

MICRO-SOLID PHASE EXTRACTION METHODS  
BASED ON MOLECULARLY IMPRINTED POLYMER FOR  
DETERMINATION OF TOLUENE METABOLITES IN URINE  
SAMPLES

MOHAMAD RAIZUL BIN ZINALIBDIN

A thesis submitted in fulfilment of the  
requirements for the award of the degree of  
Doctor of Philosophy

Faculty of Science  
Universiti Teknologi Malaysia

JANUARY 2019

## DEDICATION

Dedicated goes to

My mother and father, **Madianah Binti Abu and Zinalibdin Bin Abdul Rahim**, whose sacrifice, prayer;

My **Brothers and sisters**, whose support and encouragement;

And

My wife, **Tity Ernie Suryati Binti Ahmad**, My Children, **Nur Insyirah Binti Mohamad Raizul, Muhammad Fahimy Bin Mohamad Raizul, Nur Sumayyah Binti Mohamad Raizul** whose patience;

Lead me to achieve my doctoral degree.

Not forgotten My late son, **Muhammad Raqimy Bin Mohamad Raizul**, your memory strengthens my soul

## **ACKNOWLEDGEMENT**

First and foremost, all praise is due to Allah (SWT), the Almighty for with His mercy has given me the strength and time to complete this work.

I would like to express my sincere gratitude to my supervisor and co-supervisors, Assoc. Prof. Dr. Jafariah Jaafar, Assoc. Prof. Dr. Zaiton Abdul Majid and Prof. Dr. Mohd Marsin Sanagi for their guidance and endless supports throughout the study, the advice, inspiration, scientific excitement and unwavering patient that enabled me to approach this PhD work positively.

It is with deep appreciation and heartfelt thanks to my beloved family, whose value to me only grows with time. Their overwhelming displays of love and encouragement have given me the strength to complete my research and thesis writing. For my helpful and compassionate friends who have always been there for me especially during my 'recovery moments', thank you isn't enough to express how grateful I am. May Allah bless you all! Not forgetting the staff of the Department of Chemistry, Faculty of Science, UTM, I would like to extend my complete gratitude to them for their prompt and timely helps.

I am also indebted to the Public Services Department Malaysia (Jabatan Perkhidmatan Awam, Malaysia) for the financial support and Universiti Teknologi Malaysia for the Grant for Research University (GUP). Special thanks to UTM's library in supplying free databases and e-Journal. Finally, yet importantly I would like to thank everyone who has directly or indirectly contributed to the completion of my graduate study.

## ABSTRACT

Toluene is one of the most widely used solvents in various industries. Workers exposure to toluene can be monitored using hippuric acid and *o*-cresol as urine biomarkers for biological exposure index. However, extraction of biological fluids often results in sample emulsification in liquid-liquid extraction (LLE) while also reducing the efficiency of sorbent materials in solid phase extraction (SPE). Therefore, it is imperative to solve the emulsification problem and enhance the reusability and selectivity of SPE sorbent. In this study, a new SPE method was developed using molecularly imprinted polymers (MIPs) based on hippuric acid (MISPE-HA) and *o*-cresol (MISPE-Cre). The MIPs were prepared by sol-gel polymerization of hippuric acid (0.3 mmol) or *o*-cresol (0.5 mmol), 3-propyltrimethoxymethacrylate (800  $\mu$ L), ethanol (12 mL), tetraethoxysilane (TEOS) (12 mL) and catalyst (400  $\mu$ L of hydrochloric acid). BET analysis for MISPE-HA and MISPE-Cre demonstrated surface areas of 388.64 m<sup>2</sup>/g and 574.90 m<sup>2</sup>/g, with pore volumes of 0.4638 cm<sup>3</sup>/g and 0.5314 cm<sup>3</sup>/g, respectively. Furthermore, both MISPE samples displayed pore diameters of 4.70 nm and 2.20 nm, respectively, which proved the presence of mesoporosity in their structures. MISPE-HA showed a higher adsorption capacity (9.67 mg/g) compared to MISPE-Cre (0.99 mg/g). The results indicated that the kinetics of adsorption obeyed well the pseudo-second order model and Scatchard plot for adsorption isotherm. In order to improve analyte mobility and reusability, new dispersive solid phase extraction (DISPE) methods were developed based on surface imprinted hippuric acid grafted onto silica gel (MIP@SiO<sub>2</sub>-HA) and surface imprinted *o*-cresol grafted onto silica gel (MIP@SiO<sub>2</sub>-Cre). The MIP@SiO<sub>2</sub> materials were prepared by hippuric acid (0.3 mmol) or *o*-cresol (0.5 mmol), 3-propyltrimethoxymethacrylate (800  $\mu$ L), tetraethoxysilane (10 mL), activated silica gel (0.1 g) and acetic acid (0.01 mol/L). Nitrogen adsorption showed that both MIP@SiO<sub>2</sub>-HA and MIP@SiO<sub>2</sub>-Cre were mesoporous materials with pore diameters of 4.70 nm and 4.76 nm, respectively. BET study showed that, MIP@SiO<sub>2</sub>-HA had a surface area of 122.10 m<sup>2</sup>/g and pore volume of 0.4638 cm<sup>3</sup>/g, while MIP@SiO<sub>2</sub>-Cre exhibited surface area of 106.90 m<sup>2</sup>/g and pore volume of 0.1991 cm<sup>3</sup>/g. MIP@SiO<sub>2</sub>-HA indicated a good adsorption capacity (9.68 mg/g) compared to that of MIP@SiO<sub>2</sub>-Cre, which had a low adsorption capacity of 0.99 mg/g. MIP@SiO<sub>2</sub> was also found to be fitted well with pseudo-second order model for kinetic adsorption and Scatchard plot for adsorption isotherm. The MIP@SiO<sub>2</sub> and MISPE method validation indicated good limit of detection (LOD) and good limit of quantification (LOQ) with relative standard deviation (RSD) less than 5% and high extraction recovery more than 90%, which complied with ISO 17025:2005. The results demonstrated that these adsorbents have potential applications in the determination of toluene metabolites in urine samples due to the improved selectivity, better elution ability for target molecule and high reusability.

## ABSTRAK

Toluena adalah salah satu pelarut yang digunakan secara meluas dalam pelbagai industri. Pendedahan pekerja terhadap toluena boleh dipantau menggunakan asid hipurik dan *o*-kresol sebagai biopenanda urin bagi indeks pendedahan biologi. Walau bagaimanapun, pengekstrakan cecair biologi sering mengakibatkan pengemulsian sampel dalam pengekstrakan cecair-cecair, di samping itu juga mengurangkan kecekapan bahan penjerap dalam pengekstrakan fasa pepejal (SPE). Oleh itu, adalah amat penting bagi mengatasi masalah pengemulsian dan meningkatkan kebolegunaan dan kepilihan penjerap SPE. Dalam kajian ini, kaedah SPE baharu telah dibangunkan menggunakan polimer cetakan molekul (MIP) berasaskan asid hipurik (MISPE-HA) dan *o*-kresol (MISPE-Cre). Bahan MIPs telah disediakan secara pempolimeran sol-gel asid hipurik (0.3 mmol) atau *o*-kresol (0.5 mmol), 3-propiltrimetoksietakrilat (800  $\mu$ L), etanol (12 mL), tetraetoksisilana (TEOS) (12 mL) dan mangkin (400  $\mu$ L asid hidroklorik). Analisis BET bagi MISPE-HA dan MISPE-Cre menunjukkan luas permukaan 388.64 m<sup>2</sup>/g dan 574.90 m<sup>2</sup>/g, dengan isipadu liang masing-masing sebanyak 0.4638 cm<sup>3</sup>/g dan 0.5314 cm<sup>3</sup>/g. Tambahan lagi, kedua-dua sampel MISPE memaparkan diameter liang masing-masing 4.70 nm dan 2.20 nm, yang membuktikan kehadiran mesoliang di dalam strukturnya. MISPE-HA menunjukkan kapasiti penjerapan lebih tinggi (9.67 mg/g) berbanding dengan MISPE-Cre (0.99 mg/g). Hasil kajian menunjukkan kinetik penjerapan mematuhi dengan baik model tertib pseudo-kedua dan taburan Scatchard bagi isoterma penjerapan. Bagi memperbaiki pergerakan analit dan kebolegunaan semula, kaedah baharu pengekstrakan sebaran fasa pepejal (DISPE) telah dibangunkan berdasarkan cetakan asid hipurik tercantum di permukaan gel silika (MIP@SiO<sub>2</sub>-HA) dan cetakan *o*-kresol tercantum di permukaan gel silika (MIP@SiO<sub>2</sub>-Cre). MIP@SiO<sub>2</sub> disediakan menggunakan asid hipurik (0.3 mmol) atau *o*-kresol (0.5 mmol), 3-propiltrimetoksietakrilat (800  $\mu$ L), tetraetoksisilana (10 mL), gel silika teraktif (0.1 g) dan asid asetik (0.01 mol /L). Penjerapan nitrogen menunjukkan bahawa kedua-dua MIP@SiO<sub>2</sub>-HA dan MIP@SiO<sub>2</sub>-Cre adalah bahan mesoliang dengan diameter liang masing-masing adalah 4.70 nm dan 4.76 nm. Kajian BET menunjukkan MIP@SiO<sub>2</sub>-HA mempunyai luas permukaan 122.10 m<sup>2</sup>/g dan isipadu liang 0.4638 cm<sup>3</sup>/g manakala MIP@SiO<sub>2</sub>-Cre mempamerkan luas permukaan 106.90 m<sup>2</sup>/g dan isipadu liang 0.1991 cm<sup>3</sup>/g. MIP@SiO<sub>2</sub>-HA menunjukkan kapasiti penjerapan yang baik (9.68 mg/g) berbanding dengan MIP@SiO<sub>2</sub>-Cre yang mempunyai kapasiti penjerapan yang rendah iaitu 0.99 mg/g. MIP@SiO<sub>2</sub> juga didapati sesuai dipadankan dengan tertib pseudo-kedua bagi penjerapan kinetik dan taburan Scatchard untuk isoterma penjerapan. Validasi kaedah bagi MIP@SiO<sub>2</sub> dan MISPE menunjukkan had pengesanan (LOD) dan had kuantifikasi (LOQ) yang baik dengan sisihan piawai relatif (RSD) kurang daripada 5% dan perolehan semula lebih daripada 90%, yang mematuhi standard ISO17025:2005. Keputusan menunjukkan bahan penjerap ini berpotensi diaplikasikan bagi penentuan metabolit toluena dalam sampel urin kerana kepilihan yang bertambah baik, keupayaan elusi molekul sasaran yang lebih baik dan kebolegunaan semula yang tinggi.

## CONTENT

CHAPTER	TITLE	PAGE
	<b>DECLARATION</b>	<b>ii</b>
	<b>DEDICATION</b>	<b>iii</b>
	<b>ACKNOWLEDGEMENT</b>	<b>iv</b>
	<b>ABSTRACT</b>	<b>v</b>
	<b>ABSTRAK</b>	<b>vi</b>
	<b>CONTENT</b>	<b>vii</b>
	<b>LIST OF TABLES</b>	<b>xxi</b>
	<b>LIST OF FIGURES</b>	<b>xxvii</b>
	<b>LIST OF ABBREVIATION</b>	<b>xl</b>
	<b>LIST OF SYMBOLS</b>	<b>xliv</b>
	<b>LIST OF APPENDIXES</b>	<b>xlvi</b>
<b>1</b>	<b>INTRODUCTION</b>	<b>1</b>
	1.1 Background of Research	1
	1.2 Problem Statements	5
	1.3 Objective of Research	7
	1.4 Scopes of Research	8
	1.5 Significance of Research	10
<b>2</b>	<b>LITERATURE REVIEW</b>	<b>13</b>
	2.1 Introduction	13
	2.2 Industrialization exposure impact	16

2.2.1	Occupational exposure limit	19
2.2.1.1	Occupational exposure limit for toluene	21
2.2.1.2	Biological exposure index	23
2.3	Toluene and its metabolites	25
2.3.1	Metabolism and potential biomarker historical background	25
2.3.2	Toluene metabolism	27
2.3.3	Hippuric acid sample pre-treatment prior to HPLC	31
2.3.4	Hippuric acid sample pre-treatment and derivatization procedure prior to gas chromatography	32
2.3.5	Cresol pre-treatment prior to gas chromatography	34
2.3.6	Analysis by High Performance Liquid Chromatography	35
2.3.7	Analysis by Gas Chromatography	45
2.4	Molecularly Imprinted Polymer	54
2.4.1	Molecularly imprinting approach	55

2.4.2	Synthesize method of MIPs	59
2.5	Sol-gel Polymerization	65
2.5.1	Application of the MIP using sol-gel method	70
2.6	Surface Imprinting	74
2.6.1	Application of the surface imprinting	76
2.7	Summary	77
<b>3</b>	<b>EXPERIMENTAL</b>	<b>80</b>
3.1	Operational research framework	80
3.2	Reagents and Apparatus	82
3.2.1	Reagents	82
3.2.2	Apparatus	83
	3.2.2.1 HPLC-UV	83
	3.2.2.2 GC-FID	83
3.3	Preparation of molecularly imprinted polymer solid phase extraction (MISPE)	84
3.3.1	MISPE hippuric acid template molecule (MISPE- HA)	84
3.3.2	MISPE <i>o</i> -cresol template molecule (MISPE-Cre)	85
3.3.3	Characterization of MISPE- HA, MISPE-Cre, NISPE- HA and NISPE-Cre	86



3.3.3.1	Fourier Transform Infrared (FTIR)	86
3.3.3.2	Thermal-gravimetry analysis- Differential thermal analysis (TGA- DTA)	87
3.3.3.3	Nitrogen Adsorption	87
3.3.3.4	X-ray diffraction analysis (XRD)	87
3.3.3.5	Field Emission Scanning Electron Microscopy-Energy Dispersive X-ray (FESEM-EDX)	88
3.3.4	Binding experiment for MISPE-HA and MISPE-Cre	88
3.3.4.1	Optimization of solid to liquid ratio	88
3.3.4.2	Optimum pH environment	89
3.3.4.3	Batch adsorption experiment	89
3.3.4.4	Kinetic Adsorption	91
3.3.4.5	Selectivity experiment	91

3.3.4.6	Reusability experiment	93
3.3.4.7	Solid phase extraction studies via MISPE	94
3.3.5	Method verification and validation of MISPE	95
3.3.5.1	MISPE hippuric acid as a template molecule	95
3.3.5.2	MISPE <i>o</i> -cresol as a template molecule	96
3.4	Preparation of surface imprinted grafted onto silica gel (MIP@SiO <sub>2</sub> )	97
3.4.1	Silica gel activation	97
3.4.2	Synthesize of surface imprinted hippuric acid grafted onto silica gel (MIP@SiO <sub>2</sub> -HA)	98
3.4.3	Synthesize of surface imprinted <i>o</i> -cresol grafted onto silica gel (MIP@SiO <sub>2</sub> - Cre)	98
3.4.4	Characterization of MIP@SiO <sub>2</sub> -HA and MIP@SiO <sub>2</sub> -Cre	99

3.4.5	Binding experiment for MIP@SiO <sub>2</sub> -HA and MIP@SiO <sub>2</sub> -Cre	101
3.4.5.1	Optimization of several parameter for MIP@SiO <sub>2</sub> -HA, NIP@SiO <sub>2</sub> -HA, MIP@SiO <sub>2</sub> -Cre and NIP@SiO <sub>2</sub> -Cre	101
3.4.5.2	Batch adsorption experiment	102
3.4.5.3	Kinetic Adsorption	103
3.4.5.4	Selectivity experiment	104
3.4.5.5	Reusability experiment	105
3.4.5.6	DISPE via MIP@SiO <sub>2</sub>	106
3.4.6	Method verification and validation of MIP@SiO <sub>2</sub>	108
3.4.6.1	MIP@SiO <sub>2</sub> hippuric acid as a template molecule	108
3.4.6.2	MIP@SiO <sub>2</sub> <i>o</i> -cresol as a template molecule	109
3.5	Sampling of Real sample	110
3.5.1	Sampling at Company A	110

3.5.2	Sampling at Department B	112
3.6	Analysis of Real sample	113
3.6.1	Hydrolyse of cresol metabolites	113
3.6.2	SPE	113
3.6.3	DISPE	114
<b>4</b>	<b>MOLECULARLY IMPRINTED POLYMER SOLID PHASE EXTRACTION FOR EXTRACTION OF HIPPURIC ACID AND O-CRESOL IN URINE</b>	<b>116</b>
4.1	Introduction	116
4.2	Result and Discussion of Molecularly imprinted polymer solid phase extraction for hippuric acid (MISPE- HA)	119
4.2.1	Preparation of MISPE-HA	119
4.2.2	Characterization of MISPE- HA, NISPE-HA and unleached MISPE-HA	122
4.2.2.1	Fourier transform infrared (FTIR)	122
4.2.2.2	Thermal-gravimetry- Differential thermal analysis (TGA- DTA)	125

4.2.2.3	Nitrogen Adsorption	127
4.2.2.4	X-ray diffraction analysis (XRD)	130
4.2.2.5	Field Emission Electron Microscopy (FESEM)	132
4.2.2.6	Energy Dispersive X-ray (EDX)	134
4.2.3	Binding studies of MISPE-HA and NISPE-HA	135
4.2.3.1	Optimization of solid-liquid ratio	135
4.2.3.2	Optimization of pH environment	137
4.2.3.3	Effect of the hippuric acid concentration on the adsorption process	139
4.2.3.4	Effect of the contact time	141
4.2.3.5	Kinetic study on adsorption of hippuric acid by MISPE-HA and NISPE-HA	142

4.2.3.5.1	Pseudo first order kinetic model	142
4.2.3.5.2	Pseudo second order kinetic model	145
4.2.3.6	Adsorption Isotherm	147
4.2.3.6.1	Langmuir isotherm model	148
4.2.3.6.2	Freundlich isotherm model	151
4.2.3.6.3	Scatchard Plot	153
4.2.3.7	Selectivity experiment	156
4.2.3.8	Reusability analysis	157
4.2.3.9	SPE for MISPE-HA	158
4.2.3.9.1	Elution solvent	158
4.2.3.9.2	Effect of solvent volume	159
4.2.4	Validation of proposed method	160
4.2.5	Recovery	161

4.2.6	Real urine sample analysis	162
4.3	Result and discussion of Molecularly imprinted polymer solid phase extraction for <i>o</i> -cresol (MISPE-Cre)	168
4.3.1	Preparation of MISPE-Cre	168
4.3.2	Characterization of MISPE-Cre, NISPE-Cre and unleached MISPE-Cre	171
4.3.2.1	FTIR	171
4.3.2.2	TGA-DTA	173
4.3.2.3	Nitrogen adsorption	175
4.3.2.4	XRD	176
4.3.2.5	FESEM	177
4.3.2.6	EDX	179
4.3.3	Binding studies of NISPE-Cre and MISPE-Cre	180
4.3.3.1	Optimization of solid liquid ratio	180
4.3.3.2	Optimization of pH environment	182
4.3.3.3	Effect of the <i>o</i> -cresol concentration on the adsorption process	183
4.3.3.4	Effect of the contact time	184
4.3.3.5	Kinetic study on adsorption of <i>o</i> -cresol	185

	by MISPE-Cre and NISPE-Cre	
4.3.3.5.1	Pseudo first order kinetic model	186
4.3.3.5.2	Pseudo second order kinetic model	188
4.3.3.6	Adsorption Isotherm	190
4.3.3.6.1	Langmuir isotherm model	190
4.3.3.6.2	Freundlich isotherm model	192
4.3.3.6.3	Scatchard Plot	194
4.3.3.7	Selectivity experiment	196
4.3.3.8	Reusability analysis	198
4.3.3.9	Solid phase extraction	199
4.3.3.9.1	Elution solvent	199
4.3.3.9.2	Effect of solvent volume	200



4.3.4	Validation of proposed method	202
4.3.5	Recovery	203
4.3.6	Real urine sample analysis	203
4.4	Summary	207
<b>5</b>	<b>MIP@SiO<sub>2</sub> via SURFACE IMPRINTED POLYMER GRAFTED ONTO SILICA GEL ADSORBENT FOR EXTRACTION OF HIPPURIC ACID AND O-CRESOL IN URINE</b>	<b>212</b>
5.1	Introduction	212
5.2	Result and Discussion of Surface imprinted hippuric acid grafted onto silica gel (MIP@SiO <sub>2</sub> -HA)	215
5.2.1	Preparation of MIP@SiO <sub>2</sub> -HA	215
5.2.2	Characterization of MIP@SiO <sub>2</sub> - HA, NIP@SiO <sub>2</sub> -HA and unleached MIP@SiO <sub>2</sub> -HA	219
5.2.3	Binding studies of MISPE-HA and NISPE-HA	229
5.2.3.1	Optimization of several parameters of MIP@SiO <sub>2</sub> -HA and NIP@SiO <sub>2</sub> -HA	229
5.2.3.2	Kinetic study on adsorption of hippuric acid by MIP@SiO <sub>2</sub> -HA and NIP@SiO <sub>2</sub> -HA	233

5.2.3.3	Adsorption Isotherm	236
5.2.3.4	Selectivity experiment	243
5.2.3.5	Reusability analysis	244
5.2.3.6	DISPE for MIP@SiO <sub>2</sub> - HA	246
5.2.4	Validation of proposed method	248
5.2.5	Recovery	249
5.2.6	Real urine sample analysis	249
5.3	Result and discussion of Surface imprinted <i>o</i> -cresol grafted onto silica gel (MIP@SiO <sub>2</sub> -Cre)	255
5.3.1	Preparation of MIP@SiO <sub>2</sub> -Cre	255
5.3.2	Characterization of MIP@SiO <sub>2</sub> - Cre, NIP@SiO <sub>2</sub> -Cre and unleached MIP@SiO <sub>2</sub> -Cre	258
5.3.3	Binding studies of MIP@SiO <sub>2</sub> - Cre and NIP@SiO <sub>2</sub> -Cre	265
5.3.3.1	Optimization of several parameters of MIP@SiO <sub>2</sub> -Cre and NIP@SiO <sub>2</sub> -Cre	265
5.3.3.2	Kinetic study on adsorption of <i>o</i> -cresol by MIP@SiO <sub>2</sub> -Cre and NIP@SiO <sub>2</sub> -Cre	269
5.3.3.3	Adsorption Isotherm	271
5.3.3.4	Selectivity experiment	275
5.3.3.5	Reusability analysis	277

5.3.3.6	DISPE for MIP@SiO <sub>2</sub> - Cre	278
5.3.4	Validation of proposed method	281
5.3.5	Recovery	281
5.3.6	Real urine sample analysis	282
5.4	Summary	285
<b>6</b>	<b>COMPARISON THE DEVELOPED EXTRACTION METHODS AND PREVIOUS METHODS</b>	<b>289</b>
6.1	Comparison between MISPE and MIP@SiO <sub>2</sub>	289
6.2	Comparison between previous methods for determination of hippuric acid	291
6.3	Comparison between previous methods for determination of <i>o</i> -cresol	293
<b>7</b>	<b>CONCLUSION AND RECOMMENDATION</b>	<b>295</b>
7.1	Conclusion	295
7.2	Recommendation	300
	<b>REFERENCES</b>	<b>302</b>
	Appendices A -B	327-330

**LIST OF TABLES**

<b>TABLE NO</b>	<b>TITLES</b>	<b>PAGE</b>
2.1	Occupational exposure limit and recommendation exposure limit of United States (Administration, Occupational, safety and Health, Department of Labor, United States)	21
2.2	ACGIH Toluene Biological Exposure Index (BEI) Information	24
2.3	Condition sample pre-treatment and HPLC separation of toluene metabolites in biological samples	40
2.4	Condition for sample pre-treatment and GC separation for toluene metabolites in biological sample	48
2.5	Molecularly imprinted polymer imprinting approach	56
2.6	Method of synthesis molecularly imprinted polymers	60
3.1	Sampling point urine sample for Company A	112
3.2	Sampling point urine sample for Department B	112

4.1	The adsorption capacity and the imprint factor of the prepared polymer using different amounts of monomer and ethanol (12 mL) as the reaction solvent and TEOS (12 mL) as the cross-linker	122
4.2	Surface area and pore analysis of NISPE, unleached MISPE and MISPE by nitrogen adsorption	130
4.3	Peaks assignment in the XRD patterns of unleached MISPE-HA	132
4.4	Percentage of carbon, silicon and oxygen in NISPE-HA, unleached MISPE-HA and MISPE-HA	134
4.5	Pseudo-first-order constants and coefficients of determination by MISPE-HA and NISPE-HA	144
4.6	Pseudo-second-order constants and coefficients of determination by MISPE and NISPE	147
4.7	Langmuir isotherm constants and coefficients of determination for hippuric acid study at $298 \pm 2$ K	150
4.8	Freundlich isotherm constants and coefficients of determination for hippuric acid at $298 \pm 2$ K	153
4.9	Recognition properties of MISPE-HA and NISPE-HA	156

4.10	The LOD, LOQ and RSD values of hippuric acid via extraction using MISPE-HA	161
4.11	Analytical results for hippuric acid in blank urine samples	162
4.12	HPLC-UV analysis of MISPE-HA extraction of hippuric acid in urine	163
4.13	The adsorption capacity and the imprint factor of the prepared polymer using different amounts of monomer and ethanol (12 mL) as the reaction solvent and TEOS (12 mL) as the cross-linker	170
4.14	Surface area and pore analysis of NISPE-Cre, unleached MISPE-Cre and MISPE-Cre Leached by nitrogen adsorption analysis	176
4.15	Percentage of carbon, silicon and oxygen in NISPE-Cre, unleached MISPE-Cre and MISPE-Cre leached	179
4.16	Pseudo-first-order constants and coefficients of determination by MISPE-Cre and NISPE-Cre	187
4.17	Pseudo-second-order constants and coefficients of determination by MISPE-Cre and NISPE-Cre	190
4.18	Langmuir isotherm constants and coefficients of determination for <i>o</i> -cresol study at $298 \pm 2$ K	192

4.19	Freundlich isotherm constants and coefficients of determination for <i>o</i> -cresol at $298 \pm 2$ K	194
4.20	Recognition properties of MISPE-Cre and NISPE-Cre	196
4.21	The LOD, LOQ and RSD values of <i>o</i> -cresol via extraction using MISPE-Cre	202
4.22	Analytical results for <i>o</i> -cresol in blank urine samples	203
4.23	GC-FID analysis of MISPE-Cre extraction of hippuric acid in urine	204
5.1	The adsorption capacity and the imprint factor of the prepared polymer using different amounts of monomer, ethanol (12 mL) as the reaction solvent and TEOS (12 mL) as the cross-linker	218
5.2	Surface area and pore analysis of NIP@SiO <sub>2</sub> -HA, MIP@SiO <sub>2</sub> -HA unleached and MIP@SiO <sub>2</sub> -HA by nitrogen adsorption	224
5.3	XRD peaks assignment for unleached MIP@SiO <sub>2</sub>	226
5.4	Percentage of carbon, silicon and oxygen in activated silica gel, NIP@SiO <sub>2</sub> -HA, unleached MIP@SiO <sub>2</sub> -HA and MIP@SiO <sub>2</sub> -HA	228
5.5	Comparison of the pseudo-first-order and pseudo-second-order adsorption rate	236

	constants, initial adsorption rate and calculated and experimental $q_e$ values for MIP@SiO <sub>2</sub> -HA and NIP@SiO <sub>2</sub> -HA	
5.6	Isotherm constants of three isotherm models for hippuric acid adsorption onto MIP@SiO <sub>2</sub> -HA and NIP@SiO <sub>2</sub> -HA	242
5.7	Recognition properties of MIP@SiO <sub>2</sub> -HA and NIP@SiO <sub>2</sub> -HA	243
5.8	The LOD, LOQ and RSD values of hippuric acid via extraction using MIP@SiO <sub>2</sub> -HA	248
5.9	Analytical results for hippuric acid in blank urine samples	249
5.10	HPLC-UV analysis of MIP@SiO <sub>2</sub> -HA extraction of hippuric acid in urine	250
5.11	The adsorption capacity and the imprint factor of the prepared polymer using different amounts of monomer, ethanol (10 mL) as the reaction solvent and TEOS (4 mmol) as the cross-linker	257
5.12	Surface area and pore analysis of activated silica, NIP@SiO <sub>2</sub> -Cre, unleached MIP@SiO <sub>2</sub> -Cre and MIP@SiO <sub>2</sub> -Cre leached by nitrogen adsorption analysis	261
5.13	Percentage of carbon, silicon and oxygen in activated silica gel, NIP@SiO <sub>2</sub> -Cre, unleached MIP@SiO <sub>2</sub> -Cre and MIP@SiO <sub>2</sub> -Cre leached	265



5.14	Comparison of the pseudo-first-order and pseudo-second-order adsorption rate constants, initial adsorption rate and calculated and experimental $q_e$ values for MIP@SiO <sub>2</sub> -Cre and NIP@SiO <sub>2</sub> -Cre	271
5.15	Isotherm constants of three isotherm models for <i>o</i> -cresol adsorption onto MIP@SiO <sub>2</sub> -Cre and NIP@SiO <sub>2</sub> -Cre	274
5.16	Recognition properties of MIP@SiO <sub>2</sub> -Cre and NIP@SiO <sub>2</sub> -Cre	276
5.17	The LOD, LOQ and RSD values of <i>o</i> -cresol via extraction using MIP@SiO <sub>2</sub> -Cre	281
5.18	Analytical results for <i>o</i> -cresol in blank urine samples	282
5.19	GC-FID analysis of MIP@SiO <sub>2</sub> -Cre extraction of <i>o</i> -cresol in urine	283
6.1	Comparison between MISPE-HA, MISPE-Cre, MIP@SiO <sub>2</sub> -HA and MIP@SiO <sub>2</sub> -Cre for binding study and optimization of extraction method	290
6.2	Determination of hippuric acid using different methods	292
6.3	Determination of <i>o</i> -cresol using different methods	293

**LIST OF FIGURES**

<b>FIGURE NO</b>	<b>TITLES</b>	<b>PAGE</b>
2.1	Metabolism of toluene (Inoue <i>et al.</i> , 2004)	30
2.2	Sol-gels process: step 1 illustrates hydrolysis occurring at room temperature while condensation follows in step 2 at 105°C. R', R'' and R''' are functionalities on the starting monomers	68
2.3	Surface imprinted polymerization technique	75
3.1	Operational research framework	81
3.2	Molecularly imprinted polymer solid phase extraction procedure	95
3.4	DISPE for extraction of hippuric acid via MIP@SiO <sub>2</sub> -HA and o-cresol by MIP@SiO <sub>2</sub> -Cre	108
4.1	Synthesis of hippuric acid molecularly imprinted solid phase extraction (Bagheri and Piri-Moghadam, 2012; Moein <i>et al.</i> , 2014a)	121

4.2	FTIR absorption spectra for (a) NISPE-HA (b) Unleached MISPE-HA and (c) MISPE-HA	124
4.3	Thermal gravimetry and Differential thermal analysis of (a) NISPE-HA (b) MISPE-HA and (c) Unleached MISPE-HA	126
4.4	Nitrogen adsorption isotherms of (a) NISPE-HA, (c) MISPE-HA and (e) Unleached MISPE-HA and pore size distribution curves were plotted from BJH desorption for (b) NISPE-HA, (d) MISPE-HA and (f) Unleached MISPE-HA	129
4.5	XRD diffractogram for (a) NISPE-HA, (b) MISPE-HA and (c) Unleached MISPE-HA	131
4.6	Field Emission Scanning Electron Micrographs of (a) NISPE-HA, (b) Unleached MISPE-HA d and (c) MISPE-HA	133
4.7	Effect of solid-liquid ratio on the sorption capacity of MISPE-HA and NISPE-HA	135
4.8	Effect of pH on the adsorption capacity of hippuric acid on NISPE-HA and MISPE-HA	139

4.9	Effect of initial concentration on the adsorption of hippuric acid onto MISPE-HA and NISPE-HA (adsorption conditions: adsorbent dose= 50 mg, solution volume= 10 mL, time = 90 min, pH= 3)	140
4.10	Effect of contact time on the adsorption capacity of hippuric acid by MISPE-HA and NISPE-HA	141
4.11	Pseudo-first order kinetic plot of hippuric acid adsorption onto MISPE-HA at 50 mg/L concentration	143
4.12	Pseudo-first order kinetic plot of hippuric acid adsorption onto NISPE-HA at 50 mg/L concentration	144
4.13	Pseudo-second-order kinetics plot of hippuric acid adsorption onto MISPE-HA at 50 mg/L concentration and $298 \pm 2$ K	145
4.14	Pseudo-second-order kinetics plot of hippuric acid adsorption onto NISPE-HA at 50 mg/L concentration and $298 \pm 2$ K	146
4.15	Langmuir isotherm plot of adsorption of hippuric acid onto NISPE-HA at 298 K	149

4.16	Langmuir isotherm plot of adsorption of hippuric acid onto MISPE-HA at 298 K	149
4.17	Freundlich isotherm plot of adsorption of hippuric acid onto NISPE-HA at $298 \pm 2$ K	152
4.18	Freundlich isotherm plot of adsorption of hippuric acid onto MISPE-HA at $298 \pm 2$ K	152
4.19	Scatchard plots for binding of hippuric acid to MISPE-HA at $298 \pm 2$ K	154
4.20	Scatchard plots for binding of hippuric acid to NISPE-HA at $298 \pm 2$ K	155
4.21	Absorption-desorption cycles of hippuric acid- MISPE-HA	158
4.22	Effect of organic solvent elution on NISPE-HA and MISPE-HA analysis. Conditions: 50 mg of absorbents; conditioning solvent: 1 mL methanol, 1 mL acetonitrile and 1 mL of water and adjusted to pH 3; washing step: 3 mL water; no salt addition	159
4.23	Effect of solvent volume on NISPE-HA and MISPE-HA analysis. Conditions: 50 mg of MISPE-HA or NISPE-HA; conditioning solvent: 1	160

- mL methanol, 1 mL acetonitrile and 1 mL water and adjusted to pH 3; washing step: 3 mL water; elution solvent: 3 mL of methanol, no salt addition
- 4.24 HPLC chromatogram of 50 mg/L hippuric acid standard. Conditions: 150 mm x 2.0 mm i.d. C18 column, isocratic mode of distilled water, acetonitrile and acetic acid (84:16:0.025), mobile phase flow rate at 1.0 mL/min and UV detection at 225 nm 164
- 4.25 HPLC chromatogram of positive urine sample No.1, 2 and 3 from company A. Conditions: 150 mm x 2.0 mm i.d. C18 column, isocratic mode of distilled water, acetonitrile and acetic acid (84:16:0.025), mobile phase flow rate at 1.0 mL/min and UV detection at 225 nm 165
- 4.26 HPLC chromatogram of positive urine sample No.14, 15, 22, 23 and 34 from Department B. Conditions: 150 mm x 2.0 mm i.d. C18 column, isocratic mode of distilled water, acetonitrile and acetic acid (84:16:0.025), mobile phase flow rate at 1.0 mL/min and UV detection at 225 nm 167

4.27	Synthesis of <i>o</i> -cresol molecularly imprinted solid phase extraction (Bagheri and Piri-Moghadam, 2012)	169
4.28	FTIR spectrum for (a) NISPE-Cre, (b) Unleached MISPE-Cre and (c) MISPE-Cre	172
4.29	Thermal gravimetry and Differential thermal analysis of (a) NISPE-Cre (b) MISPE-Cre and (c) Unleached MISPE-Cre	174
4.30	XRD diffractogram for (a) NISPE-Cre, (b) MISPE-Cre and (c) Unleached MISPE-Cre	177
4.31	FESEM of (a) Unleached MISPE-Cre, (b) MISPE-Cre leached and (c) NISPE-Cre	178
4.32	Adsorption properties in five different solid-liquid ratio for MISPE-Cre and NISPE-Cre	180
4.33	Function of pH values on equilibrium adsorption amount	183
4.34	Adsorption isotherm of MISPE-Cre and NISPE-Cre toward <i>o</i> -cresol	184
4.35	Effect of contact time on the adsorption capacity of <i>o</i> -cresol by MISPE-Cre and NISPE-Cre	185

4.36	Pseudo-first order kinetic plot of <i>o</i> -cresol adsorption onto MISPE-Cre at 2.0 mg/L concentration	186
4.37	Pseudo-first order kinetic plot of <i>o</i> -cresol adsorption onto NISPE-Cre at 2.0 mg/L concentration	187
4.38	Pseudo-second-order kinetics plot of <i>o</i> -cresol adsorption onto MISPE-Cre at 2 mg L <sup>-1</sup> concentration and 298 ± 2 K	188
4.39	Pseudo-second-order kinetics plot of <i>o</i> -cresol adsorption onto NISPE-Cre at 2 mg L <sup>-1</sup> concentration and 298 ± 2 K	189
4.40	Langmuir isotherm plot of adsorption of <i>o</i> -cresol onto MISPE-Cre at 298 K	191
4.41	Langmuir isotherm plot of adsorption of <i>o</i> -cresol onto NISPE-Cre at 298 K	191
4.42	Freundlich isotherm plot of adsorption of <i>o</i> -cresol onto MISPE-Cre at 298 ± 2 K	193
4.43	Freundlich isotherm plot of adsorption of <i>o</i> -cresol onto NISPE-Cre at 298 ± 2 K	193
4.44	Scatchard plots for binding of <i>o</i> -cresol to MISPE-Cre at 298 ± 2 K	195
4.45	Scatchard plots for binding of <i>o</i> -cresol to NISPE-Cre at 298 ± 2 K	195



4.46	Absorption-desorption cycles of <i>o</i> -cresol- MISPE-Cre	198
4.47	Effect of organic solvent elution on NISPE-Cre and MISPE-Cre analysis. Conditions: 30 mg of MISPE-Cre and NISPE-Cre; conditioning solvent: 1 mL methanol, 1 mL acetonitrile and 1 mL water and adjusted to pH 7; washing step: 3 mL water; no salt addition	200
4.48	Effect of solvent volume on NISPE-Cre and MISPE-Cre analysis. Conditions: 30 mg of MISPE-Cre or NISPE-Cre; conditioning solvent: 1 mL methanol, 1 mL acetonitrile and 1 mL water and adjusted to pH 7; washing step: 3 mL water; elution solvent: 3 mL of methanol, no salt addition	201
4.49	GC chromatogram of 2 mg/L <i>o</i> -cresol and <i>p</i> -cresol standard. GC condition: DB-Wax column, 30 m x 0.20 mm I.D x 0.20 µm film thickness. The temperature program used was as follows: 50°C hold for 1 min, 40°C/min to 210°C hold for 3 min injector temperature 210°C, carrier	205

	gas at flow rate 2 mL/min and nitrogen make-up gas at 15 mL/min	
4.50	GC chromatogram of urine sample 1, sample 2 and sample 3. GC condition: DB-Wax column, 30 m x 0.20 mm I.D x 0.20 $\mu$ m film thickness. The temperature program used was as follows: 50°C hold for 1 min, 40°C/min to 210°C hold for 3 min injector temperature 210°C, carrier gas at flow rate 2 mL/min and nitrogen make-up gas at 15 mL/min	206
5.1	MIP@SiO <sub>2</sub> of hippuric acid mechanism (Moein <i>et al.</i> , 2014a; Liao <i>et al.</i> , 2016)	217
5.2	Fourier Transform Infrared (FTIR) spectrums of (a) activated silica, (b) MIP@SiO <sub>2</sub> -HA leached, (c) NIP@SiO <sub>2</sub> -HA and (d) Unleached MIP@SiO <sub>2</sub> -HA	220
5.3	Thermal gravimetry and Differential thermal analysis of (a) NIP@SiO <sub>2</sub> -HA (b) MIP@SiO <sub>2</sub> -HA and (c) Unleached MIP@SiO <sub>2</sub> -HA	221
5.4	Nitrogen adsorption isotherms of (a) NIP@SiO <sub>2</sub> -HA, (b) Unleached MIP@SiO <sub>2</sub> -HA and (c) MIP@SiO <sub>2</sub> -HA, and pore size distribution curves	223

	were plotted from BJH desorption for (b) NIP@SiO <sub>2</sub> -HA, (d) Unleached MIP@SiO <sub>2</sub> -HA and (e) MIP@SiO <sub>2</sub> -HA	
5.5	XRD diffractogram for (a) NIP@SiO <sub>2</sub> -HA, (b) MIP@SiO <sub>2</sub> -HA and (c) Unleached MIP@SiO <sub>2</sub> -HA	225
5.6	FESEM of (a) activated silica gel (b) NIP@SiO <sub>2</sub> -HA (c) Unleached MIP@SiO <sub>2</sub> -HA and (d) MIP@SiO <sub>2</sub> -HA leached	227
5.7	Effect of the solid-liquid ratios (a), pH (b), initial concentration (c) and contact time (d) on the adsorption capacity of hippuric acid by MIP@SiO <sub>2</sub> -HA and NIP@SiO <sub>2</sub> -HA	232
5.8	Pseudo-first order kinetic plot adsorption on MIP@SiO <sub>2</sub> -HA (a), Pseudo-first order kinetic plot adsorption on NIP@SiO <sub>2</sub> -HA (b), Pseudo-second order kinetic plot adsorption on MIP@SiO <sub>2</sub> -HA (c) and Pseudo-second order kinetic plot adsorption on NIP@SiO <sub>2</sub> -HA (d) for 50 mg/L hippuric acid and 298 ± 2 K	235
5.9	Langmuir isotherm plot for (a) MIP@SiO <sub>2</sub> -HA, (b) NIP@SiO <sub>2</sub> -HA, Freundlich isotherm plot for (c)	239

	MIP@SiO <sub>2</sub> -HA and (d) NIP@SiO <sub>2</sub> -HA	
5.10	Scatchard plots for binding of hippuric acid to (a) MIP@SiO <sub>2</sub> -HA and (b) NIP@SiO <sub>2</sub> -HA	240
5.11	Adsorption-desorption cycles of hippuric acid-MIP@SiO <sub>2</sub>	245
5.12	Effect of the (a) dispersive solvents, (b) solvent volume and (c) sonication time on the extraction performance of hippuric acid 50 mg/L via MIP@SiO <sub>2</sub> -HA and NIP@SiO <sub>2</sub> -HA	247
5.13	HPLC chromatogram of positive urine sample No.1, 2 and 3 from company A. Conditions: 150 mm x 2.0 mm i.d. C18 column, isocratic mode of distilled water, acetonitrile and acetic acid (84:16: 0.025), mobile phase flow rate at 1.0 mL/min and UV detection at 225 nm	252
5.14	HPLC chromatogram of positive urine sample No.14, 15, 22, 23 and 34 from Department B. Conditions: 150 mm x 2.0 mm i.d. C18 column, isocratic mode of distilled water, acetonitrile and acetic acid (84:16: 0.025), mobile phase flow rate at 1.0 mL/min and UV detection at 225 nm	254

5.15	Scheme of the synthesis of MIP@SiO <sub>2</sub> -Cr (Liao <i>et al.</i> , 2016)	256
5.16	FTIR absorption spectra for (a) NIP@SiO <sub>2</sub> -Cre (b) Unleached MIP@SiO <sub>2</sub> -Cre and (c) MIP@SiO <sub>2</sub> -Cre	259
5.17	Thermal gravimetry and Differential thermal analysis of (a) NIP@SiO <sub>2</sub> -Cre (b) MIP@SiO <sub>2</sub> -Cre and (c) Unleached MIP@SiO <sub>2</sub> -Cre	260
5.18	XRD diffractogram for (a) NIP@SiO <sub>2</sub> -Cre, (b) MIP@SiO <sub>2</sub> -Cre and (c) unleached MIP@SiO <sub>2</sub> -Cre	262
5.19	FESEM of (a) activated silica gel (b) NIP@SiO <sub>2</sub> -Cre (c) Unleached MIP@SiO <sub>2</sub> -Cre and (d) MIP@SiO <sub>2</sub> -Cre	264
5.20	Effect of the solid-liquid ratios (a), pH (b), initial concentration (c) and contact time (d) on the adsorption capacity of hippuric acid by MIP@SiO <sub>2</sub> -Cre and NIP@SiO <sub>2</sub> -Cre	268
5.21	Pseudo-first order kinetic plot adsorption on MIP@SiO <sub>2</sub> -Cre (a), Pseudo-first order kinetic plot adsorption on NIP@SiO <sub>2</sub> -Cre (b), Pseudo-second order kinetic plot adsorption on MIP@SiO <sub>2</sub> -Cre (c) and	270

	Pseudo-second order kinetic plot adsorption on NIP@SiO <sub>2</sub> -Cre (d) for 2 mg/L <i>o</i> -cresol and 298 ± 2 K	
5.22	Langmuir isotherm plot for (a) MIP@SiO <sub>2</sub> -Cre, (b) NIP@SiO <sub>2</sub> -Cre, Freundlich isotherm plot for (c) MIP@SiO <sub>2</sub> -Cre and (d) NIP@SiO <sub>2</sub> -Cre	272
5.23	Scatchard plots for binding of <i>o</i> -cresol to (a) MIP@SiO <sub>2</sub> -Cre and (b) NIP@SiO <sub>2</sub> -Cre	273
5.24	Adsorption-desorption cycles of <i>o</i> -cresol-MIP@SiO <sub>2</sub>	278
5.25	Effect of the (a) dispersive solvents, (b) solvent volume and (c) sonication time on the extraction performance of <i>o</i> -cresol 2 mg/L via MIP@SiO <sub>2</sub> -Cre and NIP@SiO <sub>2</sub> -Cre	280
5.26	GC chromatogram of urine sample 1, sample 2 and sample 3. GC condition: DB-Wax column, 30 m x 0.20 mm I.D x 0.20 μm film thickness. The temperature program used was as follows: 50°C hold for 1 min, 40°C/min to 210°C hold for 3 min injector temperature 210°C, carrier gas at flow rate 2 mL/min and nitrogen make-up gas at 15 mL/min	284

**LIST OF ABBREVIATION**

USECHH	-	Use and Standards of Exposure of Chemicals Hazardous to Health
OSHA	-	Occupational Safety and Health Act
OEL	-	Occupational exposure limits
NIOSH	-	National Institute Occupational Safety and Health
BEI	-	Biological exposure index
PEL	-	Permissible exposure limit
ACGIH	-	American Conference of Governmental Industrial Hygienists
DOSH	-	Department of Occupational Safety and Health
SAX	-	Strong basic anion exchange
SPE	-	Solid Phase Extraction
MIP	-	Molecularly imprinted polymer
SPME	-	Solid Phase Microextraction
LLE	-	Liquid-liquid Extraction
MISPE	-	Molecularly imprinted solid phase extraction
HPLC-UV	-	High performance liquid chromatography-ultra violet
GC-FID	-	Gas chromatography- flame ionization detector
MIP@SiO <sub>2</sub>	-	Molecularly imprinted surface grafted silica gel
3-PMTMOS	-	3-(propylmethacrylate)trimethoxysilane
TEOS	-	Tetraethyl orthosilicate

FTIR	-	Fourier Transform Infra-red
FESEM-EDX	-	Field Emission Scanning Electron Microscopy- Energy dispersive X-ray
XRD	-	X-ray Diffraction
TGA-DTA	-	Thermogravimetry Analysis – Differential Thermal Analysis
BET	-	Brunauer–Emmett–Teller
NGO	-	Non-governmental association
MEK	-	Methyl ethyl ketone
AIHA	-	American Industrial Hygiene Association
TLV	-	Threshold Limit Value
SBMA	-	S-benzyl mercapturic acid
TWA	-	Time Weighted Average
WHO	-	World Health Organization
UV-Vis	-	Ultra Violet - Visible
Pyr	-	Pyridine
N.R	-	Not reported
HCl	-	Hydrochloric acid
Na <sub>2</sub> SO <sub>4</sub>	-	Sodium sulphate
HA	-	Hippuric acid
MeOH	-	Methanol
ACT	-	Acetone
ACN	-	Acetonitrile
HOAC	-	Acetic acid
NaCl	-	Sodium chloride
LLLME	-	Liquid-liquid-liquid Microextraction
CW/DVB	-	Carbowax-divinylbenzene



DI-SPME	-	Direct immersion – Solid phase microextraction
CLRP	-	Controlled/living free radical polymerization
SiO <sub>2</sub>	-	Silica oxide
APTES	-	3-aminopropyl triethoxysilane
PTMOS	-	phenyltriethoxy silane
NSAID	-	non-steroidal anti-inflammatory drug
TMSPMA	-	3-(trimethoxysilyl)propylmethacrylate
NIX	-	none imprinted xerogel
MIX	-	Molecularly imprinted xerogel
PSM	-	Polysulfone membrane
MHSM	-	Molecularly imprinted sol gel hollow fibre membrane
RSD	-	Relative standard deviation
MAA	-	Methacrylic acid
EGDMA	-	Ethylene glycol dimethacrylic
NGDA	-	Nordihydroguaiaretic acid
SMIP	-	Surface molecularly imprinted polymers
MISPE-HA	-	Molecularly imprinted polymer- hippuric acid
NISPE-HA	-	Non-imprinted polymer -hippuric acid
MISPE-Cre	-	Molecularly imprinted polymer- o-cresol
NISPE-Cre	-	Non-imprinted polymer – o-cresol
LOD	-	Limit of detection
LOQ	-	Limit of quantification
IPA	-	Isopropanol
MeOH	-	Methanol
EtOH	-	Ethanol
N <sub>2</sub>	-	Nitrogen

He	-	Helium
Na <sub>2</sub> SO <sub>4</sub>	-	Sodium sulphate
JCPDS	-	Joint Committee on Powder Diffraction Standard

**LIST OF SYMBOLS**

ppm	-	part per million
mg/L	-	milligram per liter
g/g	-	gram per gram
$\mu\text{g/L}$	-	microgram per liter
nm	-	nanometer
mg/mL	-	milligram per milliliter
mL	-	milliliter
$\mu\text{L}$	-	microliter
mg	-	milligram
mL/min	-	milliliter per minute
M	-	molar
pmol/mL	-	picomol per milliliter
cm	-	centimeter
mm	-	millimeter
$^{\circ}\text{C}$	-	degree Celsius
$\Theta$	-	theta
$\alpha$	-	alpha
$\beta$	-	beta
$\text{\AA}$	-	angstrom
$\pi$	-	pi
nmol/L	-	nanomole per liter
ng/mL	-	nanogram per milliliter
$\gamma$	-	gamma

mg/g	-	milligram per gram
$\mu\text{g}/\text{mg}$	-	microgram per milligram
$\mu\text{m}$	-	micrometer
mol/L	-	mole per liter
mmol	-	millimole
$\text{m}^2/\text{g}$	-	meter square per gram
$\text{cm}^3/\text{g}$	-	centimeter square per gram

**LIST OF APPENDICES**

<b>APPENDIX</b>	<b>TITLE</b>	<b>PAGE</b>
A	List of Publication from this Study	326
B	List of Presentations Related to this Study	328

# CHAPTER 1

## INTRODUCTION

### 1.1 Background of Research

Workplace chemical exposure has been a huge problem for most industries handling chemicals hazardous to health. Numerous chemicals has been widely used as solvent in industries electric and electronic industry, printing factory, shoe factory, chemical industry and paint industry. Among the solvents, xylene (Chen *et al.*, 1994; Ohashi *et al.*, 2006), toluene (Kawai *et al.*, 1996; Caldwell *et al.*, 2000; Cok *et al.*, 2003) and benzene (Rauscher *et al.*, 1994; Ghittori *et al.*, 1995; Inoue *et al.*, 2000) are the common solvents used. These chemicals are listed as hazardous and carcinogens under Use and Standards of Exposure of Chemicals Hazardous to Health Regulation 2000 which is enforced by Department of Occupational Safety and Health (DOSH) Malaysia (OSHA Act 514, 1994).

Occupational exposure limits (OEL) are important features for control of substances hazardous to health. Personal air sampling is routinely conducted on workers to determine the level of exposures.

These samples are collected and analyzed using validated sampling and analytical methods (NIOSH Manual of Analytical Methods, 2017). Besides that, biological exposure monitoring to chemicals in the workplace is an important component of exposure assessment. As biological monitoring is generally described as the collection of specimens of tissue or body fluid, for estimating the chemical composition of the body's internal environment, it should be employed in conjunction with ambient air monitoring. Judgments on the acceptable level of a chemical or its metabolite in biological samples are based on reference value of biological exposure index (BEI).

Toluene has been one of the most widely used organic solvent in the industries since this 21<sup>st</sup> century (Moon *et al.*, 2001; Inoue *et al.*, 2004). It has been reported that exposure to high concentrations of toluene will lead to a series of diseases such as acute and chronic respiratory effects, functional alterations of the central nervous system, mucous and dermal irritations and chromosome aberrations (Ogata *et al.*, 1969; King *et al.*, 1981; Chen *et al.*, 1994). The exposure to high concentrations of toluene has been reported by many researchers (Ogata and Taguchi, 1986, 1987; Inoue *et al.*, 1991; Ohashi *et al.*, 2006; Ukai *et al.*, 2007) by detecting toluene exposure through screening of worker's blood and urine.

Occupational Safety and Health Act (OSHA) and National Institute Occupational Safety and Health (NIOSH) of America have declared the permissible exposure limit (PEL) for toluene in general industry as 200 mg/L and 100 mg/L, respectively (National Institute

Occupational, Safety and Health, 1977). However, the American Conference of Governmental Industrial Hygienists (ACGIH) strictly aligned the OEL on toluene as 20 mg/L (American Conference of Governmental Industrial Hygienists, 1995). In Malaysia, the OEL for toluene is 50 mg/L under USECHH 2000. The current American Conference of Governmental Industrial Hygienists Biological Exposure Index (ACGIH BEI) values for toluene are 0.5 mg/L *o*-cresol and 1.6 g/g creatinine hippuric acid in post-shift urine sample. Nevertheless, BEI for toluene is not established in the Guidelines on Medical Surveillance in Malaysia but indirect measurement of creatinine value is used (Guidelines on Medical Surveillance, 2009).

It has been noted by many researchers that the main biomarkers for toluene exposure are hippuric acid and *o*-cresol. Ukai *et al.*, 2007 studied on hippuric acid and *o*-cresol as biomarker of choice for long term occupational toluene exposure. However, for a short term exposure, the better indicator in urine are unmetabolized toluene and benzylmercapturic acid. They analyzed five urine workers who were occupationally exposed to 50 mg/L toluene per hour and the results were as follows: hippuric acid 30 mg/L, *o*-cresol 15.4 µg/L, benzylmercapturic acid 1.51 µg/L, unmetabolized toluene 1.47µg/L and benzyl alcohol 0.15 µg/L. The hippuric acid and *o*-cresol are in compliance with BEI.

Blood, plasma and urine are much more complex than many other samples due to the presence of proteins, salts and various organic compounds with similar chemistry to the analytes of interest. Thus, the extraction methods for biological samples have been



difficult and complicated. In addition, the extraction of biological fluids has always been challenging, frequently produced the emulsification of the solvents and destruction of sorbent packs. Liquid-liquid extraction is a common method via a NIOSH test method (NIOSH Manual of Analytical Method 5th Edition, 2017) for toluene metabolites extraction using ethyl acetate or diethyl ether as solvent for extraction of hippuric and cresol isomer. However, this method is tedious and multiple step and emulsification occurred when the biological sample matrix is dirty. Another extraction method was established via solid phase extraction (SPE) using strong basic anion exchange (SAX) and polymerically bonded octadecyl (C18) as sorbent pack for extraction of hippuric acid and cresol isomer (Rauscher *et al.*, 1994; Bieniek, 1996). Biological fluids containing thousands of chemical and dirty sample will destroy the SPE sorbent pack. Besides that, SAX cartridges is very expensive and increase the cost of extraction. Thus, a new extraction technique should be developed using lower volume of sample and solvent complying to the green chemistry extraction.

One of the alternative ways to extract and pre-concentrate the toluene metabolites is using molecularly imprinted polymer (MIP). In addition, MIP offer higher selectivity for the analysis of complex samples. MIP is a new generic technology for the introduction of recognition properties into synthetic polymers. Nowadays, this technique has attracted attention of scientists engaged in the separation and extraction of biological fluids. This is due to the many advantages in comparison to SPE, solid phase microextraction

(SPME) and liquid-liquid extraction (LLE) which include superior stability, low cost, and ease of preparation (Moein *et al.*, 2014b).

The need of this study is to develop simple, fast and green extraction methods for determination of toluene metabolites. MIP was synthesized via sol-gel polymerization and used for a packed sorbent for molecularly imprinted solid phase extraction (MISPE). Another extraction technique was developed using synthesized molecularly surface imprinting via sol-gel polymerization grafted into silica gel (MIP@SiO<sub>2</sub>). This technique improved the recognition sensitivity, higher binding capacity, faster mass transfer and binding kinetic, and the elution ability for target molecule. Among support particles, activated silica gel shows promising characteristics due to the high stability, chemical inertness and non- swelling properties. These materials were applied to the extraction of toluene metabolites from urine sample prior to high performance liquid chromatography-ultra violet (HPLC-UV) and gas chromatography- flame ionization detector (GC-FID). The reproducibility, extraction performance, sensitivity and selectivity were investigated.

## **1.2 Problem Statements**

Rapid industrial development in Malaysia contributes to the national economic growth and prosperity. However, this development also contributes to environmental problems either in workplaces or environments. In reality, production processes in

factories nowadays are still lacking in occupational, safety and health in their working environments. Thus, the workers are easily exposed to chemical hazards, especially solvent used in the production line, due to the lack of enforcement to protect the workers in the workplace or due to the ignorance of the employer.

In Malaysia, DOSH was established to enforce the OSHA 1994 for the protection of workers in the workplace. One of the subsection of this Act describes the responsibility of employers to formulate and implement the safe system of work. Monitoring BEI via biological sample such as blood and urine is one of the mechanisms to ensure the health of workers handling chemical hazardous to health.

For monitoring BEI, urine is the simplest sample to collect from workers. However, urine itself contains a large number of chemicals and emulsification upon addition of solvent is one of the challenges for extraction of the targeted analyte in urine sample (El-Beqqali and Abdel-Rehim, 2016). Thus, a selective sorbent is required to extract analyte from urine samples prior to analysis. SPE is the most useful sample preparation method via a simple extraction step and without emulsification of sample, but it is relatively expensive, limited life-time, requires large amounts of organic solvent and carry over can be a problem (Moein *et al.*, 2014b). These problems led to the current quest by researchers for new extraction techniques.

In recent years, MIPs in extraction technique have attained considerable attention because of its ability to increase the selectivity

of the sample preparation process. MIP prepared via sol-gel polymerization have significant potential whereby this technique exhibited a lower total uptake with significantly lower non-specific binding. Besides, sol-gels do not exhibit the same degree of swelling as other MIPs prepared by other methods (Farrington and Regan, 2009). Most MIP bulk polymerization result in low binding capacity, poor sites accessibility to target molecule and irregular shape. Surface imprinting technology was developed to avoid the common limitation of bulk polymerization (Liao *et al.*, 2016). The fundamental strategy of surface imprinting was to locate the imprinting sites on the surface of materials, which improved the recognition sensitivity and the elution ability for target molecules, and the prepared MIPs produced high stability, chemical inertness and non-swelling properties.

This research is targeted to prepare the MISPE and MIP@SiO<sub>2</sub> for extraction of toluene metabolites from urine samples. The use of these materials in developing techniques for extraction, detection and degradation of pesticides has been reported (Giraldo *et al.*, 2007; Taira *et al.*, 2012; Yang *et al.*, 2013). However, to date, there has been no report on the use of these materials for extraction of toluene metabolites. Thus, this research takes an initiative to synthesis these materials for extraction of toluene metabolites.

### 1.3 Objectives of Research

The aim of this research is to prepare materials for the extraction of selected toluene metabolites, namely hippuric acid and *o*-cresol from urine samples by using MISPE and MIP@SiO<sub>2</sub>. The specific objectives of the research are the following:

1. To prepare and characterize molecularly imprinted polymer based on hippuric acid for solid phase extraction (MISPE-HA) and surface molecularly imprinted polymer based on hippuric acid grafted on silica gel (MIP@SiO<sub>2</sub>-HA) via sol-gel polymerization.
2. To develop extraction methods based on MISPE-HA and MIP@SiO<sub>2</sub>-HA combined with HPLC-UV for the determination of hippuric acid in urine samples.
3. To prepare and characterize molecularly imprinted polymer based on *o*-cresol for solid phase extraction (MISPE-Cre) and surface molecularly imprinted polymer based on *o*-cresol grafted on silica gel (MIP@SiO<sub>2</sub>-Cre) via sol-gel polymerization.
4. To develop extraction methods based on MISPE-Cre and MIP@SiO<sub>2</sub>-Cre combined with GC-FID for the determination of *o*-cresol in urine samples.

## 1.4 Scopes of the Research

This work studies on the development of dispersive solid phase extraction (DSPE) utilizing new micro-solid phase sorbents, namely MISPE and MIP@SiO<sub>2</sub>. The performances of the developed methods in the determination of toluene metabolites in urine samples were examined.

The first part of this research embarked on synthesis of MIP for sorbent pack of SPE via sol-gel polymerization using template molecule hippuric acid and *o*-cresol. In the second part of this research, molecularly imprinted polymer surface grafted on silica gel was prepared using sol-gel polymerization of hippuric acid and *o*-cresol for extraction of toluene metabolites via DSPE. Preparation of MISPE and MIP@SiO<sub>2</sub> materials were investigated.

In this research, all sorbents types were synthesized by sol-gel method. This method were chosen as it provides one step reaction process at room temperature which can generate material ready for fast shaping. In this study, 3-(propylmethacrylate)trimethoxysilane (3-PMTMOS) was used as the precursor and the results were compared to those of previously investigated precursors such as tetraethoxy silane (TEOS) and 3-(triethoxysilyl)-propylamine (Moein *et al.*, 2015b). The efficiency of adsorption for both materials were examined and calculated by the extraction of hippuric acid and *o*-cresol.

The physical properties of the materials were characterized using Fourier Transform Infra-red (FTIR), Field Emission Scanning Electrone Microscopy - Energy Dispersive X-ray analysis (FESEM-EDX) , X-ray Diffraction (XRD), Thermogravimetry Analysis-Differential Thermal analysis (TGA-DTA), Nitrogen Adsorption and Brunauer–Emmett–Teller (BET) surface area analysis.

Several important extraction parameters such as extraction time, solid to liquid ratio, pH, solvent, selectivity and repeatability were optimized. Binding kinetic, adsorption isotherm and adsorption mechanism were studied and explored to ensure the adsorption type and capacity of the materials. Moreover, the performance of the developed method were evaluated, validated and applied to the determination of these toluene metabolites in urine samples.

## 1.5 Significance of the Research

NIOSH Manual of Analytical Methods 8301 used LLE as the sample preparation method for the determination of hippuric acid, *o*-cresol and methyl hippuric acid in urine. However, this sample preparation method involved multiple steps and suffers from some drawbacks which are tedious, time consuming and poor detection limit. The results showed that *p*-cresol and *o*-cresol isomers eluted together by this method (Matsui *et al.*, 1978). Other extraction techniques used disposable SPE cartridges containing strong basic anion-exchange material (SAX), which was preconditioned with

methanol and water. Lower urine sample volume was introduced and eluted with 10% aqueous acetic acid (Rauscher *et al.*, 1994).

The urinary cresol is present in urine sample usually as sulfate or glucuronic acid conjugates. Hydrolysis and neutralization procedures are needed for changing this sulfate or glucuronic acid conjugates to cresol isomer. Acid hydrolysis was the first step encountered to produce unconjugated *o*-cresol and extracted with methylene chloride, concentrated by evaporation and the sample was analysed by gas chromatography-flame ionization detector (Truchon *et al.*, 1996). However for SPE, acid hydrolysis is still required in this step followed by adjusting the mixture to a basic condition with sodium hydroxide. A SPE cartridge was conditioned by methanol and distilled water and eluted with acetonitrile-methanol (1:1 v/v) (Bieniek, 1996).

For SPE method, the packed sorbent used for hippuric acid (SAX) and *o*-cresol (C18) are relatively expensive. Biological sample can effect the sorbent physical properties and the sorbent deteriorated after handling a few samples. It was reported this will reduce the efficiency of packed sorbent. (Moein *et al.*, 2014a). Besides that, SPE has other disadvantages such as limited life-time, high detection limit, incomplete removal of interferences and sample carry (Toulabi *et al.*, 2010).

Ukai *et al.*, 2007 reported that the urinary toluene metabolites of hippuric acid 30 mg/L, *o*-cresol 15.4 µg/L, benzylmercapturic acid 1.51 µg/L, unmetabolized toluene 1.47µg/L and benzyl alcohol 0.15



## REFERENCES

- Abd Rahim, M., Wan Ibrahim, W.A., Ramli, Z., Sanagi, M.M., and Aboul-Enein, H.Y. (2016) New Sol–Gel Hybrid Material in Solid Phase Extraction Combined with Liquid Chromatography for the Determination of Non-steroidal Anti-inflammatory Drugs in Water Samples. *Chromatographia*, **79**, 421–429. Springer Berlin Heidelberg.
- Agus Salim M.B., Malina, O, Hisanaga, N., Hirata M, zainul A. (2004) Neuropathy Due to Organic Solvent Exposure: Three Reported Cases From Pahang, Malaysia. *Journal of Occupational Safety and Health*, **1**, 19–22.
- Ahmadi, F., Asgharloo, H., Sadeghi, S., Gharehbagh-Aghababa, V., and Adibi, H. (2009) Post-derivatization procedure for determination of hippuric acid after extraction by an automated micro solid phase extraction system and monitoring by gas chromatography. *Journal of chromatography. B, Analytical technologies in the biomedical and life sciences*, **877**, 2945–51.
- AKGÜR, S.A., ÖZTÜRK, P., KURTULMUŞ, Y., KARALİ, H., and ERTÜRK, S. (2001) Medicolegal aspects of blood-urine toluene and urinary ortho-cresol concentrations in toluene exposure. *Turkish Journal of Medical Sciences*, **31**, 415–419.
- American Conference of Hygienists Governmental Industrial. (1995) Threshold limit values for chemical substances and physical

agents and biological exposure indices (BEIs). P. in: *Values, Threshold Limit*.

- Amorim, L.C.A. and Alvarez-Leite, E.M. (1997) Determination of o-cresol by gas chromatography and comparison with hippuric acid levels in urine samples of individuals exposed to toluene. *Journal of Toxicology and Environmental Health Part A*, **50**, 401–408. Taylor & Francis.
- An, F. and Gao, B. (2009) Adsorption characteristics of Cr (III) ionic imprinting polyamine on silica gel surface. *Desalination*, **249**, 1390–1396.
- Angerer, J., Schildbach, M., and Krämer, A. (1997) S-p-Toluymlcapturic acid in the urine of workers exposed to toluene: A new biomarker for toluene exposure. *Archives of Toxicology*, **72**, 119–123.
- Antti-Poika, M. (2010) Handbook of Organic Industrial Solvents.
- Ariffin M, Yatim N.I, T.N.M. (2015) Selective Surface Characteristics and Extraction Performance of a Nitro-Group Explosive Molecularly Imprinted Polymer. *Malaysian Journal of Analytical Sciences*, **19**, 574–585.
- Ashley, E.K., Ph, D., and Connor, P.F.O. (2016) *NIOSH Manual of Analytical Methods ( NMAM )*, 5th Edition Forew o rd. P. in: *NIOSH Manual of Analytical Methods*. 1-860 pp.
- Asman, S., Mohamad, S., and Sarih, N.M. (2015) Exploiting F-Cyclodextrin in molecular imprinting for achieving recognition of benzylparaben in aqueous media. *International Journal of Molecular Sciences*, **16**, 3656–3676.

- Aydin, H. and Baysal, G. (2006) Adsorption of acid dyes in aqueous solutions by shells of bittim (*Pistacia khinjuk* Stocks). *Desalination*, **196**, 248–259.
- Babbar, N. and Oberoi, H.S. (2014) Influence of different solvents in extraction of phenolic compounds from vegetable residues and their evaluation as natural sources of antioxidants. **51**, 2568–2575.
- Bagheri, H. and Piri-Moghadam, H. (2012) Sol-gel-based molecularly imprinted xerogel for capillary microextraction. *Analytical and Bioanalytical Chemistry*, **404**, 1597–1602.
- Bapat, G., Labade, C., Chaudhari, A., and Zinjarde, S. (2016) Silica nanoparticle based techniques for extraction, detection, and degradation of pesticides. *Advances in Colloid and Interface Science*, **237**, 1–14.
- Bieniek, G. (1996) Simultaneous determination of phenol, cresol, xylenol isomers and naphthols in urine by capillary gas chromatography. *Journal of Chromatography B: Biomedical Applications*, **682**, 167–172.
- Brinker, C.J. and Scherer, G.W. (2013) *Sol-gel science: the physics and chemistry of sol-gel processing*. P. in.: Academic press.
- Buchet, J.P. and Lauwerys, R.R. (1973) Measurement of urinary hippuric and m-methylhippuric acids by gas chromatography. *British journal of industrial medicine*, **30**, 125–128.
- Caldwell, D.J., Armstrong, T.W., Barone, N.J., Suder, J.A., and Evans, M.J. (2000) Hydrocarbon Solvent Exposure Data: Compilation and Analysis of the Literature. *AIHAJ - American*

*Industrial Hygiene Association*, **61**, 881–894.

Campbell, L., Wilson, H.K., Samuel, A.M., and Gompertz, D. (1988) Interactions of m-xylene and aspirin metabolism in man. *British journal of industrial medicine*, **45**, 127–132.

Castleman, B.I. and Ziem, G.E. (1994) American conference of governmental industrial hygienists: low threshold of credibility. *American journal of industrial medicine*, **26**, 133–143.

Chang, Y.-S., Ko, T.-H., Hsu, T.-J., and Syu, M.-J. (2009) Synthesis of an imprinted hybrid organic– inorganic polymeric sol– gel matrix toward the specific binding and isotherm kinetics investigation of creatinine. *Analytical chemistry*, **81**, 2098–2105.

Chen, F.-F., Xie, X.-Y., and Shi, Y.-P. (2013) Preparation of magnetic molecularly imprinted polymer for selective recognition of resveratrol in wine. *Journal of Chromatography A*, **1300**, 112–118.

Chen, F.F., Wang, G.Y., and Shi, Y.P. (2011) Molecularly imprinted polymer microspheres for solid-phase extraction of protocatechuic acid in *Rhizoma homalomenae*. *Journal of Separation Science*, **34**, 2602–2610.

Chen, Z., Liu, S.-J., Cai, S.-X., Yao, Y.-M., Yin, H., Ukai, H., Uchida, Y., Nakatsuka, H., Watanabe, T., and Ikeda, M. (1994) Exposure of workers to a mixture of toluene and xylenes. II. Effects. *Occupational and environmental medicine*, **51**, 47–49.

Cheng, W., Liu, Z., and Wang, Y. (2013) Preparation and application of surface molecularly imprinted silica gel for selective extraction of melamine from milk samples. *Talanta*, **116**, 396–

402.

- Choi, Y.B., Kim, N.H., Kim, S.H., Tae, G.S., and Kim, H.H. (2014) Heterogeneous electrochemical immunoassay of hippuric acid on the electrodeposited organic films. *Sensors (Basel, Switzerland)*, **14**, 18886–18897.
- Chong, S.L., Wang, D., Hayes, J.D., Wilhite, B.W., and Malik, a. (1997) Sol-gel coating technology for the preparation of solid-phase microextraction fibers of enhanced thermal stability. *Analytical chemistry*, **69**, 3889–98.
- Cok, I., Dagdelen, A., and Gökçe, E. (2003) Determination of urinary hippuric acid and o-cresol levels as biological indicators of toluene exposure in shoe-workers and glue sniffers. *Biomarkers : biochemical indicators of exposure, response, and susceptibility to chemicals*, **8**, 119–127.
- Condon, J.B. (2006) *Surface area and porosity determinations by physisorption: measurements and theory*. P. in.: Elsevier.
- De Carvalho, D., Lanchote, V.L., Bonato, P.S., Queiroz, R.H.C., Santos, A.C., and Dreossi, S.A.C. (1991) A new derivatization procedure for the analysis of hippuric acid and m-methyl-hippuric acid by gas chromatography. *International Archives of Occupational and Environmental Health*, **63**, 33–37.
- De Paiva, M.J.N., Martins, I., and De Siqueira, M.E.P.B. (2007) Analysis of ortho-cresol in urine by solid phase microextraction-capillary gas chromatography. *Journal of the Brazilian Chemical Society*, **18**, 1034–1039.
- Department of Occupational and Health, M. (2009) GUIDELINES

ON MEDICAL SURVEILLANCE. *European Urology*, 179–201.

- Din, A.T.M., Hameed, B.H., and Ahmad, A.L. (2009) Batch adsorption of phenol onto physiochemical-activated coconut shell. *Journal of Hazardous Materials*, **161**, 1522–1529.
- Du, W., Lei, C., Zhang, S., Bai, G., Zhou, H., Sun, M., Fu, Q., and Chang, C. (2014) Determination of clenbuterol from pork samples using surface molecularly imprinted polymers as the selective sorbents for microextraction in packed syringe. *Journal of Pharmaceutical and Biomedical Analysis*, **91**, 160–168.
- El-Beqqali, A. and Abdel-Rehim, M. (2016) Molecularly imprinted polymer-sol-gel tablet toward micro-solid phase extraction: I. Determination of methadone in human plasma utilizing liquid chromatography tandem mass spectrometry. *Analytica Chimica Acta*, **936**, 116–122.
- Farrington, K. and Regan, F. (2009) Molecularly imprinted sol gel for ibuprofen: An analytical study of the factors influencing selectivity. *Talanta*, **78**, 653–659.
- Fujii, T., Mabuchi, T., Kitamura, H., Kawauchi, O., Negishi, N., and Anpo, M. (1992) Fluorescence Spectra of 1-Naphthol During the Sol-Gel Process of a Mixed Aluminum-Silicon Alkoxide (Si : Al = 94 : 6). *Bulletin of the Chemical Society of Japan*, **65**, 720–727.
- Fustinoni, S., Mercadante, R., Campo, L., Scibetta, L., Valla, C., and Foà, V. (2005) Determination of urinary ortho- and meta-cresol in humans by headspace SPME gas chromatography/mass

- spectrometry. *Journal of Chromatography B: Analytical Technologies in the Biomedical and Life Sciences*, **817**, 309–317.
- Gama, M.R. and Bottoli, C.B.G. (2017) Molecularly imprinted polymers for bioanalytical sample preparation. *Journal of Chromatography B*, **1043**, 107–121.
- Gao, B., Wang, X., and Zhang, Y. (2013) Preparation of chromate anion surface-imprinted material IIP-PVI/SiO<sub>2</sub> based on polyvinylimidazole-grafted particles PVI/SiO<sub>2</sub> and its ionic recognition characteristic. *Materials Chemistry and Physics*, **140**, 478–486.
- García-Calzón, J.A. and Díaz-García, M.E. (2007) Characterization of binding sites in molecularly imprinted polymers. *Sensors and Actuators B: Chemical*, **123**, 1180–1194.
- Ghittori, S., Maestri, L., Fiorentino, M.L., and Imbriani, M. (1995) Evaluation of occupational exposure to benzene by urinalysis. *International archives of occupational and environmental health*, **67**, 195–200.
- Giraldo, L.F., López, B.L., Pérez, L., Urrego, S., Sierra, L., and Mesa, M. (2007) Mesoporous silica applications. Pp. 129–141 in: *Macromolecular Symposia*. Wiley Online Library.
- Granado, V.L. V, Guti rrez-Capit n, M., Fern ndez-S nchez, C., Gomes, M.T.S.R., Rudnitskaya, A., and Jimenez-Jorquera, C. (2014) Thin-film electrochemical sensor for diphenylamine detection using molecularly imprinted polymers. *Analytica Chimica Acta*, **809**, 141–147.

- Han, D.-M., Fang, G.-Z., and Yan, X.-P. (2005) Preparation and evaluation of a molecularly imprinted sol-gel material for on-line solid-phase extraction coupled with high performance liquid chromatography for the determination of trace pentachlorophenol in water samples. *Journal of Chromatography A*, **1100**, 131–136.
- Harwood, L.M., Moody, C.J., and Percy, J.M. (1999) *Experimental organic chemistry: standard and microscale*. P. in.: Blackwell science Malden, MA.
- Hasegawa, K., Shiojima, S., Koizumi, A., and Ikeda, M. (1983) Hippuric acid and o-cresol in the urine of workers exposed to toluene. *International archives of occupational and environmental health*, **52**, 197–208.
- Hench, L.L. and West, J.K. (1990) The sol-gel process. *Chemical Reviews*, **90**, 33–72.
- Ho, Y.-S. and McKay, G. (1999a) Pseudo-second order model for sorption processes. *Process biochemistry*, **34**, 451–465.
- Ho, Y.-S. and McKay, G. (2003) Sorption of dyes and copper ions onto biosorbents. *Process Biochemistry*, **38**, 1047–1061.
- Ho, Y.S. and McKay, G. (1999b) The sorption of lead (II) ions on peat. *Water research*, **33**, 578–584.
- Hoda, N., Bayram, E., and Ayranci, E. (2006) Kinetic and equilibrium studies on the removal of acid dyes from aqueous solutions by adsorption onto activated carbon cloth. *Journal of hazardous materials*, **137**, 344–351.



- Hu, C., Deng, J., Zhao, Y., Xia, L., Huang, K., Ju, S., and Xiao, N. (2014a) A novel core-shell magnetic nano-sorbent with surface molecularly imprinted polymer coating for the selective solid phase extraction of dimetridazole. *Food Chemistry*, **158**, 366–373.
- Hu, Y., Feng, T., and Li, G. (2014b) A novel solid fluorescence method for the fast determination of quercetin in biological samples based on the quercetin–Al (III) complex imprinted polymer. *Spectrochimica Acta Part A: Molecular and Biomolecular Spectroscopy*, **118**, 921–928.
- Iftikhar, A.R., Bhatti, H.N., Hanif, M.A., and Nadeem, R. (2009) Kinetic and thermodynamic aspects of Cu (II) and Cr (III) removal from aqueous solutions using rose waste biomass. *Journal of Hazardous Materials*, **161**, 941–947.
- Ikeda, M. and Ohtsuji, H. (1969) Significance of urinary hippuric acid determination as an index of toluene exposure. *British journal of industrial medicine*, **26**, 244–246.
- Inoue, O., Seiji, K., Suzuki, T., Watanabe, T., Nakatsuka, H., Satoh, H., and Ikeda, M. (1991) Simultaneous determination of hippuric acid, o-, m-, and p-methylhippuric acid, phenylglyoxylic acid, and mandelic acid by HPLC. *Bulletin of Environmental Contamination and Toxicology*, **47**, 204–210.
- Inoue, O., Seiji, K., Watanabe, T., Nakatsuka, H., Jin, C., Liu, S.-J., and Ikeda, M. (1993) Effects of smoking and drinking on excretion of hippuric acid among toluene-exposed workers. *International archives of occupational and environmental*

*health*, **64**, 425–430.

Inoue, O., Kanno, E., Kakizaki, M., WATANABE, T., HIGASHIKAWA, K., and IKEDA, M. (2000) Urinary phenylmercapturic acid as a marker of occupational exposure to benzene. *Industrial Health*, **38**, 195–204.

Inoue, O., Kanno, E., Yusa, T., Kakizaki, M., Ukai, H., Okamoto, S., Higashikawa, K., and Ikeda, M. (2002) Urinary benzylmercapturic acid as a marker of occupational exposure to toluene. *International archives of occupational and environmental health*, **75**, 341–347.

Inoue, O., Kanno, E., Kasai, K., Ukai, H., Okamoto, S., and Ikeda, M. (2004) Benzylmercapturic acid is superior to hippuric acid and o-cresol as a urinary marker of occupational exposure to toluene. *Toxicology letters*, **147**, 177–186.

Jin, Y.-F., Zhang, Y.-P., Huang, M.-X., Bai, L.-Y., and Lee, M.L. (2013) A novel method to prepare monolithic molecular imprinted polymer fiber for solid-phase microextraction by microwave irradiation. *Journal of separation science*, **36**, 1429–36.

Kabir, A., Furton, K.G., and Malik, A. (2013) Innovations in sol-gel microextraction phases for solvent-free sample preparation in analytical chemistry. *TrAC - Trends in Analytical Chemistry*, **45**, 197–218.

Kamaruzaman, S., Hauser, P.C., Sanagi, M.M., Ibrahim, W.A.W., Endud, S., and See, H.H. (2013) A simple microextraction and preconcentration approach based on a mixed matrix membrane.

*Analytica Chimica Acta*, **783**, 24–30.

Kamra, T., Chaudhary, S., Xu, C., Johansson, N., Montelius, L., Schnadt, J., and Ye, L. (2015) Covalent immobilization of molecularly imprinted polymer nanoparticles using an epoxy silane. *Journal of colloid and interface science*, **445**, 277–284.

Kawai, T., Mizunuma, K., Okada, Y., Horiguchi, S., and Ikeda, M. (1996) Toluene itself as the best urinary marker of toluene exposure. *International Archives of Occupational and Environmental Health*, **68**, 289–297.

Kia, S., Fazilati, M., Salavati, H., and Bohlooli, S. (2016) Preparation of a novel molecularly imprinted polymer by the sol–gel process for solid phase extraction of vitamin D3. *RSC Adv.*, **6**, 31906–31914.

King, M.D., Day, R.E., Oliver, J.S., Lush, M., and Watson, J.M. (1981) Solvent encephalopathy. *Br Med J (Clin Res Ed)*, **283**, 663–665.

Kira, S. (1977) Measurement by gas chromatography of urinary hippuric acid and methylhippuric acid as indices of toluene and xylene exposure. *British journal of industrial medicine*, **34**, 305–309.

Kongtip, P., Vararussami, J., and Pruktharathikul, V. (2001) Modified method for determination of hippuric acid and methylhippuric acid in urine by gas chromatography. *Journal of Chromatography B: Biomedical Sciences and Applications*, **751**, 199–203.

Krämer, A., Linnert Jr, M., Wrbitzky, R., and Angerer, J. (1999)

- Occupational chronic exposure to organic solvents XVII. Ambient and biological monitoring of workers exposed to xylenes. *International archives of occupational and environmental health*, **72**, 52–55.
- Kubota, K., Horai, Y., Kushida, K., and Ishizaki, T. (1988) Determination of benzoic acid and hippuric acid in human plasma and urine by high-performance liquid chromatography. *Journal of chromatography*, **425**, 67–75.
- Kwon, B., Kim, S.S.S., Kim, S.S.S., Lee, D.K., Park, Y.J., Kim, M.D., Lee, J.S., and Kim, S.S.S. (2011) <sup>1</sup>H NMR spectroscopic identification of a glue sniffing biomarker. *Forensic Science International*, **209**, 120–125.
- Kyzas, G.Z., Lazaridis, N.K., and Bikiaris, D.N. (2013) Optimization of chitosan and  $\beta$ -cyclodextrin molecularly imprinted polymer synthesis for dye adsorption. *Carbohydrate polymers*, **91**, 198–208.
- Lee, C., Lee, J., Lee, J., Eom, H.Y., Kim, M.K., Suh, J.H., and Yeom, H. (2009) Rapid HPLC Method for the Simultaneous Determination of Eight Urinary Metabolites of Toluene, Xylene and Styrene. *Bulletin of the Korean Chemical Society*, **30**, 2021–2026.
- Lemen, R.A., Mazzuckelli, L.F., Niemeier, R.W., and Ahlers, H.W. (1989) Occupational safety and health standards. *Annals of the New York Academy of Sciences*, **572**, 100–106.
- Li, M., Wu, Z., and Tan, J. (2012) Properties of form-stable paraffin/silicon dioxide/expanded graphite phase change

composites prepared by sol–gel method. *Applied energy*, **92**, 456–461.

- Liao, S., Zhang, W., Long, W., Hou, D., Yang, X., and Tan, N. (2016) Adsorption characteristics, recognition properties, and preliminary application of nordihydroguaiaretic acid molecularly imprinted polymers prepared by sol–gel surface imprinting technology. *Applied Surface Science*, **364**, 579–588.
- Liu, Y., Wang, F., Tan, T., and Lei, M. (2007) Study of the properties of molecularly imprinted polymers by computational and conformational analysis. *Analytica chimica acta*, **581**, 137–146.
- Lovreglio, P., Barbieri, A., Carrieri, M., Sabatini, L., Fracasso, M.E., Doria, D., Drago, I., Basso, A., D’Errico, M.N., Bartolucci, G.B., Violante, F.S., and Soleo, L. (2010) Validity of new biomarkers of internal dose for use in the biological monitoring of occupational and environmental exposure to low concentrations of benzene and toluene. *International Archives of Occupational and Environmental Health*, **83**, 341–356.
- Lowry, L.K. (1986) Biological exposure index as a complement to the TLV. *Journal of occupational medicine.: official publication of the Industrial Medical Association*, **28**, 578–582.
- Maestri, L., Ghittori, S., and Imbriani, M. (1997) Determination of specific mercapturic acids as an index of exposure to environmental benzene, toluene, and styrene. *Industrial health*, **35**, 489–501.
- Mall, I.D., Srivastava, V.C., and Agarwal, N.K. (2006) Removal of Orange-G and Methyl Violet dyes by adsorption onto bagasse

fly ash—kinetic study and equilibrium isotherm analyses. *Dyes and pigments*, **69**, 210–223.

Manzoor, S., Buffon, R., and Rossi, A.V. (2015) Molecularly imprinted solid phase extraction of fluconazole from pharmaceutical formulations. *Talanta*, **134**, 1–7.

Matsui, H., Kasao, M., and Imamura, S. (1978) High-performance liquid chromatographic determination of hippuric acid in human urine. *Journal of chromatography*, **145**, 231–236.

Mehanna, a S. and Dowling, M. (1999) Liquid chromatographic determination of hippuric acid for the evaluation of ethacrynic acid as angiotensin converting enzyme inhibitor. *Journal of pharmaceutical and biomedical analysis*, **19**, 967–973.

Michitsuji, H., Ohara, A., Yamaguchi, K., and Fujiki, Y. (1987) Effect of intake of refreshments in excretion of hippuric acid in urine. *Matsushita Med J*, **26**, 105–116.

Moein, M.M., El-Beqqali, A., Javanbakht, M., Karimi, M., Akbari-Adergani, B., and Abdel-Rehim, M. (2014a) On-line detection of hippuric acid by microextraction with a molecularly-imprinted polysulfone membrane sorbent and liquid chromatography-tandem mass spectrometry. *Journal of chromatography. A*, **1372C**, 55–62.

Moein, M.M., Said, R., Bassyouni, F., and Abdel-Rehim, M. (2014b) Solid phase microextraction and related techniques for drugs in biological samples. *Journal of analytical methods in chemistry*, **2014**.

Moein, M.M., Javanbakht, M., Karimi, M., Akbari-adergani, B., and

- Abdel-Rehim, M. (2015a) A new strategy for surface modification of polysulfone membrane by in situ imprinted sol-gel method for the selective separation and screening of *l*-Tyrosine as a lung cancer biomarker. *The Analyst*, **140**, 1939–1946.
- Moein, M.M., Javanbakht, M., Karimi, M., Akbari-adergani, B., and Abdel-Rehim, M. (2015b) Three-phase molecularly imprinted sol-gel based hollow fiber liquid-phase microextraction combined with liquid chromatography–tandem mass spectrometry for enrichment and selective determination of a tentative lung cancer biomarker. *Journal of Chromatography B*, **995–996**.
- Moein, M.M., El Beqqali, A., and Abdel-Rehim, M. (2017) Bioanalytical method development and validation: critical concepts and strategies. *Journal of Chromatography B*, **1043**, 3–11.
- Monteiro, C., Franco, J.M., Proença, P., Castañera, A., Claro, A., Vieira, D.N., and Corte-Real, F. (2014a) Qualitative and quantitative analysis of a group of volatile organic compounds in biological samples by HS-GC/FID: application in practical cases. *Forensic science international*, **243**, 137–143.
- Monteiro, C., Franco, J.M., Proença, P., Castañera, A., Claro, A., Vieira, D.N., and Corte-Real, F. (2014b) Qualitative and quantitative analysis of a group of volatile organic compounds in biological samples by HS-GC/FID: Application in practical cases. *Forensic Science International*, **243**, 137–143.

- Moon, C.-S., Lee, J.-T., Chun, J.-H., and Ikeda, M. (2001) Use of solvents in industries in Korea: experience in Sinpyeong-Jangrim industrial complex. *International archives of occupational and environmental health*, **74**, 148–152.
- Morata, T.C. (2003) Chemical exposure as a risk factor for hearing loss. *Journal of Occupational and Environmental Medicine*, **45**, 676–682.
- Morgan, M.S. (1997) The biological exposure indices: A key component in protecting workers from toxic chemicals. *Environmental Health Perspectives*, **105**, 105–115.
- National Institute Occupational, Safety and Health, A. (1977) A Guide to Their Recognition.
- Nau, C.A. (1959) Handbook of Organic Industrial Solvents. *Journal of Occupational and Environmental Medicine*, **1**, 407.
- Nayak, P.S. and Singh, B.K. (2007) Removal of phenol from aqueous solutions by sorption on low cost clay. *Desalination*, **207**, 71–79. Elsevier.
- Ncanana, Z.S. and Pullabhotla, V.S.R.R. (2018) Ozone Initiated Oxidation of Cresol Isomers Using  $\gamma$ -Al<sub>2</sub>O<sub>3</sub> and SiO<sub>2</sub> as Adsorbents. *Catalysis Letters*, 1–12.
- Ng, C., Losso, J.N., Marshall, W.E., and Rao, R.M. (2002) Freundlich adsorption isotherms of agricultural by-product-based powdered activated carbons in a geosmin–water system. *Bioresource technology*, **85**, 131–135.
- NIOSH Manual of Analytical Method 5th Edition, N. (2017) NIOSH



Manual of Analytical Methods 5th Edition--new resources and direction. *Journal of occupational and environmental hygiene*, **8**, D59.

Nise, G. (1992) Urinary excretion of o-cresol and hippuric acid after toluene exposure in rotogravure printing. *International archives of occupational and environmental health*, **63**, 377–381.

Ogata, M. and Taguchi, T. (1986) Quantitative analysis of urinary glycine conjugates by high performance liquid chromatography: excretion of hippuric acid and methylhippuric acids in the urine of subjects exposed to vapours of toluene and xylenes. *International archives of occupational and environmental health*, **58**, 121–129.

Ogata, M. and Taguchi, T. (1987) Quantitation of urinary metabolites of toluene, xylene, styrene, ethylbenzene, benzene and phenol by automated high performance liquid chromatography. *International archives of occupational and environmental health*, **59**, 263–272.

Ogata, M., Tomokuni, K., and Takatsuka, Y. (1969) Quantitative determination in urine of hippuric acid and m-or p-methylhippuric acid, metabolites of toluene and m-or p-xylene. *British journal of industrial medicine*, **26**, 330–334.

Ohashi, Y., Mamiya, T., Mitani, K., Wang, B., Takigawa, T., Kira, S., and Kataoka, H. (2006) Simultaneous determination of urinary hippuric acid, o-, m- and p-methylhippuric acids, mandelic acid and phenylglyoxylic acid for biomonitoring of volatile organic compounds by gas chromatography-mass spectrometry.

*Analytica Chimica Acta*, **566**, 167–171.

- Olsson, U., Bemg, A., Kristensson, J., Palmberg, J., and Sollenberg, J. (1990) High-performance liquid chromatographic analysis of hippuric acid in human blood plasma. *Journal of Chromatography B: Biomedical Sciences and Applications*, **532**, 45–53.
- Organization, W.H. (1996) Biological monitoring of chemical exposure in the workplace: guidelines. Geneva: World Health Organization.
- OSHA Act. (1994) Occupational Safety and Health Act 514 1994.
- Özacar, M., Şengil, İ.A., and Türkmenler, H. (2008) Equilibrium and kinetic data, and adsorption mechanism for adsorption of lead onto valonia tannin resin. *Chemical Engineering Journal*, **143**, 32–42.
- Pacenti, M., Dugheri, S., Villanelli, F., Bartolucci, G., Calamai, L., Boccalon, P., Arcangeli, G., Vecchione, F., Alessi, P., Kikic, I., and Cupelli, V. (2008) Determination of organic acids in urine by solid-phase microextraction and gas chromatography-ion trap tandem mass spectrometry previous “in sample” derivatization with trimethyloxonium tetrafluoroborate. *Biomedical chromatography : BMC*, **22**, 1155–63.
- Pan, J., Yao, H., Guan, W., Ou, H., Huo, P., Wang, X., Zou, X., and Li, C. (2011) Selective adsorption of 2,6-dichlorophenol by surface imprinted polymers using polyaniline/silica gel composites as functional support: Equilibrium, kinetics, thermodynamics modeling. *Chemical Engineering Journal*, **172**,

847–855.

- Paxman, D.G. and Robinson, J.C. (1990) Regulation of occupational carcinogens under OSHA's air contaminants standard. *Regulatory toxicology and pharmacology*, **12**, 296–308.
- Pierce, C.H., Dills, R.L., Morgan, M.S., Vicini, P., and Kaiman, D.A. (1998) Biological monitoring of controlled toluene exposure. *International Archives of Occupational and Environmental Health*, **71**, 433–444.
- Pierce, C.H., Chen, Y., Dills, R.L., Kalman, D.A., and Morgan, M.S. (2002) Toluene metabolites as biological indicators of exposure. *Toxicology Letters*, **129**, 65–76.
- Poggi, G., Giusiani, M., Palagi, U., Paggiaro, P.L., Loi, A.M., Dazzi, F., Siclari, C., and Baschieri, L. (1982) High-performance liquid chromatography for the quantitative determination of the urinary metabolites of toluene, xylene, and styrene. *International Archives of Occupational and Environmental Health*, **50**, 25–31.
- Rampey, A.M., Umpleby, R.J., Rushton, G.T., Iseman, J.C., Shah, R.N., and Shimizu, K.D. (2004) Characterization of the imprint effect and the influence of imprinting conditions on affinity, capacity, and heterogeneity in molecularly imprinted polymers using the Freundlich isotherm-affinity distribution analysis. *Analytical chemistry*, **76**, 1123–1133.
- Ramström, O., Ye, L., and Gustavsson, P.-E. (1998) Chiral recognition by molecularly imprinted polymers in aqueous media. *Chromatographia*, **48**, 197–202.
- Rauscher, D., Lehnert, G., and Angerer, J. (1994) Biomonitoring of

- occupational and environmental exposures to benzene by measuring trans, trans-muconic acid in urine. *Clinical Chemistry*, **40**, 1468–1470.
- Rouquerol, F., Rouquerol, J., Sing, K.S.W., Llewellyn, P., and Maurin, G. (2014) *Adsorption by powders and porous solids*. P. in: *Climate Change 2013 - The Physical Science Basis*. 631 pp.
- Sadeghi, S. and Jahani, M. (2014) Solid-Phase Extraction of Florfenicol from Meat Samples by a Newly Synthesized Surface Molecularly Imprinted Sol–Gel Polymer. *Food Analytical Methods*, **7**, 2084–2094.
- Saito, T. and Takeichi, S. (2002) Simultaneous detection of hippuric acid and methylhippuric acid in urine by empore??? disk and gas chromatography-mass spectrometry. *Journal of Pharmaceutical and Biomedical Analysis*, **30**, 365–370.
- Sanagi, M.M., Salleh, S., Ibrahim, W.A.W., Naim, A.A., Hermawan, D., Miskam, M., Hussain, I., and Aboul-Enein, H.Y. (2013) Molecularly imprinted polymer solid-phase extraction for the analysis of organophosphorus pesticides in fruit samples. *Journal of Food Composition and Analysis*, **32**, 155–161.
- Schenk, L., Hansson, S.O., Rudén, C., and Gilek, M. (2008) Occupational exposure limits: A comparative study. *Regulatory toxicology and pharmacology*, **50**, 261–270.
- Schettgen, T., Alt, A., Dewes, P., and Kraus, T. (2015) Simple and sensitive GC/MS-method for the quantification of urinary phenol, o- and m-cresol and ethylphenols as biomarkers of exposure to industrial solvents. *Journal of Chromatography B*:

*Analytical Technologies in the Biomedical and Life Sciences*, **995–996**, 93–100.

- Sellergren, B. (2000) *Molecularly imprinted polymers: man-made mimics of antibodies and their application in analytical chemistry*. P. in.: Elsevier.
- Sellergren, B. and Shea, K.J. (1993) Influence of polymer morphology on the ability of imprinted network polymers to resolve enantiomers. *Journal of Chromatography A*, **635**, 31–49.
- Sugita, M., Aikawa, H., Suzuki, K., Yamasaki, T., Minowa, H., Etoh, R., and Kasuga, H. (1988) Urinary hippuric acid excretion in everyday life. *The Tokai journal of experimental and clinical medicine*, **13**, 185–190.
- Surikumar, H., Mohamad, S., and Sari, N.M. (2014) Molecular imprinted polymer of methacrylic acid functionalised ??-Cyclodextrin for selective removal of 2,4-dichlorophenol. *International Journal of Molecular Sciences*, **15**, 6111–6136.
- Taira, S., Kaneko, D., Onuma, K., Miyazato, A., Hiroki, T., Kawamura-Konishi, Y., and Ichiyanagi, Y. (2012) Synthesis and characterization of functionalized magnetic nanoparticles for the detection of pesticide. *International Journal of Inorganic Chemistry*, **2012**.
- Topping, M. (2001) Occupational exposure limits for chemicals. *Occup Environ Med*, **58**, 138–144.
- Toulabi, P., Daneshfar, A., and Sahrai, R. (2010) Determination of hippuric acid in biological fluids using single drop liquid–liquid–liquid microextraction. *Analytical Methods*, **2**, 564–569.

- Truchon, G., Tardif, R., and Brodeur, J. (1996) Gas chromatographic determination of urinary o-cresol for the monitoring of toluene exposure. *Journal of analytical toxicology*, **20**, 309–312.
- Tsai, H.-A. and Syu, M.-J. (2011) Preparation of imprinted poly (tetraethoxysilanol) sol–gel for the specific uptake of creatinine. *Chemical engineering journal*, **168**, 1369–1376. Elsevier.
- Turner, N.W., Piletska, E. V, Karim, K., Whitcombe, M., Malecha, M., Magan, N., Baggiani, C., and Piletsky, S.A. (2004) Effect of the solvent on recognition properties of molecularly imprinted polymer specific for ochratoxin A. *Biosensors and Bioelectronics*, **20**, 1060–1067.
- Ukai, H., Kawai, T., Inoue, O., Maejima, Y., Fukui, Y., Ohashi, F., Okamoto, S., Takada, S., Sakurai, H., and Ikeda, M. (2007) Comparative evaluation of biomarkers of occupational exposure to toluene. *International archives of occupational and environmental health*, **81**, 81–93.
- Umberger, C.J. and Fiorese, F.F. (1963) Colorimetric method for hippuric acid. *Clinical chemistry*, **9**, 91–96.
- Van Roosmalen, P.B. and Drummond, I. (1978) Simultaneous determination by gas chromatography of the major metabolites in urine of toluene, xylenes and styrene. *British journal of industrial medicine*, **35**, 56–60.
- Veningerová, M., Prachar, V., Uhnák, J., Lukáčsová, M., and Trnovec, T. (1994) Determination of chlorinated phenols and cresols in human urine using solid-phase extraction and gas chromatography. *Journal of Chromatography B: Biomedical*

*Sciences and Applications*, **657**, 103–110.

- Veulemans, H., Van Vlem, E., Janssens, H., and Masschelein, R. (1979) Exposure to toluene and urinary hippuric acid excretion in a group of heliorotagravure printing workers. *International archives of occupational and environmental health*, **44**, 99–107.
- VILLANUEVA, M.B.G., JONAI, H., KANNO, S., and TAKEUCHI, Y. (1994) Dietary sources and background levels of hippuric acid in urine: comparison of Philippine and Japanese levels. *Industrial health*, **32**, 239–246.
- Vitali, L., Gonçalves, S., Rodrigues, V., Fávere, V.T., and Mücke, G.A. (2016) Development of a fast method for simultaneous determination of hippuric acid, mandelic acid, and creatinine in urine by capillary zone electrophoresis using polymer multilayer-coated capillary. *Analytical and Bioanalytical Chemistry*. *Analytical and Bioanalytical Chemistry*.
- Wu, N., Luo, Z., Ge, Y., Guo, P., Du, K., Tang, W., Du, W., Zeng, A., Chang, C., and Fu, Q. (2016a) A novel surface molecularly imprinted polymer as the solid-phase extraction adsorbent for the selective determination of ampicillin sodium in milk and blood samples. *Journal of Pharmaceutical Analysis*, **6**, 157–164.
- Wu, N., Luo, Z., Ge, Y., Guo, P., Du, K., Tang, W., Du, W., Zeng, A., Chang, C., and Fu, Q. (2016b) A novel surface molecularly imprinted polymer as the solid-phase extraction adsorbent for the selective determination of ampicillin sodium in milk and blood samples. *Journal of Pharmaceutical Analysis*, **6**, 157–164.
- Xie, C., Liu, B., Wang, Z., Gao, D., Guan, G., and Zhang, Z. (2008)

- Molecular imprinting at walls of silica nanotubes for TNT recognition. *Analytical chemistry*, **80**, 437–443.
- Yacob, A.R. and Zinalibdin, M.R. (2010) High Performance Liquid Chromatography Determination of Urinary Hippuric Acid and Benzoic Acid as Indices for Glue Sniffer Urine. *World Academy of Science, Engineering and Technology*, **38**, 973–978.
- Yamazaki, K., Tanaka, E., and Misawa, S. (1992) Urinary ortho-cresol concentrations as an indicator of toluene inhalation in glue-sniffers. *Journal of the Forensic Science Society*, **32**, 215–223.
- Yan, M. (2004) *Molecularly imprinted materials: science and technology*. P. in.: CRC press.
- Yang, L., Wang, G.-C., Liu, Y.-J., An, J.-J., and Wang, M. (2013) Development of a stable biosensor based on a SiO<sub>2</sub> nanosheet–Nafion–modified glassy carbon electrode for sensitive detection of pesticides. *Analytical and bioanalytical chemistry*, **405**, 2545–2552.
- Yoshida, M., Akane, A., Mitani, T., and Watabiki, T. (2005) Simple colorimetric semiquantitation method of hippuric acid in urine for demonstration of toluene abuse. *Legal Medicine*, **7**, 198–200.
- Yue-dong, Y. (1998) Simultaneous determination of creatine, uric acid, creatinine and hippuric acid in urine by high performance liquid chromatography. *Biomedical Chromatography*, **12**, 47–49.
- Yuh-Shan, H. (2004) Citation review of Lagergren kinetic rate equation on adsorption reactions. *Scientometrics*, **59**, 171–177.



- Zeng, H., Wang, Y., Liu, X., Kong, J., and Nie, C. (2012) Preparation of molecular imprinted polymers using bi-functional monomer and bi-crosslinker for solid-phase extraction of rutin. *Talanta*, **93**, 172–181.
- Zhao, F., Wang, Z., Wang, H., and Ding, M. (2011) Determination of hippuric acid in human urine by ion chromatography with conductivity detection. *Journal of Chromatography B: Analytical Technologies in the Biomedical and Life Sciences*, **879**, 296–298.
- Zhao, J. (2015) Simultaneous determination of plasma creatinine, uric acid, kynurenine and tryptophan by high-performance liquid chromatography: Method validation and in application to the assessment of renal function. *Biomedical Chromatography*, **29**, 410–415.
- Zhu, G., Wang, X., Gao, X., and Fan, J. (2015) Preparation of surface molecularly imprinted polymer and selective extraction of 1-methoxyethyl-3-methylimidazolium bis [(trifluoromethyl) sulfonyl] imide. *Monatshefte für Chemie-Chemical Monthly*, **146**, 431–440.
- Zuppi, C., Rossetti, D.V., Vitali, A., Vincenzoni, F., Giardina, B., Castagnola, M., and Messana, I. (2003) Determination of urinary hippuric acid by micellar electrokinetic capillary chromatography. *Journal of Chromatography B*, **793**, 223–228.