonitrile
AMPA	-	Aminomethylphosphonic acid
AMTCI	-	4-amino-5-(methylthio)carbonyl imidazole
AMX	-	Amoxicillin
ANT	-	Anthranilic acid
ATR	-	Attenuated total reflectance
ATR-FTIR	-	Attenuated total reflectance-Fourier transform infrared
B2EHP	-	bis-(2-ethylhexyl) phosphate
BET	-	Brunauer-Emmett-Teller
Bio-SPME-IVLP	-	Bio-solid-phase microextraction-in vivo lung perfusion
BJH	-	Barrett-Joyner-Halenda
BLM	-	Bulk liquid membrane
C^4D	-	Capacitively coupled contactless conductivity
CAGR	-	Compound annual growth rate
CAP	-	Cellulose acetate propionate
CBZ	-	Carbamazepine
CE	-	Capillary electrophoresis
CE-C^4D	-	Capillary electrophoresis-capacitively coupled contactless conductivity detection
CE-ICDOF-LED-FLD	-	Capillary electrophoresis with in-column double optical-fiber light emitting diode-induced fluorescence detection
CE-UV	-	Capillary electrophoresis-ultraviolet detection
CTA	-	Cellulose triacetate
CTB	-	Cellulose tributyrate

CV	-	Cyclic voltammetry
CZE	-	Capillary zone electrophoresis
D2EHPA	-	di-(2-ethylhexyl)phosphoric acid
DART	-	Direct analysis in real time
DBS	-	Dried blood spot
DBSA	-	Dried blood spot autosampler
DC	-	Direct current
DCM	-	Dichloromethane
DDSME	-	Drop-to-drop microextraction
DESI	-	Desorption electrospray ionisation
DI	-	Deionised
DI-SDME	-	Direct immersion single drop microextraction
DI-SPME	-	Direct immersion solid-phase microextraction
DLLME	-	Dispersive liquid-liquid microextraction
DMF	-	Digital microfluidic device
DMPK	-	Drug metabolism and pharmacokinetic
DNA	-	Deoxyribonucleic acid
DNNS	-	Dinonylnapthalene sulfonic acid
DOP	-	Dioctyl phthalate
DSC	-	Differential scanning calorimetry
DTX	-	Docetaxel
ED	-	Electrodialysis
EE	-	Electroextraction
EE-SPME	-	Electroenhanced solid-phase microextraction
ELISA	-	Enzyme-linked immunosorbent assay
ELM	-	Emulsion liquid membrane
EM-SPME	-	Electromembrane-surrounded solid-phase microextraction
EME	-	Electromembrane extraction
EME-PIM	-	Electromembrane extraction-polymer inclusion membrane
ESI	-	Electrospray ionisation
FA	-	Formic acid
FDA	-	Food drug administration
FE-SEM	-	Field emission-scanning electron microscopy

FIA-UV-Vis	-	Flow injection analysis-ultraviolet visible
FTA	-	Fast transient analysis
FTIR	-	Fourier transform infrared
GAC	-	Green analytical chemistry
GC	-	Gas chromatography
GC-MS	-	Gas chromatography-mass spectrometry
GLYP	-	Glyphosate
HCl	-	Hydrochloric acid
HF-LPME	-	Hollow fiber-liquid-phase microextraction
HIP	-	Hippuric acid
HPLC	-	High-performance liquid chromatography
HPLC-DAD	-	High-performance liquid chromatography-diode array detector
HPLC-FLD	-	High-performance liquid chromatography-fluorescence detector
HS-SPME	-	Headspace-solid-phase microextraction
HSA	-	Human serum albumin
IBU	-	Ibuprofen
IC	-	Ion chromatography
IM	-	Imatinib
InC.	-	Incorporated
IV	-	Intraveneous
KTP	-	Ketoprofen
LC	-	Liquid chromatography
LC-MS/MS	-	Liquid chromatography tandem mass spectrometry
LED	-	Light emitting diode
LIF	-	Laser-induced fluorescence
LLE	-	Liquid-liquid extraction
LOD	-	Limit of detection
LOQ	-	Limit of quantification
LPME	-	Liquid-phase microextraction
LTG	-	Lamotrigine
m/z	-	Mass-to-charge ratio

MB	-	Methylene blue
MDMA	-	3,4-methylenedioxy-N-methylamphetamine
MeOH	-	Methanol
MG	-	Malachite green
MMS	-	Miniature mass spectrometer
MRM	-	Multiple reaction monitoring
MS	-	Mass spectrometry
mSPE	-	Magnetic solid-phase extraction
NaOH	-	Sodium hydroxide
NAX	-	Naproxen
NIC	-	Nicotinic acid
NSAID	-	Non-steroidal anti-inflammatory drugs
OFAT	-	one-factor-at-a-time
PADs	-	Paper-based analytical devices
PAN	-	Polyaniline
PCL	-	poly ε-caprolactone
PCR	-	Polymerase chain reaction
PDMS	-	Polydimethylsiloxane
PEDOT	-	Poly(3,4-ethylenedioxythiophene)
pH	-	Power of hydrogen
PIM	-	Polymer inclusion membrane
POC	-	Point-of-care
POU	-	Point-of-use
ppb	-	parts per billion
PPy	-	Polypyrrole
Pt	-	Platinum
PTFE	-	Polytetrafluoroethylene
PVC	-	Polyvinyl chloride
PVDF	-	Polyvinylidene fluoride
PVDF-HFP	-	Poly(vinylidene fluoride-hexafluoropropylene)
RNA	-	Ribonucleic acid
RSD	-	Relative standard deviation
RT	-	Room temperature

RTK	-	Rapid test kit
SAL	-	Salicylic acid
SALLE	-	Salting-out liquid-liquid extraction
SCAP	-	Sample card and preparation
SEM	-	Scanning electron microscopy
SERS	-	Surface-enhanced Raman spectroscopy
SIA-HPLC-MS	-	Sequential injection analysis-high performance liquid chromatography-mass spectrometry
SLM	-	Supported liquid membrane
SPE	-	Solid-phase extraction
SPE-LC-MS/MS	-	Solid-phase extraction-liquid chromatography tandem mass spectrometry
SPME	-	Solid-phase microextraction
TBEP	-	Tris(2-butoxyethyl)phosphate
TDM	-	Therapeutic drug monitoring
TEHP	-	Tris(2-ethylhexyl)phosphate
TGA	-	Thermogravimetric analyser
THF	-	Tetrahydrofuran
TIC	-	Total ion chromatogram
TOMATS	-	Trioctylmethylammonium thiosalicylate
TPU	-	Polyurethane
UPLC	-	Ultra-performance liquid chromatography
UV-Vis	-	Ultraviolet-visible
VAMS	-	Volumetric absorptive microsampling
VPA	-	Valproic acid

LIST OF SYMBOLS

°	-	degree
%	-	percent
%T	-	percent transmittance
%v/v	-	%volume per volume
%w/v	-	%weight per volume
°C	-	degree celcius
°C/min	-	degree celcius per minute
cm^{-1}	-	wavenumber
g/L	-	gram per liter
g/mol	-	gram per mole
J/g	-	Joule per gram
K	-	Kelvin
kV	-	kilovolt
L/Hr	-	liter per hour
m^2/g	-	square meter per gram
mAH	-	milliamp hour
mg/L	-	milligram per liter
mg/mL	-	milligram per milliliter
mL/g	-	mililiter per gram
mL/min	-	mililiter per minute
mM	-	milimolar
MΩ-cm	-	megaohm-centimeter
ng/mL	-	nanogram per mililiter
ppm	-	parts per million
rpm	-	revolutions per minute
r^2	-	regression coefficient
x g	-	times gravity
wt% (m/m)	-	percent by weight (mass per mass)
V	-	volt
V/cm	-	volt per centimeter

µA	-	microampere
µg/L	-	microgram per liter
µg/mL	-	microgram per mililiter
µM	-	micromolar

CHAPTER 1

INTRODUCTION

1.1 Background of the Study

Biological fluid samples—such as whole blood, plasma, urine and saliva—generally contain complex matrices which are known to be major obstacles to obtaining an accurate analytical outcome. These matrices, ranging from various inorganic ions and endogenous and exogenous organic molecules to biomacromolecules, are the coexisting impurities that can be adsorbed by the pores of the stationary phase in the analytical columns and adhere to the inner surface of the pipelines. This may lead to a reduction in the lifetime and efficiency of the column as well as to the blockage of the pipeline system (1). Many efforts have been made—in sample collection, sample preparation and instrumental analysis—to improve the existing analytical procedure, remove the coexisting interfering substances, accelerate the procedure, and improve the accuracy of the final analytical outcome. Despite tremendous advances having been made in the development of state-of-the-art analytical instruments for the qualitative and quantitative analysis of analytes in biological fluid samples, sample preparation remains a challenging task for researchers.

Sample preparation is a multi-step procedure that is employed with the purpose of pre-concentration of samples; the process involves the removal of the complex matrices and impurities in order to obtain a higher purity of the target analyte and thus more accurate results. Analytes may be lost by applying inappropriate sample preparation methods. Hence, an appropriate sample preparation method that is able to purify and pre-concentrate target analytes is a fundamental element of reliable sample analysis, particularly for samples with an extremely low concentration of target analytes. Traditional sample preparation methods such as liquid-liquid extraction (LLE) and solid-phase extraction (SPE) are widely employed prior to the analysis of biological fluid samples (2). However, the procedures of LLE and SPE are tedious,

time-consuming and harmful to the environment due to the high volume of solvent consumption. Miniaturized versions of the above sample preparation methods—namely, solid-phase microextraction (SPME), liquid-phase microextraction (LPME) and hollow fiber-liquid-phase microextraction (HF-LPME)—were introduced, offering such advantages as reduced analysis time, low solvent consumption, high extraction efficiency and environmental friendliness (3, 4).

The majority of the miniaturized sample preparation techniques—including SPME, LPME and liquid membrane extraction—are based on passive diffusion, which results in poor efficiency of extraction and unsatisfactory selectivity, particularly for the extraction of polar compounds and charged molecules. Traditionally, derivatisation, alkalization or acidification have been employed to decrease the polarity of the polar compounds in order to improve the partition coefficients of the analytes, hence, the efficiency of extraction (5). However, the derivatisation steps involve expensive and toxic reagents, while some analytes are unstable in extremely acidic or alkali conditions. Therefore, auxiliary energy—including acoustic wave, microwave, thermal and electrical energy—is introduced to the sample preparation procedure to promote energy exchange and the mass transfer of molecules, thus accelerating the entire extraction process, reducing the operation time and improving the efficiency (6). Among these field-assisted techniques, the appeal of electric field driven sample preparation has increased significantly in recent years due to the considerable improvement in the efficiency of extraction that it brings (7).

Electromembrane extraction (EME) is a novel LPME technique that was developed in 2006 by Pedersen-Bjergaard and Rasmussen (8, 9). EME has demonstrated that analytes can be electrokinetically isolated across a supported liquid membrane (SLM). The SLM is a porous hydrophobic supported membrane with a microliter volume of water-immiscible organic solvent immobilised in the pores. Notably, target analytes are ionised and extracted from the sample (donor solution) through an SLM, to an acceptor solution, in the presence of an electric field. In EME, the direct current (DC) electric potential across the SLM is the driving force for electrokinetic migration (10). Moreover, the electric potential controls the selectivity of the extraction as ions move based on the direction of the electric field. For the

extraction of cations, the cathode is placed in the acceptor solution, and the cations move towards the cathode trapped in the acceptor solution; for the extraction of anions, meanwhile, the anode is placed in the acceptor solution and the anions move towards the anode (direction of acceptor solution) (2). The EME method has considerable technical and analytical advantages over the conventional sample preparation methods. A comparative study has been made between HF-LPME and EME in the extraction of polar drugs from biological samples (11). The results proved that EME demonstrated a higher pre-concentration factor and a shorter extraction time; thus it was concluded to be a more efficient method than HF-LPME for extracting and pre-concentrating polar drugs from complex matrices (11). The success of EME in sample preparation has attracted considerable attention regarding the application of electric fields in sample preparation techniques.

For instance, electromembrane-surrounded solid-phase microextraction (EM-SPME) is an electrically enhanced solid-phase extraction technique that combines the merits of EME and SPME. In EM-SPME, the conductive SPME fiber acts as one of the electrodes and is located in the lumen of the hollow fiber, together with the SLM and the acceptor solution (12, 13). Under the application of an electric field, the analytes in the sample solution migrate through the SLM into the acceptor solution and are further adsorbed by the SPME fibers in the hollow fiber lumen (14). EM-SPME demonstrated better sample clean-up than the conventional SPME approach since the SLM around the SPME fibers acts as a "first line of defence" filter that prohibits the complex matrices from entering the acceptor solution, hence avoiding the risk of fiber saturation in the analysis of samples with complicated matrices (15, 16).

The polymer inclusion membrane (PIM) was introduced as an alternative to the SLM in the EME system. The PIMs demonstrated higher mechanical stability and improved mechanical robustness in the presence of an electric field (17). The PIM is a self-supporting liquid membrane that consists of a base polymer, a plasticizer and a carrier. The base polymer is the backbone of the PIM and provides mechanical support, while the plasticizer acts as a solvent in the membrane, providing it with flexibility and softness. The carrier in the PIM plays a significant role as it is the extractant that determines the selectivity of the PIM. A carrier, generally an ionic liquid, initiates ion

exchange reactions and forms ion complexes with target ions. Generally, the PIM is prepared by dissolving the base polymer, plasticizer and carrier in a volatile organic solvent, before casting the membrane solution as a thin film following the evaporation of the solvent. To date, a number of studies on the applications of PIMs in the aforementioned EME system have been published, in which the extraction of target analytes from feed solutions to the acceptor solution through the PIM under the application of an electric field has been demonstrated (18-20). These studies have proven the enhanced stability of the PIM over the SLM; hence, PIMs are more preferred in separation technologies nowadays, particularly in the EME system. Due to the presence of carriers and plasticizers, PIMs have good ionic conductivity and are thus effective ion transporters. By employing the concept of electrophoresis, the electrokinetic extraction of small drug analytes from a dried blood spot (DBS) matrix was conducted on a dried PIM in the absence of liquid reagents (21). The analytes were migrated and extracted through the thin film PIM under the application of an electric field, emphasising the potential and suitability of PIMs for electroextraction. The portability of the developed electrokinetic extraction method allowed for in-transit sample preparation, thus reducing the turn-around time for the analysis (21).

In the sample preparation approaches published thus far, PIMs have generally taken the form of thin films (19, 20, 22, 23) or hollow fibers (24, 25). In this study, the PIM was cast on a non-conductive glass capillary substrate using the dip-coating method and was applied in the form of a miniaturized sampling probe for electrokinetic extraction. No acceptor phase was involved and the voltage was directly applied onto the conductive PIM surface. The PIM-based sampling probe simultaneously acted as an electrode and as the extraction phase in the electrokinetic extraction. The extraction efficiency of this developed PIM sampling probe was studied using a charged fluorescent dye molecule, Rhodamine 6G, under the application of an electric field. The PIM sampling probe is potentially capable of extracting, preconcentrating and storing target analytes prior to the sample analysis.

With the rising awareness of the importance of immediate decentralised sample processing to prevent degradation and adulteration of samples, a large amount of research has been conducted on the development of miniaturized and portable

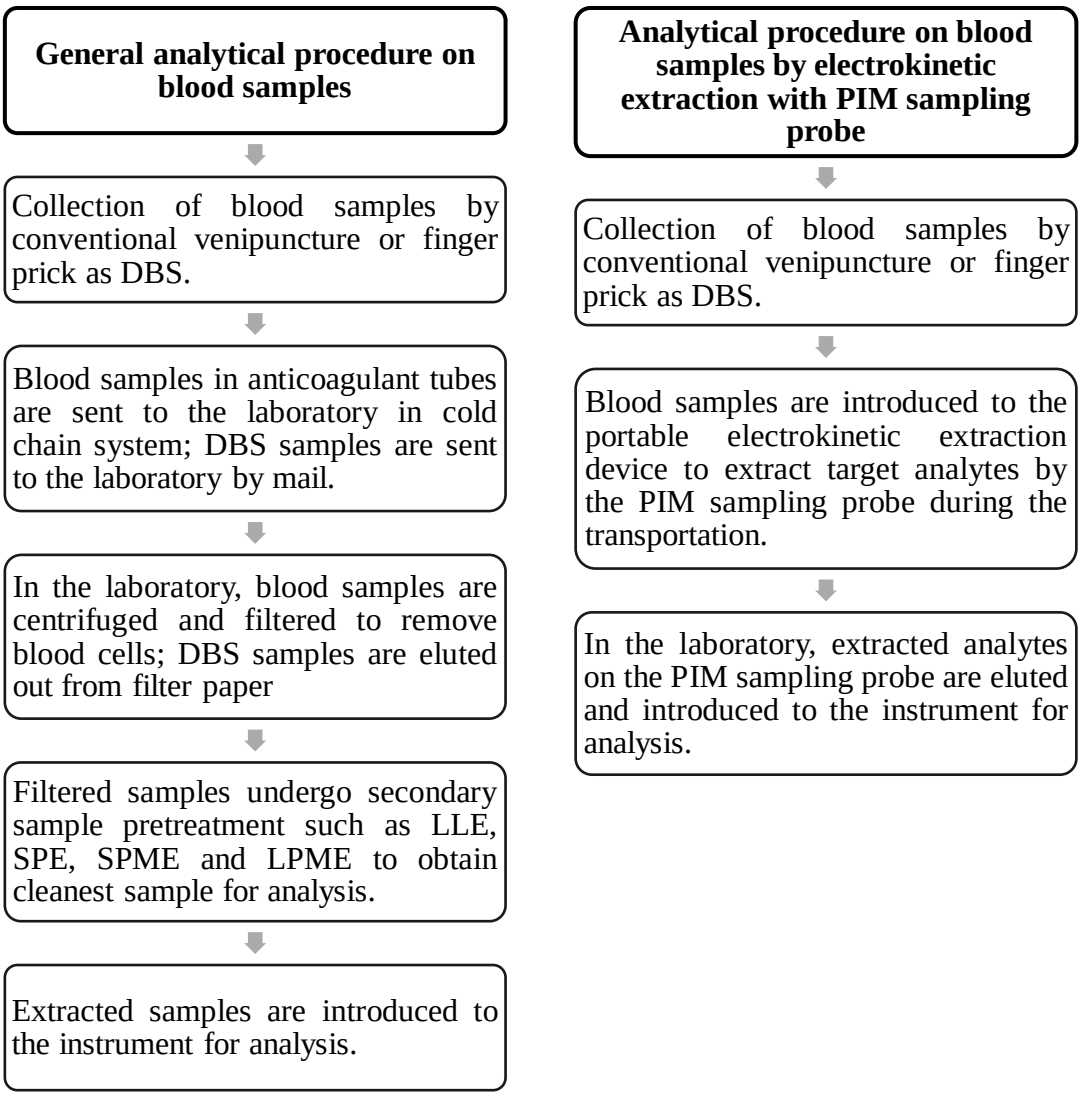
analytical methods. For example, a number of portable and easy-to-use point-of-care (POC) diagnostic tests have been developed for the immediate analysis of patients' samples at the sampling point for disease screening (26).

Patient-centric microsampling techniques such as DBS and volumetric absorptive microsampling (VAMS) (27) are less invasive blood sampling methods that extract only a microliter volume of capillary blood from a finger prick. These approaches allow patients to conduct self-sampling in a familiar and convenient environment (such as the home) thus reducing their frequency of visits to health facilities (28). They have particular benefits for newborn screening, therapeutic drug monitoring (TDM) and disease diagnostics (29). In spite of the advantages and the wide range of applications of microsampling techniques, the majority of the DBS samples are sent by mail and processed in the laboratory. The general sample for preparation of DBS samples comprises sonication, centrifugation, extraction and reconstitution; these processes are required to be conducted in the designated laboratory. To date, the development of a portable sample preparation technique for processing DBS samples during the transportation is hitherto unreported.

In this study, the optimised PIM sampling probe was employed to electrokinetically extract the anticancer drug doxorubicin from DBS, human plasma and human serum samples. Doxorubicin has demonstrated a narrow therapeutic window and serious side effects of cardiotoxicity (30). It is essential that the drug intake is monitored, particularly for childhood cancer survivors treated with doxorubicin, in order to prevent severe cardiac adverse events. In contrast to the existing analytical workflow, the sampling probe allows sample preparation to be conducted during transportation, with an electric field as its driving force. Consequently, it accelerates the entire analytical workflow for TDM and facilitates immediate analytical outcomes for patients. Figure 1.1 shows the comparison between the general analytical workflow and the analytical procedure by using electrokinetic extraction with a PIM sampling probe. PIM sampling probe simplifies the analytical workflow by electrokinetic extraction of analytes during the transportation of samples to the laboratory. To enable the portability of the electrokinetic extraction set up for sample preparation during transportation, the PIM sampling probe was further

integrated into a portable battery-powered electrokinetic extraction device. This study focuses on the design and demonstration of the prototype of the device. A lower voltage was applied to the PIM sampling probe for the electrokinetic extraction. The extracted analytes on the PIM sampling probe were further desorbed and quantified using liquid chromatography with tandem mass spectrometry (LC-MS/MS) in the laboratory in order to obtain more accurate and higher-sensitivity analytical results.

Figure 1.1 Comparison between the general analytical workflow and the analytical procedure by using electrokinetic extraction with PIM sampling probe.



1.2 Problem Statement

Nowadays, the preanalytical stage of sample collection and preparation is a time-consuming procedure, and samples are generally processed and analysed following their arrival in the laboratory. Many test results are inconclusive, sometimes incorrect, due to the fact that the sample was not stabilised for analysis by the central laboratory test. Moreover, there are also significant challenges in terms of the transportation of stabilised samples as the transportation network between the sampling site and many central laboratories are inadequate. The inconvenience of transporting samples from rural, remote areas to the centralised laboratory services may lead to low-quality of analytical and diagnostic services. In addition, traditional methods for the collection of liquid samples in bulk quantities involve high-cost cold chain systems during transportation to preserve the liquid samples for subsequent pretreatment and analysis in the laboratory. Sample preparation steps that employ huge amounts of toxic solvents for extraction are not environmentally friendly and are commonly performed in the laboratory after samples have been received. Patients are required to frequently visit healthcare premises for their routine medical check-ups, which is inconvenient, particularly for the elderly, children and patients who require TDM. Even before the outbreak of the COVID-19 pandemic, the decentralisation and miniaturization of sample preparation procedures have been a topic of great interest to analytical scientists.

The world has had to change and evolve in a variety of ways in order to adapt to the ongoing COVID-19 pandemic. Since the outbreak of the pandemic, there has been a growing trend for the decentralisation and digitalisation of healthcare services since most people have deemed visiting healthcare facilities for routine visits to be a high-risk course of action (31). Therefore, the only way for sample collection and preparation methods to keep up with this trend will be to introduce a simple and user-friendly portable device for patients to be able to closely monitor their conditions during pandemics.

The DBS method is a less-invasive and patient-friendly microsampling technique that can be easily performed by patients at home with adequate training.

Blood samples are collected from a finger prick and dried on a paper substrate for at least three hours before being sent to the laboratory for sample preparation and instrumental analysis. DBS samples are relatively stable; hence they are sent by mail to the laboratory, avoiding the need for a cold-chain system. Nevertheless, the surrounding humidity and moisture content might destroy the DBS samples. DBS require a significant sample preparation procedure to meet satisfactory levels of reliability on analytical results. In the laboratory, DBS samples are desorbed with suitable solvents and are further processed using traditional sample preparation techniques, including protein precipitation and LLE (32, 33). To date, no research has been conducted on the processing of DBS samples during transportation using an electric field, hence the motivation for this study.

A portable battery-powered electrokinetic extraction device using electric field driven sample preparation was developed in this study. The device is designed for the electrokinetic extraction of drug analytes from DBS, human plasma and human serum samples during sample transportation. The core of the device is a miniaturized PIM-based sampling probe, the PIM being fabricated using a simple dip-coating method. The PIM is robust, with a strong mechanical structure, and exhibits excellent performance in adsorbing targeted analytes in sample matrices under an electric field. In addition, the new sampling probe is quick and easy to manufacture and, most importantly, cost-effective, thus benefiting end users in the commercial market.

1.3 Objectives of the Study

This project focuses on the development of a new self-assembly PIM sampling probe for electric field driven extraction. The aims of this proposal are:

(a) To evaluate the capability and performance of a new self-assembly PIM sampling probe—comprising a PIM dip-coated on a non-conductive glass capillary substrate—in an electric field driven extraction.

(b) To determine the optimal PIM composition and operating conditions for the electric field driven extraction of target charged analytes on the PIM sampling probe.

(c) To validate and apply the developed approach as a new method of electrokinetic extraction of doxorubicin from human plasma, human serum and DBS samples.

(d) To investigate the efficiency and feasibility of the PIM sampling probe integrated with a portable battery-powered device for electrokinetic extraction of doxorubicin.

1.4 Scopes of the Research

In this study, a PIM-based sampling probe was developed to electrokinetically extract doxorubicin from DBS, human plasma and human serum samples. The potential of the PIM sampling probe for application in electrokinetic extraction was investigated and monitored using the cationic fluorescent dye, Rhodamine 6G. In this work, the PIM sampling probes were designed, prepared and characterised for electrokinetic extraction. The PIM sampling probes were characterised using field emission-scanning electron microscopy (FE-SEM), nitrogen adsorption/ desorption analysis and attenuated total reflectance-Fourier transform infrared (ATR-FTIR) spectroscopy analysis. The thermal stability and phase transitions of the PIM were studied using thermogravimetric analysis (TGA) and differential scanning calorimetry (DSC) (Objective a).

A series of optimisation studies were performed to determine the composition of the PIM, including the amount of carrier (1-ethyl-3-methylimidazolium bis(trifluoromethylsulfonyl)imide [EMIM][NTf_2]) in the range of 0 to 75 mg, the amount of plasticizer (2-nitrophenyloctyl ether [2-NPOE]) in the range of 0 to 150 mg, the amount of base polymer (cellulose triacetate [CTA]) in the range of 50 to 150 mg, and the thickness of the PIM on the glass capillary (controlled by the number of dipping cycles of the glass capillary in the membrane solution). The electrophoretic parameters, such as voltage applied (0 – 500 V) and pH of the solution (pH 3 – pH 8), were optimised to determine the effectiveness of the extraction. Analytical performances of the developed PIM sampling probe on the extraction of Rhodamine 6G dye were evaluated and visualised using a handheld fluorescence microscope. The

fluorescence intensity of the extracted Rhodamine 6G was quantified using ImageJ software (Objective b).

The performance of the PIM sampling probe was further evaluated by extracting the fluorescent anticancer drug doxorubicin from real biological fluid samples under the application of an electric field. The doxorubicin in DBS, human plasma and human serum samples was electrokinetically extracted using the PIM sampling probe and then desorbed in the suitable desorption solvent for LC-MS/MS analysis. The LC-MS/MS procedure assessed the practicability and reliability of the PIM sampling probe by quantifying the desorption of extracted doxorubicin from the sampling probe. The probe was applied to the electrokinetic extraction of doxorubicin from human plasma, human serum and DBS samples in order to evaluate and validate the analytical performance of the developed method. The method validation parameters include linearity and range, the limit of quantification (LOQ), the limit of detection (LOD), recovery and repeatability (Objective c). Ultimately, the PIM sampling probe was integrated into a portable battery-powered electrokinetic extraction device prototype to improve the portability of the setup, and the effectiveness of electrokinetic extraction of the developed device was evaluated (Objective d).

1.5 Significance of the Study

In this study, the electrokinetic extraction capabilities of a portable miniaturized PIM-based sampling probe were investigated. The developed PIM sampling probe is envisioned to provide significant improvements in sampling efficiency and sample preparation procedures as well as in the transportation of samples in the presence of an electric field. In addition, the presence of this electric field in the extraction process means that liquid samples can be extracted with solvents in microliter volumes, resulting in a more environmentally friendly, cost-effective procedure.

Furthermore, the process of shipping conventional samples is both costly and time-consuming, particularly in some large countries; thus, our developed portable battery-powered electrokinetic extraction device brings significant benefits to the nation. The ability to conduct sample preparation during transportation using this portable device will reduce the incidence of mortality resulting from delayed diagnoses. It will shorten the time required to reach a clinical outcome following sample collection. Since the analytes are extracted and stored in the PIM sampling probe during sample transportation, it will also lead to reduced shipping costs. Transformation of remote clinical outposts into high-quality analytical and diagnostic units is also achievable with the introduction of this device. In addition, with this novel portable sampling probe, Malaysia can position itself as a pioneer in the development of portable sample preparation technology.

From an economic standpoint, the global sample preparation market size is estimated to increase at a compound annual growth rate (CAGR) of 6.6% from $7.57 billion in 2021 to $8.06 billion in 2022. The market is projected to rise to $9.84 billion in 2026 at a CAGR of 5.1% (34). Therefore, the development of this portable electrokinetic extraction device into a commercially viable product is expected to bring significant economic gains to the country as one of the advanced sample preparation techniques.

1.6 Outline of the Thesis

This thesis comprises five chapters.

Chapter 1 gives a detailed description of the research background, problem statement, objectives, scope and significance of the project.

Chapter 2 provides a review of the literature on existing miniaturized sample preparation techniques, electric field enhanced sample preparation techniques, PIMs, miniaturized blood sampling techniques, miniaturized analytical tools, TDM and doxorubicin.

Chapter 3 describes the method of fabrication of the PIM sampling probe, the set parameters for each characterisation technique, the sample and standard solution preparation procedure, the electrokinetic extraction platform, the electrokinetic extraction procedure in real sample matrices, and the quantification assays using HPLC-FLD and LC-MS/MS.

Chapter 4 discusses the configuration and physical characteristics of the PIM sampling probe, followed by a discussion on the optimisations of PIM composition, the number of dipping cycles in the PIM sampling probe fabrication process and operating conditions for electrokinetic extraction. The application of the developed electrokinetic extraction setup was firstly demonstrated through the visualisation of the fluorescent dye Rhodamine 6G extracted on the PIM sampling probe. This chapter also reports the application of the PIM sampling probe in the electrokinetic extraction of the anticancer drug doxorubicin from biological fluid samples, including human plasma, human serum and DBS samples. Spiked doxorubicin was electrokinetically extracted using the PIM sampling probe; this was followed by desorption of the extracted analytes in a suitable solvent for LC-MS/MS analysis. The PIM sampling probe was further integrated into a portable battery-powered device for in-transit sample preparation. The performance of the PIM sampling probe in the electrokinetic extraction of doxorubicin in real sample matrices was validated.

Chapter 5 provides a conclusion to the thesis. The main elements of the study—including the design, the optimal conditions and the applications of the electrokinetic extraction technique—are summarised. Suggestions for future studies are also presented.

REFERENCES

1. Xu, L., Hu, Z. S., Duan, R., Wang, X., Yang, Y. S., Dong, L. Y., Wang, X. H. Advances and applications of in-tube solid-phase microextraction for analysis of proteins. *J Chromatogr A*. 2021. 1640: 461962.
2. Chen, L., Wang, J., Xu, T., Feng, X., Huang, C., Shen, X. Recent sample pretreatment methods for determination of selective serotonin reuptake inhibitors (SSRIs) in biological samples. *J Pharm Biomed Anal*. 2021. 206: 114364.
3. Soltani, S., Jouyban, A. Biological sample preparation: attempts on productivity increasing in bioanalysis. *Bioanalysis*. 2014. 6(12): 1691-1710.
4. Yamini, Y., Rezazadeh, M., Seidi, S. Liquid-phase microextraction – The different principles and configurations. *TrAC Trends in Analytical Chemistry*. 2019. 112: 264-272.
5. Zeng, J., Zou, J., Song, X., Chen, J., Ji, J., Wang, B., Wang, Y., Ha, J., Chen, X. A new strategy for basic drug extraction in aqueous medium using electrochemically enhanced solid-phase microextraction. *J Chromatogr A*. 2011. 1218(2): 191-196.
6. Xia, L., Yang, J., Su, R., Zhou, W., Zhang, Y., Zhong, Y., Huang, S., Chen, Y., Li, G. Recent Progress in Fast Sample Preparation Techniques. *Analytical Chemistry*. 2019. 92(1): 34-48.
7. Wu, J., Huang, Y., Huang, X. Efficient trap of polar aromatic amines in environmental waters by electroenhanced solid phase microextraction based on porous monolith doped with carboxylic carbon nanotubes. *Separation and Purification Technology*. 2022. 282: 120067.
8. Pedersen-Bjergaard, S., Huang, C., Gjelstad, A. Electromembrane extraction-Recent trends and where to go. *J Pharm Anal*. 2017. 7(3): 141-147.
9. Pedersen-Bjergaard, S., Rasmussen, K. E. Electrokinetic migration across artificial liquid membranes. New concept for rapid sample preparation of biological fluids. *J Chromatogr A*. 2006. 1109(2): 183-190.
10. Gjelstad, A., Rasmussen, K. E., Pedersen-Bjergaard, S. Electrokinetic migration across artificial liquid membranes Tuning the membrane chemistry to different types of drug substances. *J Chromatogr A*. 2006. 1124(1-2): 29-34.
11. Fotouhi, L., Yamini, Y., Molaei, S., Seidi, S. Comparison of conventional hollow fiber based liquid phase microextraction and electromembrane extraction efficiencies for the extraction of ephedrine from biological fluids. *J Chromatogr A*. 2011. 1218(48): 8581-8586.
12. Rezazadeh, M., Yamini, Y., Seidi, S., Ebrahimpour, B. Electromembrane surrounded solid phase microextraction: a novel approach for efficient extraction from complicated matrices. *J Chromatogr A*. 2013. 1280: 16-22.
13. Rezazadeh, M., Yamini, Y., Seidi, S. Application of a new nanocarbonaceous sorbent in electromembrane surrounded solid phase microextraction for analysis of amphetamine and methamphetamine in human urine and whole blood. *J Chromatogr A*. 2015. 1396: 1-6.
14. Rezazadeh, M., Yamini, Y., Seidi, S., Tahmasebi, E., Rezaei, F. Electromembrane Surrounded Solid Phase Microextraction Followed by Injection Port Derivatisation and Gas Chromatography-Flame Ionisation

Detector Analysis for Determination of Acidic Herbicides in Plant Tissue. *Journal of Agricultural and Food Chemistry*. 2014. 62(14): 3134-3142.
15. Shamsayei, M., Yamini, Y., Asiabi, H., Rezazadeh, M., Seidi, S. Electromembrane surrounded solid-phase microextraction using a stainless-steel wire coated with a nanocomposite composed of polypyrrole and manganese dioxide. *Microchimica Acta*. 2017. 184(8): 2697-2705.
16. Mohammadkhani, E., Yamini, Y., Rezazadeh, M., Seidi, S. Electromembrane surrounded solid phase microextraction using electrochemically synthesized nanostructured polypyrrole fiber. *J Chromatogr A*. 2016. 1443: 75-82.
17. Nghiem, L., Mornane, P., Potter, I., Perera, J., Cattrall, R., Kolev, S. Extraction and transport of metal ions and small organic compounds using polymer inclusion membranes (PIMs). *Journal of Membrane Science*. 2006. 281(1-2): 7-41.
18. See, H. H., Stratz, S., Hauser, P. C. Electro-driven extraction across a polymer inclusion membrane in a flow-through cell. *J Chromatogr A*. 2013. 1300: 79-84.
19. Schmidt-Marzinkowski, J., See, H. H., Hauser, P. C. Electric Field Driven Extraction of Inorganic Anions Across a Polymer Inclusion Membrane. *Electroanalysis*. 2013. 25(8): 1879-1886.
20. See, H. H., Hauser, P. C. Electro-driven extraction of low levels of lipophilic organic anions and cations across plasticized cellulose triacetate membranes: Effect of the membrane composition. *Journal of Membrane Science*. 2014. 450: 147-152.
21. Nanthasurasak, P., See, H. H., Zhang, M., Guijt, R. M., Breadmore, M. C. In-Transit Electroextraction of Small-Molecule Pharmaceuticals from Blood. *Angew Chem Int Ed Engl*. 2019. 58(12): 3790-3794.
22. Kaya, A., Onac, C., Alpoguz, H. K. A novel electro-driven membrane for removal of chromium ions using polymer inclusion membrane under constant D.C. electric current. *J Hazard Mater*. 2016. 317: 1-7.
23. Meng, X., Li, J., Lv, Y., Feng, Y., Zhong, Y. Electro-membrane extraction of cadmium(II) by bis(2-ethylhexyl) phosphate/kerosene/polyvinyl chloride polymer inclusion membrane. *J Hazard Mater*. 2020. 386: 121990.
24. Mamat, N. A., See, H. H. Development and evaluation of electromembrane extraction across a hollow polymer inclusion membrane. *J Chromatogr A*. 2015. 1406: 34-39.
25. Mamat, N. A., See, H. H. Simultaneous electromembrane extraction of cationic and anionic herbicides across hollow polymer inclusion membranes with a bubbleless electrode. *J Chromatogr A*. 2017. 1504: 9-16.
26. Paul, R., Ostermann, E., Wei, Q. Advances in point-of-care nucleic acid extraction technologies for rapid diagnosis of human and plant diseases. *Biosens Bioelectron*. 2020. 169: 112592.
27. Denniff, P., Spooner, N. Volumetric absorptive microsampling: a dried sample collection technique for quantitative bioanalysis. *Anal Chem*. 2014. 86(16): 8489-8495.
28. Rysava, L., Dvorak, M., Kuban, P. Dried Blood Spot Self-Sampling with Automated Capillary Electrophoresis Processing for Clinical Analysis. *Angew Chem Int Ed Engl*. 2021. 60(11): 6068-6075.
29. Tey, H. Y., See, H. H. A review of recent advances in microsampling techniques of biological fluids for therapeutic drug monitoring. *J Chromatogr A*. 2021. 1635: 461731.

30. Tacar, O., Sriamornsak, P., Dass, C. R. Doxorubicin: an update on anticancer molecular action, toxicity and novel drug delivery systems. *Journal of Pharmacy and Pharmacology*. 2013. 65(2): 157-170.
31. Rudge, J., Kushon, S. Volumetric absorptive microsampling: its use in COVID-19 research and testing. *Bioanalysis*. 2021. 13(24): 1851-1863.
32. Amara, A. B., Else, L. J., Tjia, J., Olagunju, A., Puls, R. L., Khoo, S., Back, D. J. A validated method for quantification of efavirenz in dried blood spots using high-performance liquid chromatography-mass spectrometry. *Ther Drug Monit*. 2015. 37(2): 220-228.
33. Weber, J., Oberfeld, S., Bonse, A., Telger, K., Lingg, R., Hempel, G. Validation of a dried blood spot method for therapeutic drug monitoring of citalopram, mirtazapine and risperidone and its active metabolite 9-hydroxyrisperidone using HPLC-MS. *J Pharm Biomed Anal*. 2017. 140: 347-354.
34. Company, T. B. R. Sample Preparation Global Market Report 2022 – By Product (Sample Preparation Instruments, Consumables, Sample Preparation Kit), By Application (Proteomics, Genomics, Epigenomics), By End User (Pharmaceutical, Biotechnology, Molecular Diagnostics) – Market Size, Trends, And Global Forecast 2022-2026. 2022.
35. Wilde, C., Out, D., Johnson, S., Granger, D. A. Sample Collection, Including Participant Preparation and Sample Handling. *The Immunoassay Handbook* 2013: 427-440.
36. Navitha Reddy, G., Dilip Zagade, A., Sengupta, P. Current direction and advances in analytical sample extraction techniques for drugs with special emphasis on bioanalysis. *Bioanalysis*. 2019. 11(4).
37. Hansen, F. A., Pedersen-Bjergaard, S. Emerging Extraction Strategies in Analytical Chemistry. *Anal Chem*. 2020. 92(1): 2-15.
38. Insight, C. M. Sample Preparation Market, by Product Type (Reagents & Kits, Instruments), By Application (Research Application, Diagnosis), by End User (Pharmaceuticals and Biotechnology Companies, Pathology & Diagnostic Laboratories, Academics & Research Laboratories), and by Region (North America, Latin America, Europe, Asia Pacific, Middle East, and Africa)- Size, Share, Outlook, and Opportunity Analysis, 2022 - 2030. 2022 July 2022.
39. Kabir, A., Locatelli, M., Ulusoy, H. Recent Trends in Microextraction Techniques Employed in Analytical and Bioanalytical Sample Preparation. *Separations*. 2017. 4(4): 36.
40. Tajabadi, F., Ghambarian, M. Carrier-mediated extraction: Applications in extraction and microextraction methods. *Talanta*. 2020. 206: 120145.
41. Gorynski, K. A critical review of solid-phase microextraction applied in drugs of abuse determinations and potential applications for targeted doping testing. *TrAC Trends in Analytical Chemistry*. 2019. 112: 135-146.
42. Jalili, V., Barkhordari, A., Ghiasvand, A. A comprehensive look at solid-phase microextraction technique: A review of reviews. *Microchemical Journal*. 2020. 152: 104319.
43. Jalili, V., Barkhordari, A., Ghiasvand, A. New extraction media in microextraction techniques. A review of reviews. *Microchemical Journal*. 2020. 153: 104386.
44. Kim, U. J., Karthikraj, R. Solid-phase microextraction for the human biomonitoring of environmental chemicals: Current applications and future perspectives. *J Sep Sci*. 2021. 44(1): 247-273.

45. Peng, S., Huang, X., Huang, Y., Huang, Y., Zheng, J., Zhu, F., Xu, J., Ouyang, G. Novel solid-phase microextraction fiber coatings: A review. *J Sep Sci*. 2022. 45(1): 282-304.
46. Abdolhosseini, S., Ghiasvand, A. R., Heidari, N. Comparison of the Conventional and Electroenhanced Direct-Immersion Solid-Phase Microextraction for Sampling of Nicotine in Biological Fluids of the Human Body. *Molecules*. 2018. 23(5): 1171
47. Barroso, M., Moreno, I., Fonseca, B. d., Queiroz, J. A., Gallardo, E. Role of microextraction sampling procedures in forensic toxicology. *Bioanalysis*. 2012. 4(14): 1805–1826.
48. Barahona, F., Turiel, E., Martin-Esteban, A. Supported liquid membrane-protected molecularly imprinted fibre for solid-phase microextraction of thiabendazole. *Anal Chim Acta*. 2011. 694(1-2): 83-89.
49. Sajid, M., Khaled Nazal, M., Rutkowska, M., Szczepanska, N., Namiesnik, J., Plotka-Wasylka, J. Solid Phase Microextraction: Apparatus, Sorbent Materials, and Application. *Critical Reviews in Analytical Chemistry*. 2019. 49(3): 271-288.
50. Ingle, R. G., Zeng, S., Jiang, H., Fang, W.-J. Current developments of bioanalytical sample preparation techniques in pharmaceuticals. *Journal of Pharmaceutical Analysis*. 2022. 12(4): 517-529.
51. Rasmussen, K. E., Pedersen-Bjergaard, S. Developments in hollow fibre-based, liquid-phase microextraction. *TrAC Trends in Analytical Chemistry*. 2004. 23(1): 1-10.
52. Wang, H., Liu, X., Tu, M., Xu, X., Yang, S., Chen, D. Current sample preparation methods and analytical techniques for the determination of synthetic antioxidants in edible oils. *J Sep Sci*. 2022.:1-13.
53. Amico, D., Tassone, A., Pirrone, N., Sprovieri, F., Naccarato, A. Recent applications and novel strategies for mercury determination in environmental samples using microextraction-based approaches: A review. *J Hazard Mater*. 2022. 433: 128823.
54. Msagati, T., Chimuka, L., Cukrowska, E. Sample preparation using liquid membrane extraction techniques. *Water SA*. 2008. 34: 421-428.
55. Yurtov, E. V., Koroleva, M. Y. Liquid membranes for extraction. *Petroleum Chemistry*. 2015. 54(8): 581-594.
56. Huang, C., Seip, K. F., Gjelstad, A., Pedersen-Bjergaard, S. Electromembrane extraction for pharmaceutical and biomedical analysis - Quo vadis. *J Pharm Biomed Anal*. 2015. 113: 97-107.
57. Kuban, P., Slampova, A., Bocek, P. Electric field-enhanced transport across phase boundaries and membranes and its potential use in sample pretreatment for bioanalysis. *Electrophoresis*. 2010. 31(5): 768-785.
58. Hansen, F. A., Petersen, N. J., Kutter, J. P., Pedersen-Bjergaard, S. Electromembrane extraction in microfluidic formats. *J Sep Sci*. 2022. 45(1): 246-257.
59. Huang, C., Chen, Z., Gjelstad, A., Pedersen-Bjergaard, S., Shen, X. Electromembrane extraction. *TrAC Trends in Analytical Chemistry*. 2017. 95: 47-56.
60. Huang, C., Gjelstad, A., Seip, K. F., Jensen, H., Pedersen-Bjergaard, S. Exhaustive and stable electromembrane extraction of acidic drugs from human plasma. *J Chromatogr A*. 2015. 1425: 81-87.

61. Zarghampour, F., Yamini, Y., Baharfar, M., Faraji, M. Simultaneous extraction of acidic and basic drugs via on-chip electromembrane extraction using a single-compartment microfluidic device. *Analyst*. 2019. 144(4): 1159-1166.
62. Basheer, C., Lee, J., Pedersen-Bjergaard, S., Rasmussen, K. E., Lee, H. K. Simultaneous extraction of acidic and basic drugs at neutral sample pH: a novel electro-mediated microextraction approach. *J Chromatogr A*. 2010. 1217(43): 6661-6667.
63. Lee, J., Khalilian, F., Bagheri, H., Lee, H. K. Optimisation of some experimental parameters in the electro membrane extraction of chlorophenols from seawater. *J Chromatogr A*. 2009. 1216(45): 7687-7693.
64. Drouin, N., Kubáň, P., Rudaz, S., Pedersen-Bjergaard, S., Schappler, J. Electromembrane extraction: Overview of the last decade. *TrAC Trends in Analytical Chemistry*. 2019. 113: 357-363.
65. Oedit, A., Ramautar, R., Hankemeier, T., Lindenburg, P. W. Electroextraction and electromembrane extraction: Advances in hyphenation to analytical techniques. *Electrophoresis*. 2016. 37(9): 1170-1186.
66. Kuba´nˇ, P., lampova´, A. S., Bocˇek, P. Electric field-enhanced transport across phase boundaries and membranes and its potential use in sample pretreatment for bioanalysis. *Electrophoresis*. 2010. 31: 768-785.
67. See, H. H., Hauser, P. C. Electric field-driven extraction of lipophilic anions across a carrier-mediated polymer inclusion membrane. *Anal Chem*. 2011. 83(19): 7507-7513.
68. Cristina, R. H., Maria Jesus, M. V., Rut, F. T., Miguel Angel, B. L. Use of Polymer Inclusion Membranes (PIMs) as support for electromembrane extraction of non-steroidal anti-inflammatory drugs and highly polar acidic drugs. *Talanta*. 2018. 179: 601-607.
69. Meng, X., Wang, C., Ren, T., Wang, L., Wang, X. Electrodriven transport of chromium (VI) using 1-octanol/PVC in polymer inclusion membrane under low voltage. *Chemical Engineering Journal*. 2018. 346: 506-514.
70. Chaudhury, S., Bhattacharyya, A., Goswami, A. Electrodriven Transport of Cs+ through Polymer Inclusion Membrane as “Solvent Separated Ions”. *Industrial & Engineering Chemistry Research*. 2016. 55(11): 3120-3124.
71. Gjelstad, A., Pedersen-Bjergaard, S. Recent developments in electromembrane extraction. *Anal Methods*. 2013. 5(18): 4549-4557.
72. Slampova, A., Kuban, P., Bocek, P. Electromembrane extraction using stabilized constant d.c. electric current--a simple tool for improvement of extraction performance. *J Chromatogr A*. 2012. 1234: 32-37.
73. See, H. H., Hauser, P. C. Automated electric-field-driven membrane extraction system coupled to liquid chromatography-mass spectrometry. *Anal Chem*. 2014. 86(17): 8665-8670.
74. Tan, T. Y., Basheer, C., Yan Ang, M. J., Lee, H. K. Electroenhanced solid-phase microextraction of methamphetamine with commercial fibers. *J Chromatogr A*. 2013. 1297: 12-16.
75. Wu, J., Chen, H., Huang, X. Monolith/aminated graphene oxide composite-based electric field-assisted solid phase microextraction for efficient capture of phenoxycarboxylic acids herbicides in environmental waters. *J Chromatogr A*. 2021. 1653: 462407.
76. Djozan, D., Baheri, T., Pournaghi-Azar, M. H. Development of Electro Solid-Phase Microextraction and Application to Methamphetamine Analysis. *Chromatographia*. 2006. 65(1-2): 45-50.

77. Asiabi, H., Yamini, Y., Shamsayei, M. Development of electrochemically controlled packed-in-tube solid phase microextraction method for sensitive analysis of acidic drugs in biological samples. *Talanta*. 2018. 185: 80-88.
78. Davarani, S. S., Nojavan, S., Asadi, R., Banitaba, M. H. Electro-assisted solid-phase microextraction based on poly(3,4-ethylenedioxythiophen) combined with GC for the quantification of tricyclic antidepressants. *J Sep Sci*. 2013. 36(14): 2315-2322.
79. Roszkowska, A., Miekus, N., Baczek, T. Application of solid-phase microextraction in current biomedical research. *J Sep Sci*. 2019. 42(1): 285-302.
80. Maiphetlho, K., Chimuka, L., Tutu, H., Richards, H. Technical design and optimisation of polymer inclusion membranes (PIMs) for sample pre-treatment and passive sampling - A review. *Sci Total Environ*. 2021. 799: 149483.
81. Almeida, M., Cattrall, R. W., Kolev, S. D. Polymer inclusion membranes (PIMs) in chemical analysis - A review. *Anal Chim Acta*. 2017. 987: 1-14.
82. Chen, L., Wu, Y., Dong, H., Meng, M., Li, C., Yan, Y., Chen, J. An overview on membrane strategies for rare earths extraction and separation. *Separation and Purification Technology*. 2018. 197: 70-85.
83. Kuswandi, B., Nitti, F., Almeida, M. I. G. S., Kolev, S. D. Water monitoring using polymer inclusion membranes: a review. *Environmental Chemistry Letters*. 2020. 18(1): 129-150.
84. Suah, F. B. M., Ahmad, M. Preparation and characterisation of polymer inclusion membrane based optode for determination of Al(3+) ion. *Anal Chim Acta*. 2017. 951: 133-139.
85. Pereira, N., John, A. S., Cattrall, R. W., Perera, J. M., Kolev, S. D. Influence of the composition of polymer inclusion membranes on their homogeneity and flexibility. *Desalination* 2009. 236: 327–333.
86. Hoque, B., Almeida, M. I. G. S., Cattrall, R. W., Gopakumar, T. G., Kolev, S. D. Improving the extraction performance of polymer inclusion membranes by cross-linking their polymeric backbone. *Reactive and Functional Polymers*. 2021. 160: 104813.
87. Vázquez, M. I., Romero, V., Fontàs, C., Anticó, E., Benavente, J. Polymer inclusion membranes (PIMs) with the ionic liquid (IL) Aliquat 336 as extractant: Effect of base polymer and IL concentration on their physical–chemical and elastic characteristics. *Journal of Membrane Science*. 2014. 455: 312-319.
88. Almeida, M. I. G. S., Cattrall, R. W., Kolev, S. D. Recent trends in extraction and transport of metal ions using polymer inclusion membranes (PIMs). *Journal of Membrane Science*. 2012. 415-416: 9-23.
89. Mwakalesi, A. J., Potter, I. D. Removal of picloram herbicide from an aqueous environment using polymer inclusion membranes. *Journal of Environmental Chemical Engineering*. 2020. 8(5): 103936.
90. Kolev, S. D., Baba, Y., Cattrall, R. W., Tasaki, T., Pereira, N., Perera, J. M., Stevens, G. W. Solid phase extraction of zinc(II) using a PVC-based polymer inclusion membrane with di(2-ethylhexyl)phosphoric acid (D2EHPA) as the carrier. *Talanta*. 2009. 78(3): 795-799.
91. Ershad, M., Almeida, M. I. G. S., Spassov, T. G., Cattrall, R. W., Kolev, S. D. Polymer inclusion membranes (PIMs) containing purified dinonylnaphthalene sulfonic acid (DNNS) Performance and selectivity. *Separation and Purification Technology*. 2018. 195: 446-452.

92. Vera, R., Antico, E., Eguiazabal, J. I., Aranburu, N., Fontas, C. First Report on a Solvent-Free Preparation of Polymer Inclusion Membranes with an Ionic Liquid. *Molecules*. 2019. 24(10): 1845.
93. Lam, B., Wei, M., Zhu, L., Luo, S., Guo, R., Morisato, A., Alexandridis, P., Lin, H. Cellulose triacetate doped with ionic liquids for membrane gas separation. *Polymer*. 2016. 89: 1-11.
94. Benavente, J., Romero, V., Vázquez, M., Anticó, E., Fontàs, C. Electrochemical Characterisation of a Polymer Inclusion Membrane Made of Cellulose Triacetate and Aliquat 336 and Its Application to Sulfonamides Separation. *Separations*. 2018. 5(1): 1-12.
95. O'Bryan, Y., Cattrall, R. W., Truong, Y. B., Kyratzis, I. L., Kolev, S. D. The use of poly(vinylidenefluoride-co-hexafluoropropylene) for the preparation of polymer inclusion membranes. Application to the extraction of thiocyanate. *Journal of Membrane Science*. 2016. 510: 481-488.
96. Vera, R., Gelde, L., Anticó, E., Martínez de Yuso, M. V., Benavente, J., Fontàs, C. Tuning physicochemical, electrochemical and transport characteristics of polymer inclusion membrane by varying the counter-anion of the ionic liquid Aliquat 336. *Journal of Membrane Science*. 2017. 529: 87-94.
97. Huang, S., Chen, J., Zou, D. A preliminary study of polymer inclusion membrane for lutetium(III) separation and membrane regeneration. *Journal of Rare Earths*. 2020.
98. White, K. M., Smith, B. D., Duggan, P. J., Sheahan, S. L., Tyndall, E. M. Mechanism of facilitated saccharide transport through plasticized cellulose triacetate membranes. *Journal of Membrane Science*. 2001. 194(2): 165-175.
99. Hassoune, H., Rhlalou, T., Verchère, J.-F. Mechanism of transport of sugars across a supported liquid membrane using methyl cholate as mobile carrier. *Desalination*. 2009. 242(1-3): 84-95.
100. Hu, F., Hu, H., Tang, J., Qiu, X., Jin, W., Hu, J. Plasticization-induced oriented micro-channels within polymer inclusion membranes for facilitating Cu(II) transport. *Journal of Molecular Liquids*. 2020. 301: 112457.
101. Nagul, E. A., Croft, C. F., Cattrall, R. W., Kolev, S. D. Nanostructural characterisation of polymer inclusion membranes using X-ray scattering. *Journal of Membrane Science*. 2019. 588: 117208.
102. Rodríguez de San Miguel, E., Monroy-Barreto, M., Aguilar, J. C., Ocampo, A. L., de Gyves, J. Structural effects on metal ion migration across polymer inclusion membranes: Dependence of membrane properties and transport profiles on the weight and volume fractions of the components. *Journal of Membrane Science*. 2011. 379(1-2): 416-425.
103. Meng, X., Song, Y., Lv, Y., Xin, X., Ren, T., Wang, X. Study on stable mass transfer and enrichment of phenol by 1-octanol/kerosene/polyvinyl chloride polymer inclusion membrane. *Environmental Pollution*. 2019. 253: 1100-1106.
104. Li, Z., Liu, Y., Wang, B., Lang, Q., Tan, M., Lee, M., Peng, C., Zhang, Y. Insights into the facilitated transport mechanisms of Cr(VI) in ionic liquid-based polymer inclusion membrane – Electrodialysis (PIM-ED) process. *Chemical Engineering Journal*. 2020. 397: 125324.
105. Cho, Y., Cattrall, R. W., Kolev, S. D. A novel polymer inclusion membrane based method for continuous clean-up of thiocyanate from gold mine tailings water. *J Hazard Mater*. 2018. 341: 297-303.

106. Cho, Y., Xu, C., Cattrall, R. W., Kolev, S. D. A polymer inclusion membrane for extracting thiocyanate from weakly alkaline solutions. *Journal of Membrane Science*. 2011. 367(1-2): 85-90.
107. Salima, A., Ounissa, K.-S., Lynda, M., Mohamed, B. Cationic dye (MB) removal using polymer inclusion membrane (PIMs). *Procedia Engineering*. 2012. 33: 38-46.
108. Upitis, A., Peterson, J., Lukey, C., Nghiem, L. D. Metallic ion extraction using polymer inclusion membranes (PIMs): Optimising physical strength and extraction rate. *Desalination and Water Treatment*. 2012. 6(1-3): 41-47.
109. Pantuckova, P., Kuban, P., Bocek, P. In-line coupling of microextractions across polymer inclusion membranes to capillary zone electrophoresis for rapid determination of formate in blood samples. *Anal Chim Acta*. 2015. 887: 111-117.
110. Perez-Silva, I., Rodriguez, J. A., Ramirez-Silva, M. T., Paez-Hernandez, M. E. Determination of oxytetracycline in milk samples by polymer inclusion membrane separation coupled to high performance liquid chromatography. *Anal Chim Acta*. 2012. 718: 42-46.
111. Vera, R., Insa, S., Fontas, C., Antico, E. A new extraction phase based on a polymer inclusion membrane for the detection of chlorpyrifos, diazinon and cyprodinil in natural water samples. *Talanta*. 2018. 185: 291-298.
112. Ling, Y. Y., Mohd Suah, F. B. Extraction of malachite green from wastewater by using polymer inclusion membrane. *Journal of Environmental Chemical Engineering*. 2017. 5(1): 785-794.
113. Mohd Amin, N. H., Mehamod, F. S., Mohd Suah, F. B. A novel approach in simultaneous extraction of basic dyes by using a batch reactor consisting a polymer inclusion membrane. *Alexandria Engineering Journal*. 2019. 58(3): 929-935.
114. Ohshima, T., Kagaya, S., Gemmei-Ide, M., Cattrall, R. W., Kolev, S. D. The use of a polymer inclusion membrane as a sorbent for online preconcentration in the flow injection determination of thiocyanate impurity in ammonium sulfate fertilizer. *Talanta*. 2014. 129: 560-564.
115. Nagul, E. A., Fontas, C., McKelvie, I. D., Cattrall, R. W., Kolev, S. D. The use of a polymer inclusion membrane for separation and preconcentration of orthophosphate in flow analysis. *Anal Chim Acta*. 2013. 803: 82-90.
116. Garcia-Rodríguez, A., Matamoros, V., Kolev, S. D., Fontàs, C. Development of a polymer inclusion membrane (PIM) for the preconcentration of antibiotics in environmental water samples. *Journal of Membrane Science*. 2015. 492: 32-39.
117. Vera, R., Zhang, Y., Fontas, C., Almeida, M., Antico, E., Cattrall, R. W., Kolev, S. D. Automatic determination of arsenate in drinking water by flow analysis with dual membrane-based separation. *Food Chem*. 2019. 283: 232-238.
118. Yaftian, M. R., Almeida, M., Cattrall, R. W., Kolev, S. D. Flow injection spectrophotometric determination of V(V) involving on-line separation using a poly(vinylidene fluoride-co-hexafluoropropylene)-based polymer inclusion membrane. *Talanta*. 2018. 181: 385-391.
119. Zhang, L. L., Cattrall, R. W., Kolev, S. D. The use of a polymer inclusion membrane in flow injection analysis for the on-line separation and determination of zinc. *Talanta*. 2011. 84(5): 1278-1283.

120. Elias, G., Diez, S., Fontas, C. System for mercury preconcentration in natural waters based on a polymer inclusion membrane incorporating an ionic liquid. *J Hazard Mater*. 2019. 371: 316-322.
121. Nys, G., Kok, M. G. M., Servais, A.-C., Fillet, M. Beyond dried blood spot: Current microsampling techniques in the context of biomedical applications. *TrAC Trends in Analytical Chemistry*. 2017. 97: 326-332.
122. Milosheska, D., Grabnar, I., Vovk, T. Dried blood spots for monitoring and individualization of antiepileptic drug treatment. *Eur J Pharm Sci*. 2015. 75: 25-39.
123. Guthrie, R., Susi, A. A simple phenylalanine method for detecting Phenylketonuria in large populations of newborn infants. *Pediatrics*. 1963. 32(3): 338-343.
124. Babu, S., Prasad, M., Miller, M., Morrissey, M., Bhutada, A., Rojas, M., Rastogi, S. Use of serum citrulline concentrations from routine newborn screen as a biomarker for necrotizing enterocolitis. *Pediatr Surg Int*. 2019. 35(6): 715-722.
125. Samsonova, J. V., Saushkin, N. Y., Osipov, A. P., Kondakov, S. E., Fomina, S. N., Mischenko, A. V. Detection of antibodies against foot-and-mouth disease virus serotypes A, O and Asia-1 by ELISA in strip-dried samples from vaccinated bovines. *Appl Biochem Biotechnol*. 2019. 188(2): 491-497.
126. Martial, L. C., Kerkhoff, J., Martinez, N., Rodriguez, M., Coronel, R., Molinas, G., Roman, M., Gomez, R., Aguirre, S., Jongedijk, E., Huisman, J., Touw, D. J., Perez, D., Chaparro, G., Gonzalez, F., Aarnoutse, R. E., Alffenaar, J. W., Magis-Escurra, C. Evaluation of dried blood spot sampling for pharmacokinetic research and therapeutic drug monitoring of anti-tuberculosis drugs in children. *Int J Antimicrob Agents*. 2018. 52(1): 109-113.
127. Kloosterboer, S. M., de Winter, B. C. M., Bahmany, S., Al-Hassany, L., Dekker, A., Dieleman, G. C., van Gelder, T., Dierckx, B., Koch, B. C. P. Dried blood spot analysis for therapeutic drug monitoring of antipsychotics: Drawbacks of its clinical application. *Ther Drug Monit*. 2018. 40(3): 344-350.
128. Duthaler, U., Berger, B., Erb, S., Battegay, M., Letang, E., Gaugler, S., Natamatungiro, A., Mnzava, D., Donzelli, M., Krahenbuhl, S., Haschke, M. Using dried blood spots to facilitate therapeutic drug monitoring of antiretroviral drugs in resource-poor regions. *J Antimicrob Chemother*. 2018. 73(10): 2729-2737.
129. Sadler Simoes, S., Castanera Ajenjo, A., Dias, M. J. Dried blood spots combined to an UPLC-MS/MS method for the simultaneous determination of drugs of abuse in forensic toxicology. *J Pharm Biomed Anal*. 2018. 147: 634-644.
130. Baldo, M. N., Angeli, E., Gareis, N. C., Hunzicker, G. A., Murguia, M. C., Ortega, H. H., Hein, G. J. Liquid chromatography tandem-mass spectrometry (LC-MS/MS) and dried blood spot sampling applied to pharmacokinetics studies in animals: Correlation of classic and block design. *Lab Anim*. 2018. 52(2): 125-134.
131. Lee, H., Park, Y., Jo, J., In, S., Park, Y., Kim, E., Pyo, J., Choe, S. Analysis of benzodiazepines and their metabolites using DBS cards and LC-MS/MS. *Forensic Sci Int*. 2015. 255: 137-145.
132. Patteet, L., Maudens, K. E., Stove, C. P., Lambert, W. E., Morrens, M., Sabbe, B., Neels, H. The use of dried blood spots for quantification of 15 antipsychotics and 7 metabolites with ultra-high performance liquid

chromatography - tandem mass spectrometry. *Drug Test Anal*. 2015. 7(6): 502-511.
133. Denniff, P., Spooner, N. The effect of hematocrit on assay bias when using DBS samples for the quantitative bioanalysis of drugs. *Bioanalysis*. 2010. 2(8): 1385-1395.
134. Velghe, S., Deprez, S., Stove, C. P. Fully automated therapeutic drug monitoring of anti-epileptic drugs making use of dried blood spots. *J Chromatogr A*. 2019. 1601: 95-103.
135. Taylor, R. R., Hoffman, K. L., Schniedewind, B., Clavijo, C., Galinkin, J. L., Christians, U. Comparison of the quantification of acetaminophen in plasma, cerebrospinal fluid and dried blood spots using high-performance liquid chromatography-tandem mass spectrometry. *J Pharm Biomed Anal*. 2013. 83: 1-9.
136. Verplaetse, R., Henion, J. Quantitative determination of opioids in whole blood using fully automated dried blood spot desorption coupled to on-line SPE-LC-MS/MS. *Drug Test Anal*. 2016. 8(1): 30-38.
137. Torres, L. M., Rivera-Espinosa, L., Chavez-Pacheco, J. L., Navas, C. F., Demetrio, J. A., Alemon-Medina, R., Trujillo, F., Perez, M., Zapata, M. M., Cardenas, R., Salinas, C., Aquino, A., Velazquez-Cruz, R., Castillejos, M. D. A new method to quantify ifosfamide blood levels using dried blood spots and UPLC-MS/MS in paediatric patients with embryonic solid tumours. *PLoS One*. 2015. 10(11): e0143421.
138. Linder, C., Andersson, M., Wide, K., Beck, O., Pohanka, A. A LC-MS/MS method for therapeutic drug monitoring of carbamazepine, lamotrigine and valproic acid in DBS. *Bioanalysis*. 2015. 7(16): 2031-2039.
139. Raymundo, S., Muller, V. V., Andriguetti, N. B., Tegner, M., Artmann, A. C., Kluck, H. M., Franzoi, M. A., Vilela, R. M. M., Schwartsmann, G., Linden, R., Antunes, M. V. Determination of docetaxel in dried blood spots by LC-MS/MS: Method development, validation and clinical application. *J Pharm Biomed Anal*. 2018. 157: 84-91.
140. Knapen, L. M., Beer, Y., Bruggemann, R. J. M., Stolk, L. M., Vries, F., Tjan-Heijnen, V. C. G., Erp, N. P. V., Croes, S. Development and validation of an analytical method using UPLC-MS/MS to quantify everolimus in dried blood spots in the oncology setting. *J Pharm Biomed Anal*. 2018. 149: 106-113.
141. Antunes, M. V., Raymundo, S., Wagner, S. C., Mattevi, V. S., Vieira, N., Leite, R., Reginato, F., Capra, M. Z., Fogliatto, L., Linden, R. DBS sampling in imatinib therapeutic drug monitoring: from method development to clinical application. *Bioanalysis*. 2015. 7(16): 2105-2117.
142. Verheijen, R. B., Bins, S., Thijssen, B., Rosing, H., Nan, L., Schellens, J. H., Mathijssen, R. H., Lolkema, M. P., Beijnen, J. H., Steeghs, N., Huitema, A. D. Development and clinical validation of an LC-MS/MS method for the quantification of pazopanib in DBS. *Bioanalysis*. 2016. 8(2): 123-134.
143. Alsous, M. M., Hawwa, A. F., McElnay, J. C. Determination of azathioprine/6-mercaptopurine metabolites in dried blood spots: Correlation with RBC concentrations. *J Pharm Biomed Anal*. 2020. 178: 112870.
144. De Kesel, P. M., Lambert, W. E., Stove, C. P. Does volumetric absorptive microsampling eliminate the hematocrit bias for caffeine and paraxanthine in dried blood samples? A comparative study. *Anal Chim Acta*. 2015. 881: 65-73.
145. Spooner, N., Denniff, P., Michielsen, L., De Vries, R., Ji, Q. C., Arnold, M. E., Woods, K., Woolf, E. J., Xu, Y., Boutet, V., Zane, P., Kushon, S., Rudge, J. B.

A device for dried blood microsampling in quantitative bioanalysis: overcoming the issues associated blood hematocrit. *Bioanalysis*. 2015. 7(6): 653-659.
146. Protti, M., Mandrioli, R., Mercolini, L. Tutorial: Volumetric absorptive microsampling (VAMS). *Anal Chim Acta*. 2019. 1046: 32-47.
147. Tanna, S., Alalaqi, A., Bernieh, D., Lawson, G. Volumetric absorptive microsampling (VAMS) coupled with high-resolution, accurate-mass (HRAM) mass spectrometry as a simplified alternative to dried blood spot (DBS) analysis for therapeutic drug monitoring of cardiovascular drugs. *Clinical Mass Spectrometry*. 2018. 10: 1-8.
148. Velghe, S., Stove, C. P. Volumetric absorptive microsampling as an alternative tool for therapeutic drug monitoring of first-generation anti-epileptic drugs. *Anal Bioanal Chem*. 2018. 410(9): 2331-2341.
149. Andersen, I. K. L., Rosting, C., Gjelstad, A., Halvorsen, T. G. Volumetric absorptive MicroSampling vs. other blood sampling materials in LC-MS-based protein analysis - preliminary investigations. *J Pharm Biomed Anal*. 2018. 156: 239-246.
150. van den Broek, I., Fu, Q., Kushon, S., Kowalski, M. P., Millis, K., Percy, A., Holewinski, R. J., Venkatraman, V., Van Eyk, J. E. Application of volumetric absorptive microsampling for robust, high-throughput mass spectrometric quantification of circulating protein biomarkers. *Clinical Mass Spectrometry*. 2017. 4-5: 25-33.
151. Anoshkina, Y., Costas-Rodriguez, M., Vanhaecke, F. Iron isotopic analysis of finger-prick and venous blood by multi-collector inductively coupled plasma-mass spectrometry after volumetric absorptive microsampling. *J Anal Atom Spectrom*. 2017. 32(2): 314-321.
152. Denniff, P., Parry, S., Dopson, W., Spooner, N. Quantitative bioanalysis of paracetamol in rats using volumetric absorptive microsampling (VAMS). *J Pharm Biomed Anal*. 2015. 108: 61-69.
153. Delahaye, L., Dhont, E., De Cock, P., De Paepe, P., Stove, C. P. Volumetric absorptive microsampling as an alternative sampling strategy for the determination of paracetamol in blood and cerebrospinal fluid. *Anal Bioanal Chem*. 2019. 411(1): 181-191.
154. Sen, A., Wang, Y., Chiu, K., Whiley, L., Cowan, D., Chang, R. C., Legido-Quigley, C. Metabolic phenotype of the healthy rodent model using in-vial extraction of dried serum, urine, and cerebrospinal fluid spots. *Anal Chem*. 2013. 85(15): 7257-7263.
155. Pena-Pereira, F., Lavilla, I., Bendicho, C. Greening sample preparation: An overview of cutting-edge contributions. *Current Opinion in Green and Sustainable Chemistry*. 2021. 30: 100481.
156. Dixon, C., Lamanna, J., Wheeler, A. R. Direct loading of blood for plasma separation and diagnostic assays on a digital microfluidic device. *Lab Chip*. 2020. 20(10): 1845-1855.
157. Schary, W., Paskali, F., Rentschler, S., Ruppert, C., Wagner, G. E., Steinmetz, I., Deigner, H. P., Kohl, M. Open-Source, Adaptable, All-in-One Smartphone-Based System for Quantitative Analysis of Point-of-Care Diagnostics. *Diagnostics (Basel)*. 2022. 12(3): 589.
158. McCracken, K. E., Yoon, J.-Y. Recent approaches for optical smartphone sensing in resource-limited settings: a brief review. *Analytical Methods*. 2016. 8(36): 6591-6601.

159. Ding, X., Mauk, M. G., Yin, K., Kadimisetty, K., Liu, C. Interfacing Pathogen Detection with Smartphones for Point-of-Care Applications. *Anal Chem*. 2019. 91(1): 655-672.
160. Lafreniere, N. M., Shih, S. C., Abu-Rabie, P., Jebrail, M. J., Spooner, N., Wheeler, A. R. Multiplexed extraction and quantitative analysis of pharmaceuticals from DBS samples using digital microfluidics. *Bioanalysis*. 2014. 6(3): 307-318.
161. Wheeler, A. R. Chemistry. Putting electrowetting to work. *Science*. 2008. 322(5901): 539-540.
162. Kirby, A. E., Wheeler, A. R. Digital microfluidics: an emerging sample preparation platform for mass spectrometry. *Anal Chem*. 2013. 85(13): 6178-6184.
163. Shih, S. C., Yang, H., Jebrail, M. J., Fobel, R., McIntosh, N., Al-Dirbashi, O. Y., Chakraborty, P., Wheeler, A. R. Dried blood spot analysis by digital microfluidics coupled to nanoelectrospray ionisation mass spectrometry. *Anal Chem*. 2012. 84(8): 3731-3738.
164. Jebrail, M. J., Yang, H., Mudrik, J. M., Lafreniere, N. M., McRoberts, C., Al-Dirbashi, O. Y., Fisher, L., Chakraborty, P., Wheeler, A. R. A digital microfluidic method for dried blood spot analysis. *Lab Chip*. 2011. 11(19): 3218-3224.
165. Mikhail, I. E., Tehranirokh, M., Gooley, A. A., Guijt, R. M., Breadmore, M. C. Hyphenated sample preparation-electrospray and nano-electrospray ionisation mass spectrometry for biofluid analysis. *J Chromatogr A*. 2021. 1646: 462086.
166. Kirby, A. E., Lafreniere, N. M., Seale, B., Hendricks, P. I., Cooks, R. G., Wheeler, A. R. Analysis on the go: quantitation of drugs of abuse in dried urine with digital microfluidics and miniature mass spectrometry. *Anal Chem*. 2014. 86(12): 6121-6129.
167. Jayawardane, B. M., McKelvie, I. D., Kolev, S. D. A paper-based device for measurement of reactive phosphate in water. *Talanta*. 2012. 100: 454-460.
168. Noviana, E., Ozer, T., Carrell, C. S., Link, J. S., McMahon, C., Jang, I., Henry, C. S. Microfluidic Paper-Based Analytical Devices: From Design to Applications. *Chem Rev*. 2021. 121(19): 11835-11885.
169. Li, H., Steckl, A. J. Paper Microfluidics for Point-of-Care Blood-Based Analysis and Diagnostics. *Anal Chem*. 2019. 91(1): 352-371.
170. Ding, J., He, N., Lisak, G., Qin, W., Bobacka, J. Paper-based microfluidic sampling and separation of analytes for potentiometric ion sensing. *Sensors and Actuators B: Chemical*. 2017. 243: 346-352.
171. Yin, J., Suo, Y., Zou, Z., Sun, J., Zhang, S., Wang, B., Xu, Y., Darland, D., Zhao, J. X., Mu, Y. Integrated microfluidic systems with sample preparation and nucleic acid amplification. *Lab Chip*. 2019. 19(17): 2769-2785.
172. Gan, W., Zhuang, B., Zhang, P., Han, J., Li, C. X., Liu, P. A filter paper-based microdevice for low-cost, rapid, and automated DNA extraction and amplification from diverse sample types. *Lab Chip*. 2014. 14(19): 3719-3728.
173. Nilghaz, A., Shen, W. Low-cost blood plasma separation method using salt functionalized paper. *RSC Advances*. 2015. 5(66): 53172-53179.
174. Luo, Z., Lv, T., Zhu, K., Li, Y., Wang, L., Gooding, J. J., Liu, G., Liu, B. Paper-Based Ratiometric Fluorescence Analytical Devices towards Point-of-Care Testing of Human Serum Albumin. *Angewandte Chemie*. 2020. 132(8): 3155-3160.

175. Schulz, M., Iwersen-Bergmann, S., Andresen, H., Schmoldt, A. Therapeutic and toxic blood concentrations of nearly 1,000 drugs and other xenobiotics. *Crit Care*. 2012. 16(4): R136.
176. Conti, M., Matulli Cavedagna, T., Ramazzotti, E., Mancini, R., Calza, L., Rinaldi, M., Badia, L., Guardigni, V., Viale, P., Verucchi, G. Multiplexed therapeutic drug monitoring (TDM) of antiviral drugs by LC–MS/MS. *Clinical Mass Spectrometry*. 2018. 7: 6-17.
177. Calcagno, A., Motta, I., Milia, M. G., Rostagno, R., Simiele, M., Libanore, V., Fontana, S., D'Avolio, A., Ghisetti, V., Di Perri, G., Bonora, S. Dried plasma/blood spots for monitoring antiretroviral treatment efficacy and pharmacokinetics: a cross-sectional study in rural Burundi. *Br J Clin Pharmacol*. 2015. 79(5): 801-808.
178. Patsalos, P. N., Berry, D. J. Therapeutic drug monitoring of antiepileptic drugs by use of saliva. *Ther Drug Monit*. 2013. 35(1): 4-29.
179. Li, Y. Y., Jiang, Y., Cao, H. W., Lin, H., Ren, W. B., Huang, J., Zhang, J. Therapeutic drug monitoring of valproic acid using a dried plasma spot sampling device. *Journal of Mass Spectrometry*. 2020. 56(4): e4603.
180. Shokry, E., Villanelli, F., Malvagia, S., Rosati, A., Forni, G., Funghini, S., Ombrone, D., Della Bona, M., Guerrini, R., la Marca, G. Therapeutic drug monitoring of carbamazepine and its metabolite in children from dried blood spots using liquid chromatography and tandem mass spectrometry. *J Pharm Biomed Anal*. 2015. 109: 164-170.
181. Berm, E. J., Odigie, B., Bijlsma, M. J., Wilffert, B., Touw, D. J., Maring, J. G. A clinical validation study for application of DBS in therapeutic drug monitoring of antidepressants. *Bioanalysis*. 2016. 8(5): 413-424.
182. Berm, E. J. J., Paardekooper, J., Brummel-Mulder, E., Hak, E., Wilffert, B., Maring, J. G. A simple dried blood spot method for therapeutic drug monitoring of the tricyclic antidepressants amitriptyline, nortriptyline, imipramine, clomipramine, and their active metabolites using LC-MS/MS. *Talanta*. 2015. 134: 165-172.
183. Siebenhaar, M., Kullmer, K., Fernandes, N. M., Hullen, V., Hopf, C. Personalized monitoring of therapeutic salicylic acid in dried blood spots using a three-layer setup and desorption electrospray ionisation mass spectrometry. *Anal Bioanal Chem*. 2015. 407(23): 7229-7238.
184. Ghareeb, M., Akhlaghi, F. Alternative matrices for therapeutic drug monitoring of immunosuppressive agents using LC-MS/MS. *Bioanalysis*. 2015. 7(8): 1037-1058.
185. Dickerson, J. A., Sinkey, M., Jacot, K., Stack, J., Sadilkova, K., Law, Y. M., Jack, R. M. Tacrolimus and sirolimus in capillary dried blood spots allows for remote monitoring. *Pediatr Transplant*. 2015. 19(1): 101-106.
186. Alnaim, L. Therapeutic drug monitoring of cancer chemotherapy. *J Oncol Pharm Pract*. 2007. 13(4): 207-221.
187. Iacuzzi, V., Posocco, B., Zanchetta, M., Gagno, S., Poetto, A. S., Guardascione, M., Toffoli, G. Dried Blood Spot Technique Applied in Therapeutic Drug Monitoring of Anticancer Drugs: a Review on Conversion Methods to Correlate Plasma and Dried Blood Spot Concentrations. *Pharm Res*. 2021. 38(5): 759-778.
188. Hempel, G., Schulze-Westhoff, P., Flege, S., Laubrock, N., Boos, J. Therapeutic drug monitoring of doxorubicin in paediatric oncology using capillary electrophoresis. *Electrophoresis*. 1998. 19(16-17): 2939-2943.

189. Li, G., Zhao, M., Zhao, L. Ultra-performance liquid chromatography-tandem mass spectrometry for simultaneous determination of 12 anti-tumor drugs in human plasma and its application in therapeutic drug monitoring. *J Pharm Biomed Anal*. 2021. 206: 114380.
190. Stratigou, I.-C., Tsiasioti, A., Tzanavaras, P. D., Markopoulou, C. K., Fytianos, K., Zacharis, C. K. Homogeneous liquid liquid extraction using salt as mass separating agent for the ultra high pressure liquid chromatographic determination of doxorubicin in human urine. *Microchemical Journal*. 2020. 158: 105260.
191. Maliszewska, O., Plenis, A., Oledzka, I., Kowalski, P., Miekus, N., Bien, E., Krawczyk, M. A., Adamkiewicz-Drozynska, E., Baczek, T. Optimisation of LC method for the quantification of doxorubicin in plasma and urine samples in view of pharmacokinetic, biomedical and drug monitoring therapy studies. *J Pharm Biomed Anal*. 2018. 158: 376-385.
192. Sun, F., Hung, H. C., Sinclair, A., Zhang, P., Bai, T., Galvan, D. D., Jain, P., Li, B., Jiang, S., Yu, Q. Hierarchical zwitterionic modification of a SERS substrate enables real-time drug monitoring in blood plasma. *Nat Commun*. 2016. 7: 13437.
193. Wenningmann, N., Knapp, M., Ande, A., Vaidya, T. R., Ait-Oudhia, S. Insights into Doxorubicin-induced Cardiotoxicity: Molecular Mechanisms, Preventive Strategies, and Early Monitoring. *Molecular Pharmacology*. 2019. 96(2): 219-232.
194. Xu, J., Liu, Y., Yu, Y., Ni, Q., Chen, Y. Subcellular Quantification of Doxorubicin and Its Metabolite in Cultured Human Leukemia Cells Using Liquid Chromatography-Tandem Mass Spectrometry. *Analytical Letters*. 2012. 45(14): 1980-1994.
195. Harahap, Y., Amalia, S. N., Anarta, A., Ramadhan. Analysis of Doxorubicin and Doxorubicinol in Dried Blood Spot of Breast Cancer Patients for Monitoring the Cardiotoxicity of Doxorubicin. *International Journal of Pharmacy and Pharmaceutical Sciences*. 2020. 12(4):21-25.
196. Harahap, Y., Suryadi, H., Anarta, A. Development and Validation of Doxorubicin Hydrochloride and Doxorubicinol Quantification Method in Dried Blood Spot by Liquid Chromatography-Tandem Mass Spectrometry. *J Pharm Bioallied Sci*. 2020. 12(4): 406-412.
197. Yang, X., Gao, H., Qian, F., Zhao, C., Liao, X. Internal standard method for the measurement of doxorubicin and daunorubicin by capillary electrophoresis with in-column double optical-fiber LED-induced fluorescence detection. *J Pharm Biomed Anal*. 2016. 117: 118-124.
198. Fogli, S., Danesi, R., Innocenti, F., Di Paolo, A., Bocci, G., Barbara, C., Del Tacca, M. An Improved HPLC Method for Therapeutic Drug Monitoring of Daunorubicin, Idarubicin, Doxorubicin, Epirubicin, and Their 13-Dihydro Metabolites in Human Plasma. *Therapeutic Drug Monitoring*. 1999. 21(3): 367.
199. Ehsani, M., Soleymani, J., Hasanzadeh, M., Vaez-Gharamaleki, Y., Khoubnasabjafari, M., Jouyban, A. Sensitive monitoring of doxorubicin in plasma of patients, MDA-MB-231 and 4T1 cell lysates using electroanalysis method. *J Pharm Biomed Anal*. 2021. 192: 113701.
200. Guo, Y., Chen, Y., Zhao, Q., Shuang, S., Dong, C. Electrochemical Sensor for Ultrasensitive Determination of Doxorubicin and Methotrexate Based on Cyclodextrin-Graphene Hybrid Nanosheets. *Electroanalysis*. 2011. 23(10): 2400-2407.

201. Soleymani, J., Hasanzadeh, M., Eskandani, M., Khoubnasabjafari, M., Shadjou, N., Jouyban, A. Electrochemical sensing of doxorubicin in unprocessed whole blood, cell lysate, and human plasma samples using thin film of poly-arginine modified glassy carbon electrode. *Mater Sci Eng C Mater Biol Appl*. 2017. 77: 790-802.
202. Panikar, S. S., Banu, N., Escobar, E. R., Garcia, G. R., Cervantes-Martinez, J., Villegas, T. C., Salas, P., De la Rosa, E. Stealth modified bottom up SERS substrates for label-free therapeutic drug monitoring of doxorubicin in blood serum. *Talanta*. 2020. 218: 121138.
203. Dharmalingam, S. R., Ramamurthy, S., Chidambaram, K., Nadaraju, S. A Simple HPLC Bioanalytical Method for the Determination of Doxorubicin Hydrochloride in Rat Plasma: Application to Pharmacokinetic Studies. *Tropical Journal of Pharmaceutical Research*. 2014. 13(3): 409.
204. Lu, H., Yuan, G., He, Q., Chen, H. Rapid analysis of anthracycline antibiotics doxorubicin and daunorubicin by microchip capillary electrophoresis. *Microchemical Journal*. 2009. 92(2): 170-173.
205. Bojko, B., Looby, N., Olkowicz, M., Roszkowska, A., Kupcewicz, B., Reck Dos Santos, P., Ramadan, K., Keshavjee, S., Waddell, T. K., Gomez-Rios, G., Tascon, M., Gorynski, K., Cypel, M., Pawliszyn, J. Solid phase microextraction chemical biopsy tool for monitoring of doxorubicin residue during in vivo lung chemo-perfusion. *J Pharm Anal*. 2021. 11(1): 37-47.
206. Mazzucchelli, S., Ravelli, A., Gigli, F., Minoli, M., Corsi, F., Ciuffreda, P., Ottria, R. LC-MS/MS method development for quantification of doxorubicin and its metabolite 13-hydroxy doxorubicin in mice biological matrices: Application to a pharmaco-delivery study. *Biomed Chromatogr*. 2017. 31(4): e3863.
207. Ma, N., Zhang, L., Li, R., Zhou, Y., Cai, Z., Dong, C., Shuang, S. Magnetic solid-phase extraction based on a trimethylstearylammonium bromide coated Fe3O4/SiO2 composite for determination of adriamycin hydrochloride in human plasma and urine by HPLC-FLD. *Anal Methods*. 2014. 6(17): 6736-6744.
208. Chong, K. C., Thang, L. Y., Quirino, J. P., See, H. H. Monitoring of vancomycin in human plasma via portable microchip electrophoresis with contactless conductivity detector and multi-stacking strategy. *J Chromatogr A*. 2017. 1485: 142-146.
209. Chiang, T.-L., Wang, Y.-C., Ding, W.-H. Trace Determination of Rhodamine B and Rhodamine 6G Dyes in Aqueous Samples by Solid-phase Extraction and High-performance Liquid Chromatography Coupled with Fluorescence Detection. *Journal of the Chinese Chemical Society*. 2012. 59(4): 515-519.
210. Arous, O., Saoud, F. S., Kerdjoudj, H. Cellulose triacetate properties and their effect on the thin films morphology and performance. *IOP Conference Series: Materials Science and Engineering*. 2010. 12: 012001.
211. Rathika, R., Suthanthiraraj, S. A. Influence of 1-ethyl-3-methylimidazolium bis (trifluoromethyl sulfonyl) imide plasticization on zinc-ion conducting PEO/PVdF blend gel polymer electrolyte. *Journal of Materials Science: Materials in Electronics*. 2018. 29(23): 19632-19643.
212. Sellami, F., Kebiche-Senhadji, O., Marais, S., Couvrat, N., Fatyeyeva, K. Polymer inclusion membranes based on CTA/PBAT blend containing Aliquat 336 as extractant for removal of Cr(VI): Efficiency, stability and selectivity. *Reactive and Functional Polymers*. 2019. 139: 120-132.

213. Baczyńska, M., Waszak, M., Nowicki, M., Prządka, D., Borysiak, S., Regel-Rosocka, M. Characterisation of Polymer Inclusion Membranes (PIMs) Containing Phosphonium Ionic Liquids as Zn(II) Carriers. *Industrial & Engineering Chemistry Research*. 2018. 57(14): 5070-5082.
214. Kebiche-Senhadji, O., Mansouri, L., Tingry, S., Seta, P., Benamor, M. Facilitated Cd(II) transport across CTA polymer inclusion membrane using anion (Aliquat 336) and cation (D2EHPA) metal carriers. *Journal of Membrane Science*. 2008. 310(1-2): 438-445.
215. Sellami, F., Kebiche-Senhadji, O., Marais, S., Colasse, L., Fatyeyeva, K. Enhanced removal of Cr(VI) by polymer inclusion membrane based on poly(vinylidene fluoride) and Aliquat 336. *Separation and Purification Technology*. 2020. 248: 117038.
216. Neise, C., Rautenberg, C., Bentrup, U., Beck, M., Ahrenberg, M., Schick, C., Keßler, O., Kragl, U. Stability studies of ionic liquid [EMIm][NTf_2] under short-term thermal exposure. *RSC Advances*. 2016. 6(54): 48462-48468.
217. Chen, Y., Niu, Y., Gong, P., Xiao, Z., Li, G. Thermally Reversible and Irreversible Phase Transition Behaviors in Poly(ethylene oxide)/Ionic Liquid Mixtures. *Macromol Rapid Commun*. 2017. 38(23): 1700401.
218. Fontàs, C., Tayeb, R., Dhahbi, M., Gaudichet, E., Thominette, F., Roy, P., Steenkeste, K., Fontaine-Aupart, M.-P., Tingry, S., Tronel-Peyroz, E. Polymer inclusion membranes: The concept of fixed sites membrane revised. *Journal of Membrane Science*. 2007. 290(1-2): 62-72.
219. Iben Nasser, I., Ibn El Haj Amor, F., Donato, L., Algieri, C., Garofalo, A., Drioli, E., Ahmed, C. Removal and recovery of $Ag(CN)^{2-}$ from synthetic electroplating baths by polymer inclusion membrane containing Aliquat 336 as a carrier. *Chemical Engineering Journal*. 2016. 295: 207-217.
220. Rodríguez de San Miguel, E., Aguilar, J. C., de Gyves, J. Structural effects on metal ion migration across polymer inclusion membranes: Dependence of transport profiles on nature of active plasticizer. *Journal of Membrane Science*. 2008. 307(1): 105-116.
221. Ghiculesco, R. A. Abnormal laboratory results: Therapeutic drug monitoring: which drugs, why, when and how to do it. *Australian Prescriber*. 2008. 31(2): 42-44.
222. Maiphetlho, K., Shumbula, N., Motsoane, N., Chimuka, L., Richards, H. Evaluation of silver nanocomposite polymer inclusion membranes (PIMs) for trace metal transports: Selectivity and stability studies. *Journal of Water Process Engineering*. 2020. 37: 101527.
223. Wang, B., Li, Z., Lang, Q., Tan, M., Ratanatamskul, C., Lee, M., Liu, Y., Zhang, Y. A comprehensive investigation on the components in ionic liquid-based polymer inclusion membrane for Cr(VI) transport during electrodialysis. *Journal of Membrane Science*. 2020. 604: 118016.
224. Li, Y.-H., Wang, D.-M., Hsien, T.-Y., Chan, K.-Y., Lai, J.-Y. Polymer Inclusion Membranes with Strip Dispersion. *Water*. 2017. 9(6): 399.
225. Gomez-Rios, G. A., Vasiljevic, T., Gionfriddo, E., Yu, M., Pawliszyn, J. Towards on-site analysis of complex matrices by solid-phase microextraction-transmission mode coupled to a portable mass spectrometer via direct analysis in real time. *Analyst*. 2017. 142(16): 2928-2935.

LIST OF PUBLICATIONS

Journal with Impact Factor

1. **Tey, H. Y**. & See, H. H. (2021). A review of recent advances in microsampling techniques of biological fluids for therapeutic drug monitoring. *J Chromatogr A, 1635*, pp. 461731. doi:10.1016/j.chroma.2020.461731 (Q1, IF: 4.759)

2. **Tey, H. Y**., Breadmore, M. C. & See, H. H. (2022). Electrokinetic Extraction of Drug Analytes from Biological Fluids by Polymer Inclusion Membrane (PIM). *Analytical Chmistry* (Submitted) (Q1, IF: 8.008)

Intellectual Property (IP)

Patents

1. See, H. H., **Tey, H. Y.**, Breadmore, M. C. & Nur H. (2021). *Polymer Inclusion Membrane Sampling Probe for Solvent-less Electric Field Driven Extraction, Methods and Assembly Thereof* (Patent No: PI2021006275)

Conference Presentations

1. **Tey, H. Y.** & See, H. H., "Solid Sampling Probe based on a Polymer Inclusion Membrane." Oral presentation at 36th International Symposium on Microscale Separations and Bioanalysis (e-MSB 2020). September 27 – 30, 2020. Virtual symposium organised by The Society for Microscale Separations and Bioanalysis (SMSB).

2. **Tey, H. Y.** & See, H. H., "Polymer Inclusion Membrane (PIM) Sampling Probe for Electric Field Driven Extraction of Drug Analytes from Biological Fluids." Oral presentation at 38th International Symposium on Microscale Separations and Bioanalysis (MSB 2022). Liège, Belgium, July 3 – 6, 2022. Symposium organised by The Society for Microscale Separations and Bioanalysis (SMSB).

3. **Tey, H. Y.** & See, H. H., "Polymer Inclusion Membrane Sampling Probe for Electric Field Driven Extraction of Drug Analytes from Biological Fluids." Oral presentation at 2022 AOAC SEA 1st Annual Conference, Singapore, October 12 – 13, 2022. Annual Meeting organised by AOAC SEA Section.