Determination of acrylamide in brewed coffee and coffee powder using polymeric ionic liquid-based sorbent coatings in solid-phase microextraction coupled to gas chromatography-mass spectrometry

Determination of Acrylamide in Brewed Coffee and Coffee Powder using Polymeric Ionic Liquid-based Sorbent Coatings in Solid-Phase Microextraction Coupled to Gas Chromatography-Mass Spectrometry

Cecilia Cagliero,[b] Tien D. Ho,[a] Cheng Zhang,[a] Carlo Bicchi,[b] and Jared L. Anderson[a,*]

[a]Department of Chemistry, Iowa State University, Ames, IA 50011 USA

[b]Dipartimento di Scienza e Tecnologia del Farmaco, Università degli Studi di Torino, I-10125 Torino, Italy

Abstract

This study describes a simple and rapid sampling method employing a polymeric ionic liquid (PIL) sorbent coating in direct immersion solid-phase microextraction (SPME) for the trace-level analysis of acrylamide in brewed coffee and coffee powder. The crosslinked PIL sorbent coating demonstrated superior sensitivity in the extraction of acrylamide compared to all commercially available SPME coatings. A spin coating method was developed to evenly distribute the PIL coating on the SPME support and reproducibly produce fibers with a large film thickness. Ninhydrin was employed as a quenching reagent during extraction to inhibit the production of interfering acrylamide. The PIL fiber produced a limit of quantitation for acrylamide of 10 μg L^{-1} and achieved comparable results to the ISO method in the analysis of six coffee powder samples.

Keywords

Acrylamide; Coffee; Polymeric ionic liquids; Solid-phase microextraction; Gas Chromatography-Mass Spectrometry

---

*Corresponding author
Jared L. Anderson
Department of Chemistry, 1605 Gilman Hall
Iowa State University, Ames, IA 50010
andersoj@iastate.edu
515-294-8356

1. Introduction

Acrylamide is an unsaturated amide formed when carbohydrate-rich foods are subjected to high temperatures during cooking or thermal processing [1]. Initial studies on the mechanistic pathway of acrylamide formation in food proposed the Maillard reaction (non-enzymatic browning reaction) as the major chemical route [2, 3]. Acrylamide has previously been observed in several food matrices such as potato crisps, French fries, crispy bread, breakfast cereals, pastries, and coffee. The concentration levels of acrylamide are high in coffee compared to other food products, although lower levels can be expected due to dilution within coffee beverages [4]. The toxicological properties of acrylamide have been well-studied and include neurotoxicity, genotoxicity, carcinogenicity, and reproductive toxicity [5, 6]. For this reason, it is possible that maximum limits of acrylamide in food could be proposed in the future thereby requiring new analytical methods for its sensitive analysis.

Analytical methods employing high performance liquid chromatography (HPLC) coupled to mass spectrometry (MS) or tandem MS (MS/MS) for the analysis of acrylamide in food have been reported [7, 8]. Gas chromatography-mass spectrometry (GC-MS) has also been demonstrated to be a viable alternative to HPLC-MS in the analysis of acrylamide [9-11]. Regardless if the method involves HPLC or GC, the analysis of acrylamide in complex food products such as coffee requires several pretreatment/cleanup steps before the sample can be subjected to analysis. Conventional extraction techniques using solid phase extraction (SPE) have been previously applied to purify crude sample extracts prior to the analysis of acrylamide [7-9]. However, due to the multiple steps inherent to SPE, these techniques are often cumbersome and can be time-consuming. Additionally, commercial SPE cartridges may lack the selectivity needed to discriminate the target analyte(s) from other matrix components, which can result poor method accuracy and inadequate limits of detection (LOD).

Over the past few decades, solid-phase microextraction (SPME) has become a popular extraction technique in food analysis, due to its exceptional simplicity and ease-of-use [12]. SPME is an equilibrium-based extraction/pre-concentration technique that enables the consolidation of sample preparation, cleanup, and sampling into one simple step [13]. Compared to SPE, SPME is a non-exhaustive extraction technique that uses considerably smaller sample volumes and is not susceptible to analyte breakthrough. A number of studies have exploited SPME coupled to GC-MS for the determination of acrylamide using commercially available sorbent coatings [8, 14-17]. However, these approaches have been hindered by poor LODs when analyzing acrylamide at ultra trace-levels in complex matrices. Within the last few years, Anderson and co-workers have developed a contemporary class of SPME sorbent coating based on polymeric ionic liquids (PIL) [18]. PIL coatings are highly versatile as they can be custom designed to exhibit superior selectivity towards various classes of analyte(s) by altering the chemical structures of the cation and anion while also tailoring their unique combinations [18, 19]. In addition, these coatings can be bonded and crosslinked to rugged supports, such as super-elastic nitinol (NiTi) wires [20]. The physico-chemical properties (e.g., polarity and water solubility) of acrylamide are in-line with the sorbent characteristics of PIL-based SPME materials, particularly in terms of selectivity. Therefore, they can be exploited to improve the recovery of acrylamide from water matrices such as coffee brew.

We report in this manuscript a crosslinked PIL-based SPME sorbent coating that exhibits superior sensitivity over commercially available coatings for the trace-level determination of acrylamide in brewed coffee and coffee powder when coupled to GC-MS. This report represents the first SPME method for the direct immersion analysis of acrylamide in brewed coffee and is a much faster method than the currently employed ISO SPE-HPLC-MS/MS

method taken as a reference [21]. This method requires no derivatization steps, can be easily automated, and is rugged despite the complexity of the coffee matrix.

2. Experimental

2.1. Materials

Acrylamide (99.9%), ninhydrin, asparagine (98%), glucose (99.9%), menthol (99%), ethanol (99.9%) and DAROCUR 1173 (97%) were purchased from Sigma Aldrich (St. Louis, MO, USA). Deionized water (18.2 MΩ cm) was obtained from a Milli-Q water purification system (Millipore, Bedford, MA, USA).

Table 1 lists the crosslinked PIL-based SPME fiber coatings that were evaluated in this study. All IL monomers and crosslinkers were prepared according to previously published methods [19, 20] and were fully characterized by ^{1}H-NMR (see Supporting Information). Two PIL SPME fibers possessing thin sorbent coatings (~10 μm) with varying chemical composition, namely, 50% (w/w) 1,12-di(3-vinylimidazolium)dodecane dibromide [$(VIM)_2C_{12}$] 2[Br] in 1-vinyl-3-hexylimidazolium chloride [VHIM][Cl] (Fiber **1**) and 50 % [$(VIM)_2C_{12}$] 2[NTf_2] in 1-vinylbenzyl-3-hexadecylimidazolium bis[(trifluoromethyl)sulfonyl]imide [VBHDIM][NTf_2] (Fiber **2**) were fabricated on a 1 cm portion of etched and derivatized fused silica, according to previously reported methods [19]. Additional PIL-based fibers, composed of 50% (w/w) [$(VIM)_2C_{12}$] 2[NTf_2] in [VBHDIM] [NTf_2] and possessing larger film thicknesses (~30-45 μm), were prepared on derivatized NiTi wires as described previously [20]. Fiber **3** was crosslinked to a 1 cm portion of a derivatized NiTi wire and Fibers **4**-**6** were crosslinked to 1.3 cm portions of derivatized NiTi wires.

Five commercially available SPME fibers, namely, DVB/Carboxen/PDMS (2 cm, ~30-50 μm), Carboxen/PDMS (1 cm, 75 μm), PA (1 cm, 85 μm), PDMS/DVB (1 cm, 65 μm), and PDMS (1 cm, 30 μm) were also employed in this study. These fibers were supplied by Supelco (Bellafonte, PA, USA) and were operated according the manufacturer instructions.

Commercial blends of roasted coffee and single-dose coffee capsules were kindly provided by Lavazza (Turin, Italy)

2.2. Standard and sample preparation

Individual stock solutions of acrylamide, asparagine, glucose, and menthol (used as an internal standard) were prepared in a 20 mL sealed vial by dissolving 2 mg of the pure standard in deionized water to obtain a concentration of 100 mg L^{-1}. A 200 mg L^{-1} solution of acrylamide was also prepared for preliminary studies. Working standards containing acrylamide, asparagine, and/or glucose were prepared by pipetting appropriate amounts of the stock standard(s) into a 20 mL sealed vial and further diluted with deionized water to obtain a final volume of 20 mL.

Brewed coffee samples were prepared from single-dose espresso capsules (7.6 g) using a Lavazza “A modo mio” espresso machine. Brewed coffee (19 mL) was mixed with 1 mL of 2% (w/v) ethanolic ninhydrin solution in a 20 mL sealed vial. Roasted coffee powder samples were prepared by transferring 2 g of roasted coffee powder into a 20 mL sealed vial containing 16.6 mL of deionized water. Subsequently, 44 μL of the menthol internal standard stock solution and 1 mL of 2% (w/v) ethanolic ninhydrin solution were added to the suspension. Reaction quenching by ninhydrin was carried out by placing the solution vial into a water bath thermostatted at 80 °C (with constant agitation at 1500 rpm) for 10 minutes. Sampling was performed immediately after the reaction.

2.3. Sampling and quantification of acrylamide in brewed coffee and coffee powder

Sampling was carried out by directly immersing the PIL and the commercial fibers into the sample solution under the following conditions: solution temperature: 25 °C, extraction time: 60 min; sample agitation: 1500 rpm. A temperature of 25° C was selected to prevent the formation of new acrylamide in the sample at higher temperatures. The extraction time was optimized by sampling a solution of acrylamide at 5, 15, 30, 45, 60, 90 and 120 min with both the PIL and the commercial fibers. The analytes were then desorbed in the GC inlet at 220 °C for 10 min. Following each desorption step, the PIL fibers were immersed in 20 mL of deionized water for 5 min to remove any impurities extracted from the coffee matrix. The fiber was then re-conditioned in the GC inlet for 5 minutes at 220 °C to remove residual moisture from the coating. By employing this conditioning procedure, PIL fiber **6** was subjected to more than 150 analyses and found to maintain comparable extraction performance.

Quantitative analysis of acrylamide in brewed coffee and coffee powder was performed by the method of standard addition. Brewed coffee samples were fortified with 50, 100, and 250 μg L^{-1} of acrylamide by spiking appropriate amounts of the stock standard into the sample solution. Coffee powder suspensions were spiked in the same mode to obtain 0.5, 1, and 2.5 μg g^{-1}, or 1 and 2 μg g^{-1} of acrylamide (relative to 2 g of sample). All coffee samples were also subjected to reaction quenching using ninhydrin, as described earlier. Each analysis was performed in triplicate. Once a linear calibration curve was obtained from the spiking experiments, the data was then extrapolated to calculate the theoretical concentration of acrylamide in the unadulterated sample.

2.4.Instrument parameters

Analyses were performed using a Shimadzu GC 2010 - Shimadzu QP2010-PLUS GC-MS system equipped with the GC-MS Solution 2.51 software (Shimadzu, Milan, Italy). The GC split/splitless injector temperature was maintained at 220 °C and operated in split mode (5:1) during analyte desorption. Helium was used as the carrier gas and maintained at a constant flow of 1 mL min^{-1}. A Mega-FFAP-EXT column (50 m x 0.20 mm d_c x 0.20 µm d_f) (Legnano, MI, Italy) was used for the separation. The following oven temperature program was applied: initial temperature of 50 °C (held for 0.9 min), ramped to 170 °C at 2.5 °C min^{-1}, then ramped to 250 °C at 5 °C min^{-1} (held for 5 min). The MS was operated in electron ionization mode (EI) at 70 eV for all analyses. Data were initially acquired in SCAN mode (scan rate: 666 u/s, mass range: 35–350 *m/z*) to locate and identify acrylamide in the coffee samples. Subsequently, selected ion monitoring (SIM) acquisition mode was used for the detection/quantification of acrylamide (event time: 0.2 sec, target ion: 71 *m/z*, qualifier ion: 55 *m/z*). The LOD and limit of quantitation (LOQ) were determined based on a 3:1 (signal:noise) and 10:1 ratio, respectively.

3. Results and Discussion

3.1. Selection of PIL-based SPME coating and comparison with commercial coatings

Fibers **1** and **2** were employed in the direct immersion extraction of 0.5 mg L^{-1} of acrylamide in aqueous solution. As shown in Figure 1A, Fiber **2** exhibited significantly higher extraction efficiency for acrylamide, whereas no signal was observed using Fiber **1**. Five commercial fibers were also examined in the extraction of acrylamide. As shown in Figure 1B, most of the commercial coatings did not exhibit appreciable extraction of acrylamide until its concentration was increased from 0.5 mg L^{-1} to 2 mg L^{-1}. Among the commercial coatings, the Carboxen/PDMS coating provided the best extraction of acrylamide. However, the LOD and

LOQ for this fiber were found to be 500 µg L^{-1} and 1500 µg L^{-1}, respectively, which is much higher than the typical amount of acrylamide found in coffee making it not suitable for the direct immersion analysis of acrylamide in coffee. Furthermore, a side-by-side comparison shown in Figure 2 of Fiber **2** to the commercial PA and DVB/Carboxen/PDMS coatings demonstrates that Fiber **2** was the only SPME sorbent coating that can effectively extract acrylamide from real brewed coffee samples.

A spin-coating method was developed to evenly distribute large volumes of IL monomer/crosslinker mixtures onto a 1.3 cm derivatized NiTi wire prior to the polymerization step. As shown in Figure 3, a specific length of derivatized NiTi wire was dip-coated in a mixture containing IL monomer, IL-based crosslinker (50 % w/w based on the weight of IL monomer), and DAROCUR 1173 (3 % w/w) photoinitiator. Subsequently, the SPME support was spun using a high-speed motor at 1200 RPM in the UV reactor during the crosslinking of the coating mixture to the fiber surface. A smooth and even PIL coating surface was observed on the fiber after the crosslinking step, indicating that the spin coating device was capable of evenly distributing the coating on the fiber during polymerization. By immobilizing a thicker PIL film to the support, the extraction efficiency and sensitivity of acrylamide can be significantly enhanced. Fibers **3, 4**, **5**, and **6** were fabricated using this method and possessed film thicknesses of ~30-45 µm. As shown in Figure 4, all these fibers demonstrated higher extraction of acrylamide compared to Fiber **2** (film thickness ~10 µm) with a superior sensitivity for the 1.3 cm long fibers (Fibers **4**-**6**) than the 1 cm long fiber (Fiber **3**).

3.2. Analytical performance of PIL-based fibers for the extraction of acrylamide

The analytical performance of the PIL-based SPME coatings, including the analytical precision, linearity, LOQ, and fiber-to-fiber reproducibility, was evaluated by sampling aqueous solutions spiked with acrylamide at different concentration levels. The results are

summarized in table **1**. Five consecutive analyses of a 100 µg L^{-1} acrylamide solution using Fiber **4** resulted in satisfactory analytical precision, with a 10.9% relative standard deviation (RSD). The fiber-to-fiber reproducibility was also studied for Fibers **4**, **5**, and **6** wherein a 14% RSD was obtained, indicative that the spin-coating approach used for the first time in this study produces fibers with similar film thicknesses.

The linearity of the calibration curve was also studied using Fibers **4**, **5**, and **6** at three concentration levels, namely, 10, 50, and 100 µg L^{-1}. All three PIL-based fibers showed excellent linearity with correlation coefficients (R^2) above 0.98. The LOQ for acrylamide using these PIL-based fibers was determined to be 10 µg L^{-1}, considerably lower than the LOQ of 1500 µg L^{-1} obtained using the Carboxen/PDMS fiber.

3.3. Development of the sampling method for coffee samples with reaction quenching by ninhydrin

Dunovska and coworkers previously reported the formation of acrylamide in the GC inlet when analyzing real-world samples containing free asparagine and glucose [9]. A series of experiments were carried out to determine whether these precursors can be extracted using the PIL-based coatings and if they can generate additional acrylamide during the desorption step in the GC inlet. Aqueous solutions containing 50 mg L^{-1} of asparagine, 50 mg L^{-1} of glucose, and 50 mg L^{-1} of both asparagine and glucose were individually sampled and subjected to the GC inlet using Fiber **4**. As shown in Figure 5, no additional acrylamide was observed in the analysis of the aqueous glucose solution. However, a noticeable acrylamide peak was detected in the asparagine solution. The acrylamide peak is even more pronounced when sampling was performed from the asparagine/glucose mixture. These results indicate that PIL-based sorbent coatings can also extract free asparagine and glucose from the sample matrix and that acrylamide forms during the high-temperature desorption step in the GC inlet.

A quenching reaction using ninhydrin was applied to selectively degrade free asparagine to Ruhemann's purple dye and inhibit the formation of interfering acrylamide during the desorption step. The reaction was carried out by adding 2 % (w/v) ethanolic ninhydrin solution to the sample prior to the extraction step. Figure 6 illustrates the effect of ninhydrin on the generation of interfering acrylamide. Acrylamide was not detected in either the asparagine solution or the glucose/asparagine mixture containing ninhydrin. Furthermore, the responses obtained from the acrylamide standard solutions, with and without ninhydrin, were nearly identical. These results clearly indicate that the addition of ninhydrin inhibits the formation of interfering acrylamide without compromising the selectivity of the PIL-based coatings towards native acrylamide originally present within the coffee sample.

3.4. Quantification of acrylamide in brewed coffee and coffee powder

Brewed coffee and coffee powder were analyzed to examine the selectivity, sensitivity, and robustness of the PIL-based coatings in the extraction of acrylamide from commercial coffee samples. Fiber **4** was applied in the direct immersion extraction of brewed *Coffea robusta* and the concentration of acrylamide in these samples was determined by the method of standard addition. Individual brewed coffee samples were spiked with varying amounts of acrylamide (i.e. 50, 100, and 250 µg L^{-1}) to generate a calibration curve. The results produced a calibration curve with excellent linearity (R^2 > 0.98). The analytical precision for three consecutive analyses were below 7.4 % RSD at every concentration level. The extrapolated concentration of acrylamide in the brewed *Coffea robusta* coffee was determined to be 268 µg L^{-1}.

The headspace extraction of coffee powder using PIL-based coatings revealed that these sorbent phases are also capable of recovering acrylamide in this sampling mode. However, since the headspace of coffee powder samples contains a complex mixture of volatile components that often possess ion fragments at 71 and 55 *m/z*, it is not feasible to discriminate

acrylamide from these interferences. To address this, the coffee powder samples were suspended in water and analyzed by direct immersion SPME. Quantitative analysis was carried out by the method of standard addition. The suspensions, each containing 2 g of coffee powder, were fortified with 0.5, 1, and 2.5 μg g^{-1} of acrylamide to generate a calibration curve. The analyses were performed in triplicate using Fibers **4** and **6**. Good linearity of the calibration curve was obtained (R^2 > 0.98) and analytical precision was below 5.8 % (RSD) at every concentration level. The extrapolated concentration of acrylamide in the unadulterated coffee powder was 2.34 μg g^{-1} and 2.00 μg g^{-1} using Fibers **4** and **6**, respectively, and the fiber-to-fiber reproducibility was 11.1 % (RSD).

3.5. Evaluation of the method for the determination of acrylamide within coffee powder blends

The accuracy of the PIL-based SPME method was verified by analyzing six coffee powder samples composed of different coffee blends. The obtained results were then compared with the ISO SPME-LC-MS/MS method. Table S1 (Supporting Information) compares the observed concentration of acrylamide in the coffee powder samples between both methods. The developed PIL-based SPME method showed slightly higher acrylamide concentrations compared to the reference method. However, the results obtained between the two methods are comparable and the %RSD are within the limit of acceptance based on the Horwitz-Thompson equation [22].

4. Conclusions

A direct-immersion SPME method using a crosslinked PIL-based coating was applied to determine trace levels of acrylamide in brewed coffee and coffee powder. The PIL-based coating exhibited much higher sensitivity towards acrylamide compared to the commercially

available SPME sorbent coatings. A spin coating method capable of forming a thick and homogeneous coating of the PIL on the fiber support was developed to further increase the extraction efficiency. The PIL fiber produced a limit of quantitation for acrylamide of 10 μg L^{-1} and exhibited good analytical precision, linearity, and fiber-to-fiber reproducibility. Due to the ruggedness and durability of the crosslinked PIL coating, sampling can be performed in complex matrices such as coffee without sacrificing analytical precision and sensitivity. The addition of ninhydrin to the sample matrix minimized the production of interfering acrylamide from free asparagine and glucose within the sample, leading to satisfactory method accuracy.

Acknowledgements

J.L.A. acknowledges support from the Chemical Measurement and Imaging Program in the Division of Chemistry at the National Science Foundation (CHE-1413199). C.C. and C.B. acknowledge support from the Ricerca Locale (Ex 60% 2014) of the University of Turin, Turin (Italy). The authors are grateful to Lavazza SpA (Turin, Italy) for supplying coffee samples, the espresso machine, and technical support.

References

[1] A.A. Elbashir, M.M. Omar, W.A. Ibrahim, O.J. Schmitz, H.Y. Aboul-Enein, Acrylamide analysis in food by liquid chromatographic and gas chromatographic methods, Crit. Rev. Anal. Chem., 44 (2014) 107-141.

[2] D.S. Mottram, B.L. Wedzicha, A.T. Dodson, Acrylamide is formed in the Maillard reaction, Nature, 419 (2002) 448-449.

[3] R.H. Stadler, I. Blank, N. Varga, F. Robert, J. Hau, P.A. Guy, M.C. Robert, S. Riediker, Acrylamide from Maillard reaction products, Nature, 419 (2002) 449-450.

[4] EFSA CONTAM Panel (EFSA Panel on Contaminants in the Food Chain), Scientific Opinion on acrylamide in food, EFSA J. 13 (2015) 4104-4321.

[5] J. Park, L.M. Kamendulis, M.A. Friedman, J.E. Klaunig, Acrylamide-induced cellular transformation, Toxicol. Sci., 65 (2002) 177-183.

[6] J.M. Rice, The carcinogenicity of acrylamide, Mutat. Res.-Gen. Tox. En., 580 (2005) 3-20.

[7] P.C. Aguas, M.J. Fitzhenry, G. Giannikopoulos, P. Varelis, Analysis of acrylamide in coffee and cocoa by isotope dilution liquid chromatography-tandem mass spectrometry, Anal. Bioanal. Chem., 385 (2006) 1526-1531.

[8] J. Oracz, E. Nebesny, D. Zyzelewicz, New trends in quantification of acrylamide in food products, Talanta, 86 (2011) 23-34.

[9] L. Dunovska, T. Cajka, J. Hajslova, K. Holadova, Direct determination of acrylamide in food by gas chromatography-high-resolution time-of-flight mass spectrometry, Anal. Chim. Acta, 578 (2006) 234-240.

[10] A. Pittet, A. Perisset, J.M. Oberson, Trace level determination of acrylamide in cereal-based foods by gas chromatography-mass spectrometry, J. Chromatogr. A, 1035 (2004) 123-130.

[11] Y.H. Zhu, G.R. Li, Y.P. Duan, S.Q. Chen, C. Zhang, Y.F. Li, Application of the standard addition method for the determination of acrylamide in heat-processed starchy foods by gas chromatography with electron capture detector, Food Chem., 109 (2008) 899-908.

[12] E.A. Souza-Silva, E. Gionfriddo, J. Pawliszyn, A critical review of the state of the art of solid-phase microextraction of complex matrices II. Food analysis, Trac-Trend Anal. Chem., 71 (2015) 236-248.

[13] C.L. Arthur, J. Pawliszyn, Solid-Phase Microextraction with Thermal-Desorption Using Fused-Silica Optical Fibers, Anal. Chem., 62 (1990) 2145-2148.

[14] L.B. Chen, H.Z. Liu, P. Yu, J.Y. Zhao, X. Chen, Determination of Acrylamide in Foods by Solid Phase Microextraction-Gas Chromatography, Food Sci. Biotechnol., 18 (2009) 895-899.
[15] A.R. Ghiasvand, S. Hajipour, Direct determination of acrylamide in potato chips by using headspace solid-phase microextraction coupled with gas chromatography-flame ionization detection, Talanta, 146 (2016) 417-422.
[16] A.F. Lagalante, M.A. Felter, Silylation of acrylamide for analysis by solid-phase microextraction/gas chromatography/ion-trap mass spectrometry, J. Agr. Food Chem., 52 (2004) 3744-3748.
[17] M.R. Lee, L.Y. Chang, J.P. Dou, Determination of acrylamide in food by solid-phase microextraction coupled to gas chromatography-positive chemical ionization tandem mass spectrometry, Anal. Chim. Acta, 582 (2007) 19-23.
[18] T.D. Ho, A.J. Canestraro, J.L. Anderson, Ionic liquids in solid-phase microextraction: A review, Anal. Chim. Acta, 695 (2011) 18-43.
[19] T.D. Ho, H. Yu, W.T.S. Cole, J.L. Anderson, Ultraviolet photoinitiated on-fiber copolymerization of ionic liquid sorbent coatings for headspace and direct immersion solid-phase microextraction, Anal. Chem., 84 (2012) 9520-9528.
[20] T.D. Ho, B.R. Toledo, L.W. Hantao, J.L. Anderson, Chemical immobilization of crosslinked polymeric ionic liquids on nitinol wires produces highly robust sorbent coatings for solid-phase microextraction, Anal. Chim. Acta, 843 (2014) 18-26.
[21] European Standard EN 16618:2015, Food analysis - Determination of acrylamide in food by liquid chromatography tandem mass spectrometry (LC-ESI-MS/MS)
[22] M. Thompson, Recent trends in inter-laboratory precision at ppb and sub-ppb concentrations in relation to fitness for purpose criteria in proficiency testing, Analyst, 125 (2000) 385-386.

Figure Captions

Figure 1: Comparison of the extraction efficiency of acrylamide using PIL-based SPME coating and commercial fibers. A: Direct immersion extraction of acrylamide in an aqueous solution (0.5 mg L^{-1}) using Fiber **1** (pink) and Fiber **2** (black); B: Direct immersion extraction of acrylamide in an aqueous solution (2 mg L^{-1}) using the commercial fibers DVB/Carboxen/PDMS (green), Carboxen/PDMS (blue), PA (black), PDMS/DVB (pink), and PDMS (red).

Figure 2: Comparison of the direct immersion extraction of acrylamide (AA) from coffee brew prepared from a single-dose espresso capsule without addition of ninhydrin employing Fiber **2** (black), DVB/Carboxen/PDMS (pink), and PA (blue).

Figure 3: Schematic illustrating the spin-coating approach used to prepare PIL SPME fibers with larger film thicknesses (~30-45 μm). The home-made spin-coating device was fabricated using a 1.5-3 VDC high-speed motor and a 1.5 V C battery.

Figure 4: Comparison of the extraction efficiency towards acrylamide (AA) for different PIL-based SPME sorbent coatings prepared in this study. Profiles obtained by direct immersion sampling of a 500 μg L^{-1} acrylamide standard solution with Fiber **2** (green), Fiber **3** (pink), Fiber **4** (blue), Fiber **5** (brown), Fiber **6** (black).

Figure 5: Comparison between the levels of acrylamide observed in various aqueous solutions by direct immersion SPME using Fiber **4**: 0.5 mg L^{-1} of acrylamide (black); pure water (green), 50 mg L^{-1} of glucose aqueous solution (brown), 50 mg L^{-1} asparagine aqueous solution (blue) and a mixture of asparagine and glucose aqueous solution, each at 50 mg L^{-1} (pink).

Figure 6: Levels of acrylamide (AA) observed in various aqueous solutions by direct immersion SPME using Fiber **4.** (Green) 0.3 mg L^{-1} acrylamide standard solution; (pink) 0.3 mg L^{-1} acrylamide standard solution after reaction quenching by ninhydrin; (black) mixture of asparagine and glucose aqueous solution, each at 50 mg L^{-1}; (blue) mixture of asparagine and glucose aqueous solution, each at 50 mg L^{-1}, after reaction quenching by ninhydrin; (brown) 50 mg L^{-1} asparagine solution after reaction quenching by ninhydrin.

Table 1. Composition and abbreviations of all PIL-based SPME sorbent coatings examined in this study

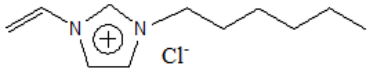
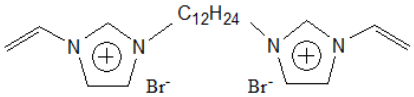
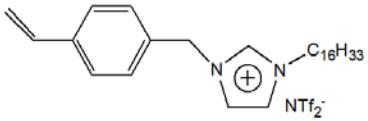
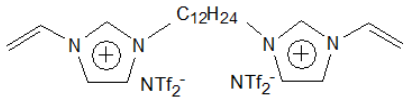
	IL monomer	IL-based crosslinker	Support	Coating method	Length of the sorbent coating	Approximate film thickness
Fiber **1**	 [VHIM][Cl]	 [(VIM)$_2$C$_{12}$] 2[Br]	Silica	Dip coating	1 cm	~10 µm
Fiber **2**	 [VBHDIM][NTf$_2$]	 [(VIM)$_2$C$_{12}$] 2[NTf$_2$]	Silica	Dip coating	1 cm	~10 µm
Fiber **3**			NiTi wire	Spin coating	1 cm	~35 µm
Fiber **4**			NiTi wire	Spin coating	1.3 cm	~35 µm
Fiber **5**			NiTi wire	Spin coating	1.3 cm	~35 µm
Fiber **6**			NiTi wire	Spin coating	1.3 cm	~35 µm

Table 2. Analytical performance of the 1.3 cm long spin coated 50% (w/w) $[(VIM)_2C_{12}]$ 2$[NTf_2]$ in [VBHDIM] $[NTf_2]$ PIL-based fiber for the extraction of acrylamide

LOQ	10 µg L^{-1}
Linearity	0.984
Analytical precision (% RSD 100 µg L^{-1})	10.9% (*n=5*)
Fiber-to-fiber reproducibility (% RSD 100 µg L^{-1})	14.0% (*n=3*)

Figure 1

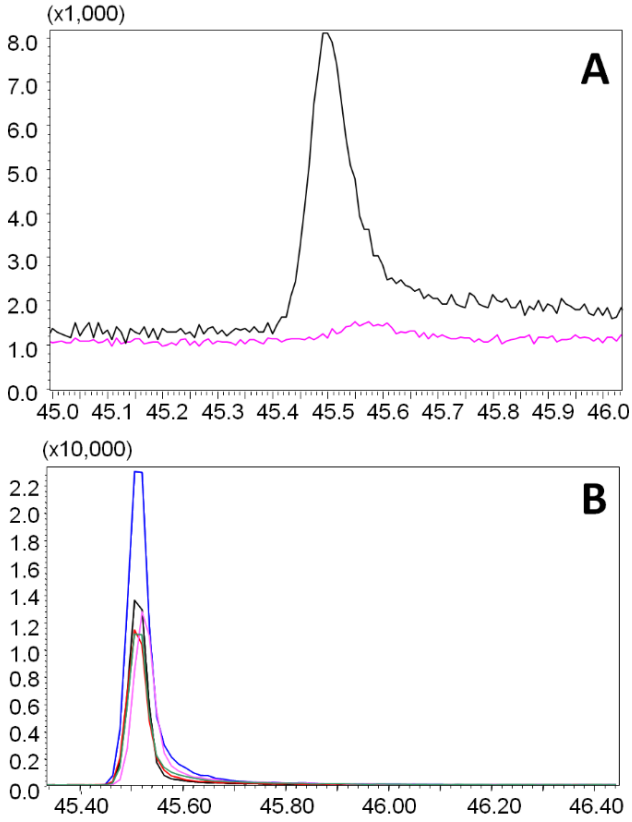

(x1,000)
8.0
7.0
6.0
5.0
4.0
3.0
2.0
1.0
0.0
A
45.0 45.1 45.2 45.3 45.4 45.5 45.6 45.7 45.8 45.9 46.0
(x10,000)
2.2
2.0
1.8
1.6
1.4
1.2
1.0
0.8
0.6
0.4
0.2
0.0
B
45.40 45.60 45.80 46.00 46.20 46.40

Figure 2

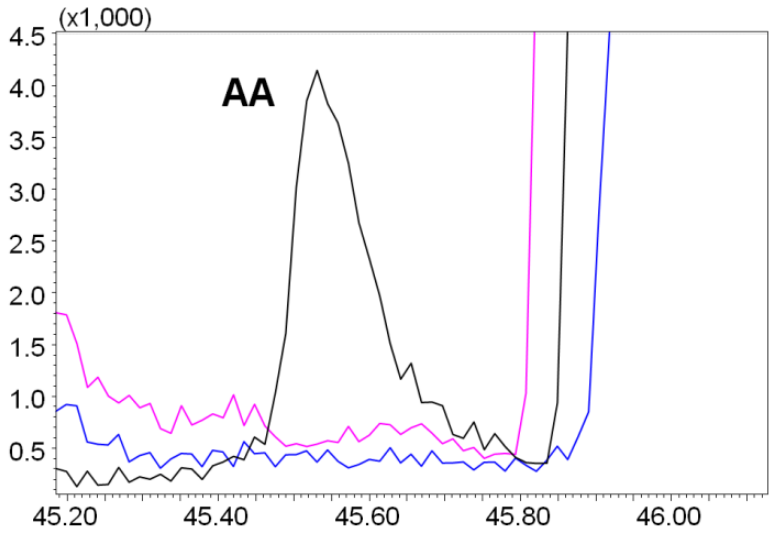


Figure 3.

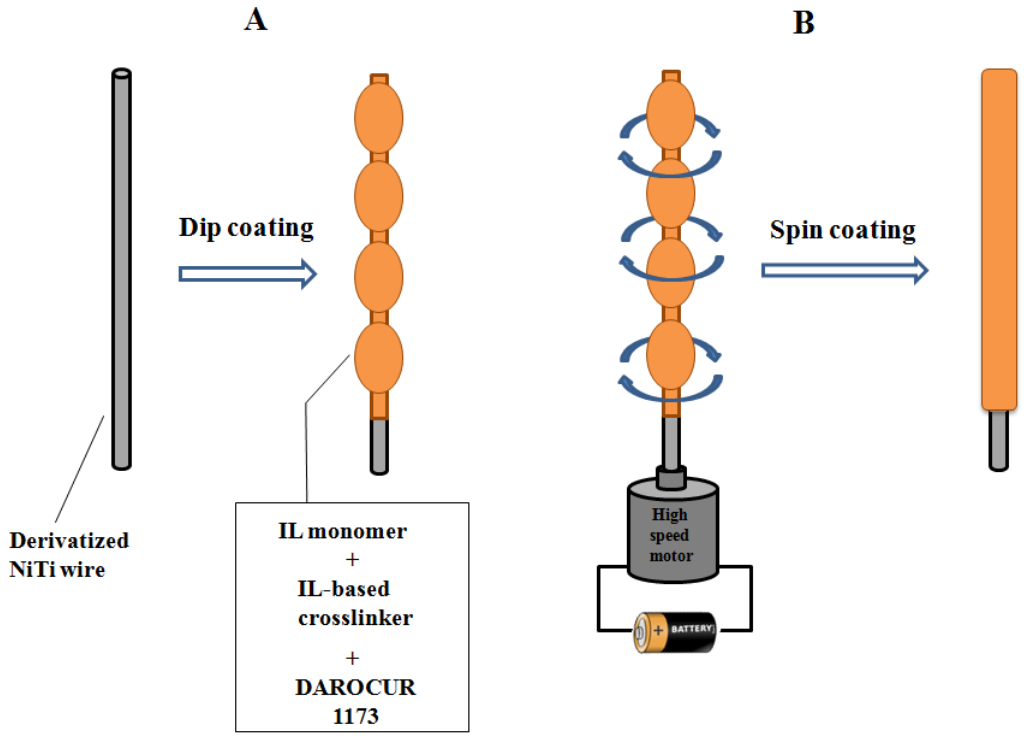

A
B
Dip coating
Spin coating
Derivatized
NiTi wire
IL monomer
+
IL-based
crosslinker
+
DAROCUR
1173
High
speed
motor
BATTERY

Figure 4

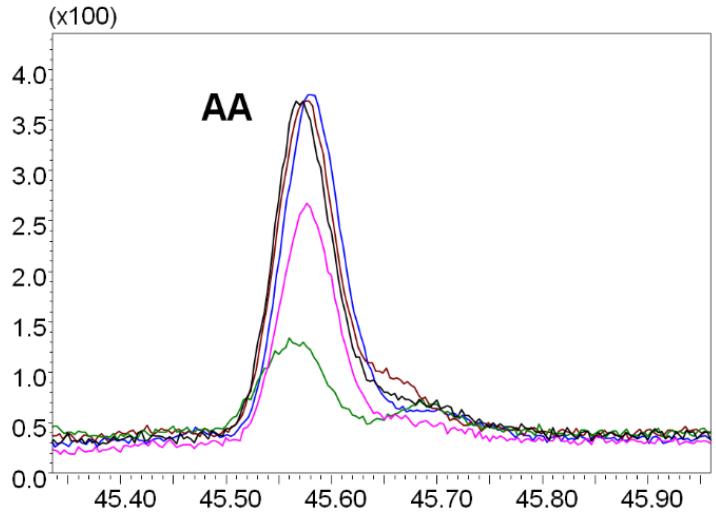


Figure 5

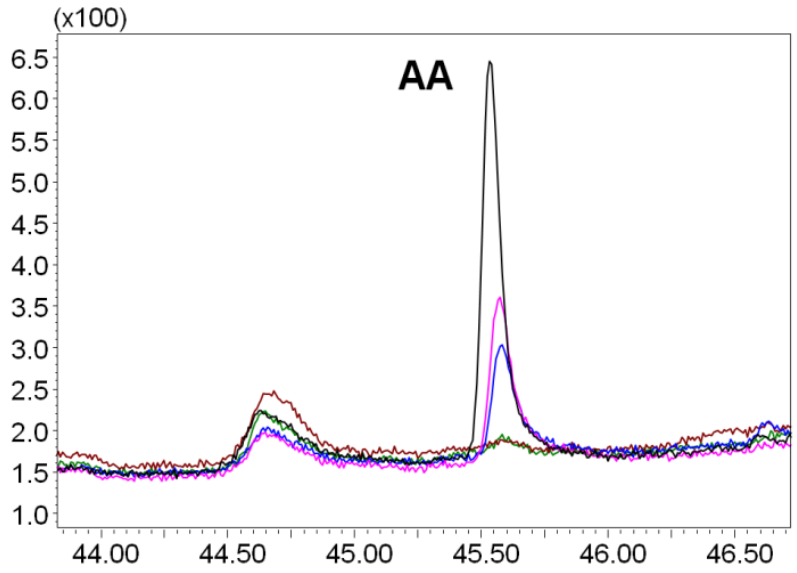
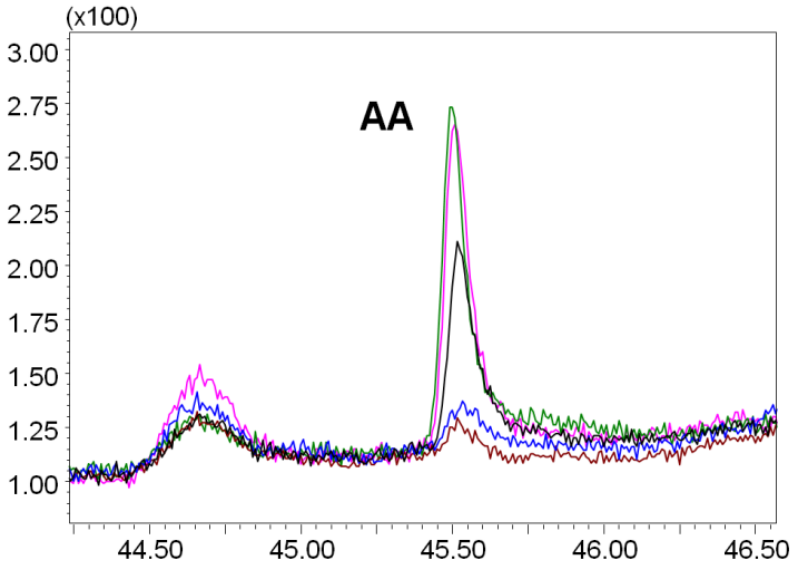

(x100)
AA
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
44.00
44.50
45.00
45.50
46.00
46.50

Figure 6


(x100)
3.00
2.75
2.50
2.25
2.00
1.75
1.50
1.25
1.00
AA
44.50
45.00
45.50
46.00
46.50

Supporting Information

Determination of Acrylamide in Brewed Coffee and Coffee Powder using Polymeric Ionic Liquid-based Sorbent Coatings in Solid-Phase Microextraction Coupled to Gas Chromatography-Mass Spectrometry

Cecilia Cagliero,[b] Tien D. Ho,[a] Cheng Zhang, [a] Carlo Bicchi,[b] and Jared L. Anderson[a]

[a] Department of Chemistry, Iowa State University, Ames, IA 50011 USA

[b]Dipartimento di Scienza e Tecnologia del Farmaco, Università degli Studi di Torino, I-10125 Torino, Italy

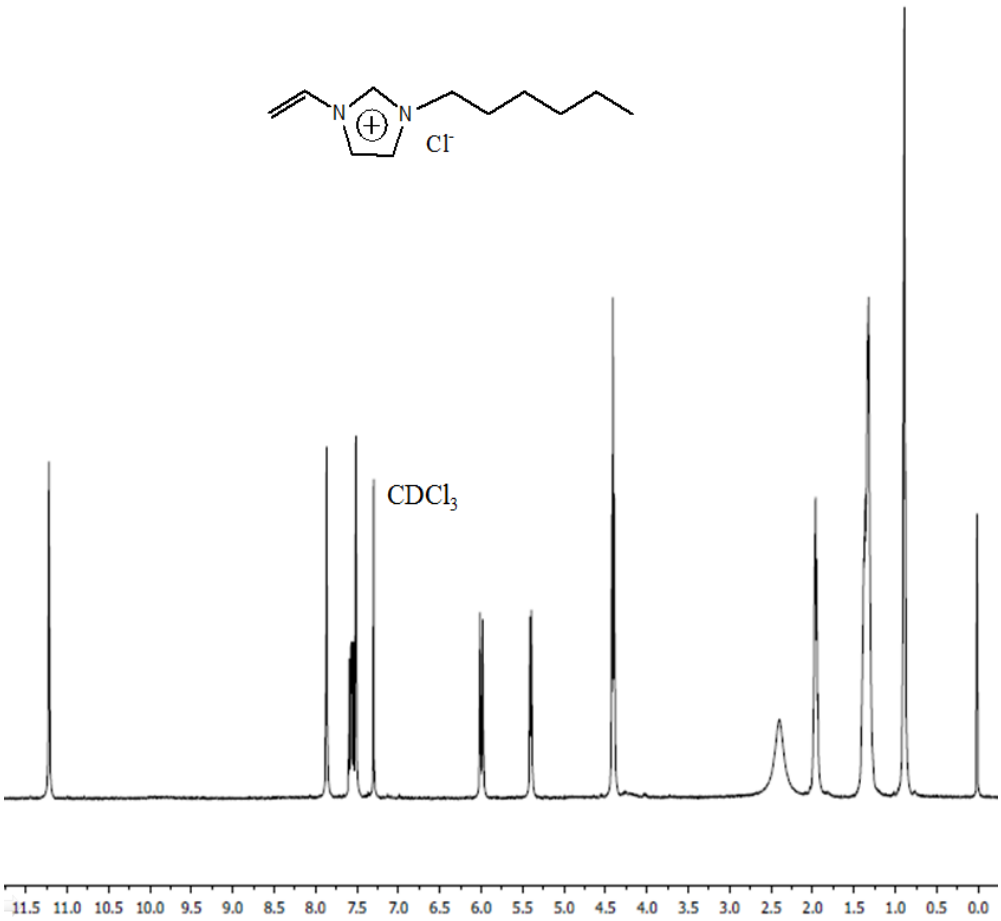


Figure S1. ^{1}H NMR (Bruker DRX-500 MHz, $CDCl_3$) spectrum of [VHIM][Cl]: 11.21 (s, 1H), 7.86 (s, 1H), 7.55 (m, 1H), 7.49 (s, 1H), 5.98 (s, 1H), 5.38 (s, 1H), 4.40 (t, 2H), 1.96 (m, 2H), 1.35 (m, 6H), 0.87 (t, 3H).

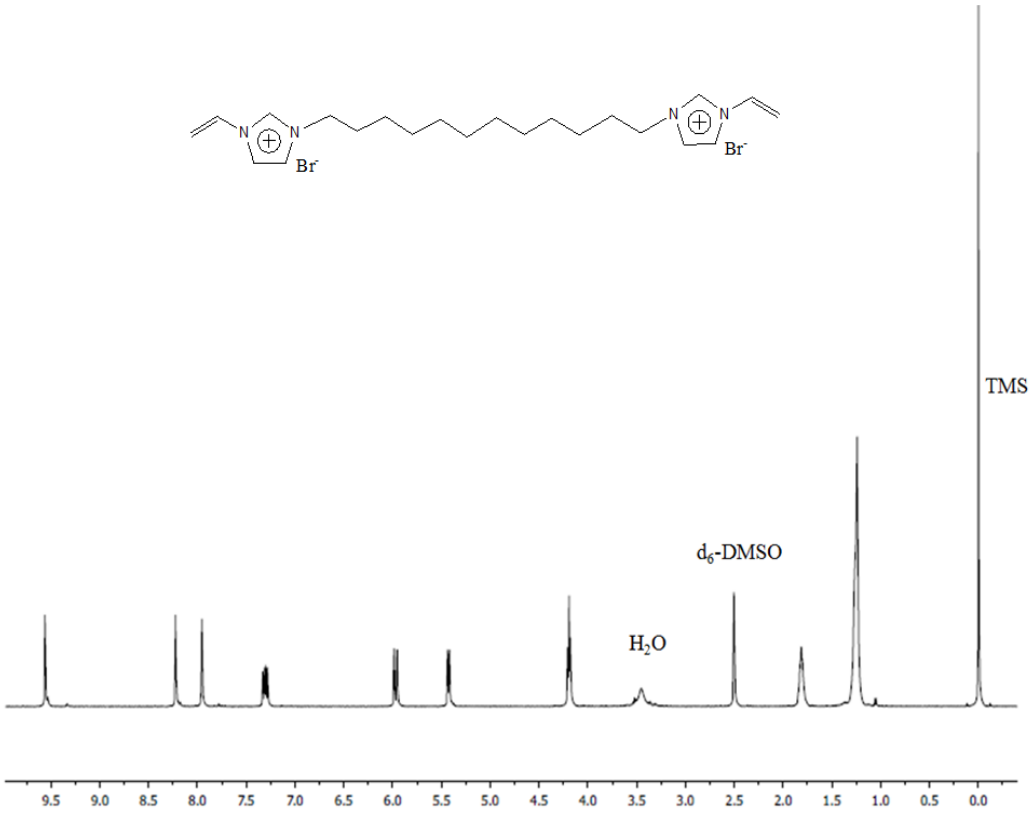


Figure S2. ^{1}H NMR (Bruker DRX-500 MHz, d_6-DMSO) spectrum of $[(VIM)_2C_{12}]$ 2[Br]: 9.55 (s, 2H), 8.22 (s, 2H), 7.95 (s, 2H), 7.30 (m, 2H), 5.97 (d, 2H), 5.42 (d, 2H), 4.18 (t, 4H), 1.80 (t, 4H), 1.24 (m, 16H).

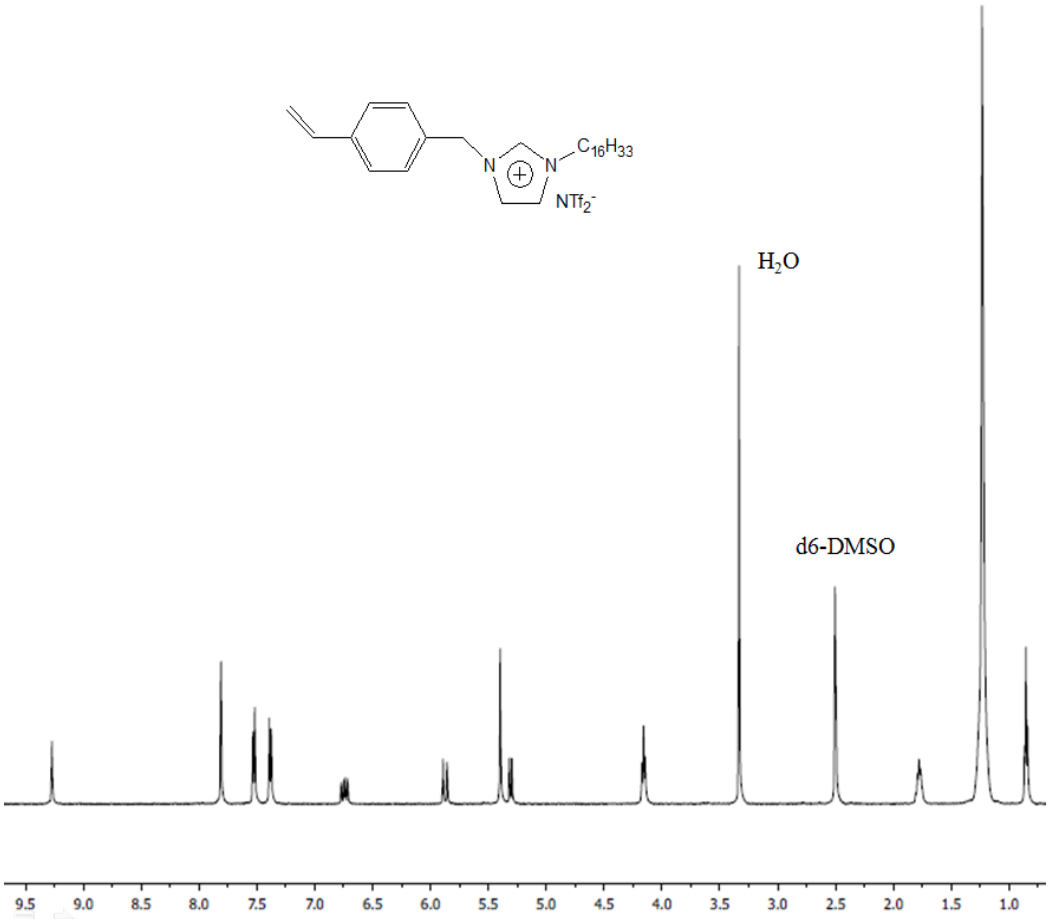


Figure S3. ^{1}H NMR (Bruker DRX-500 MHz, d_6-DMSO) spectrum of [VBHDIM][NTf_2]: 9.25 (s, 1H), 8.22 (s, 2H), 7.78 (d, 2H), 7.51 (d, 2H), 7.36 (d, 2H), 6.72 (m, 1H), 5.86 (d, 1H), 5.41 (d, 2H), 5.28 (m, 1H), 4.12 (t, 2H), 1.77 (t, 2H), 1.22 (m, 26H), 0.84 (t, 3H).

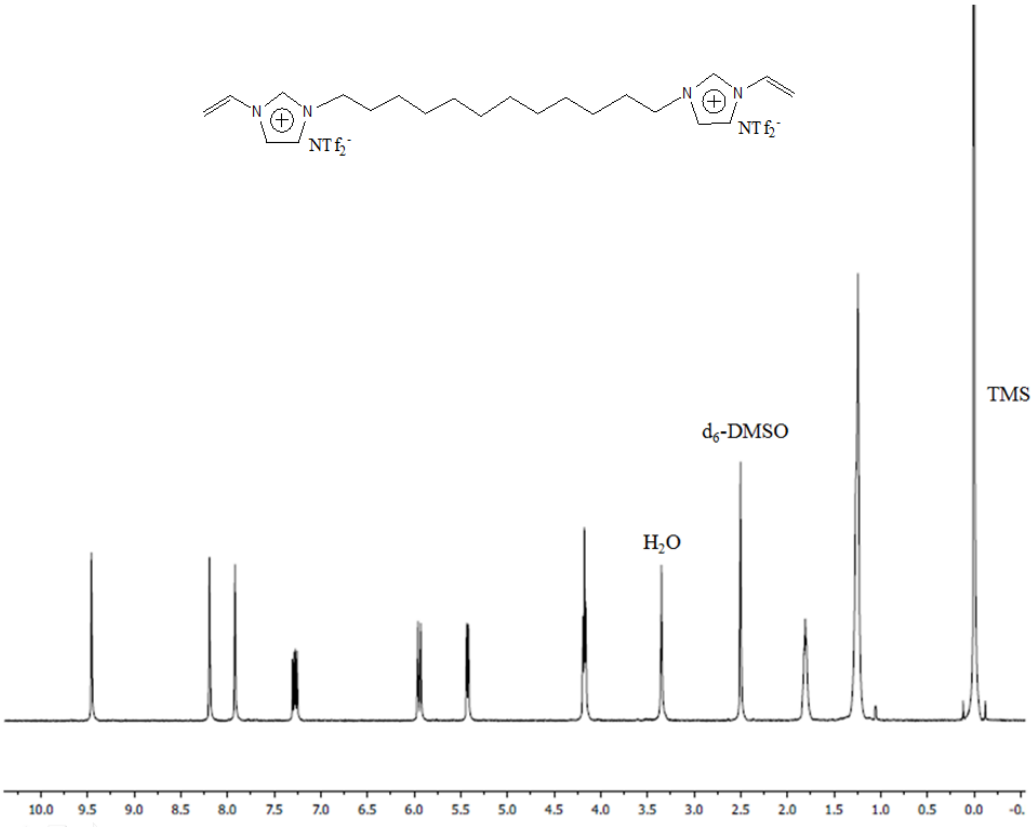


Figure S4. ^{1}H NMR (Bruker DRX-500 MHz, d_6-DMSO) spectrum of $[(VIM)_2C_{12}]$ $2[NTf_2]$: 9.43 (s, 2H), 8.18 (s, 2H), 7.91 (s, 2H), 7.26 (m, 2H), 5.93 (d, 2H), 5.40 (d, 2H), 4.16 (t, 4H), 1.79 (t, 4H), 1.23 (m, 16H).

Table S1. Comparison of the results obtained with the ISO and the PIL-SPME methods for the determination of acrylamide concentration within coffee powder blends

Sample #	ISO method results[a] (μg/kg)	PIL-SPME results (μg/kg)	% RSD	Slope ± error	Linearity
1	230	311	21.1	675.67 ± 76.74	0.987
2	252	357	24.2	585.67 ± 2.14	0.991
3	302	422	23.5	556.00 ± 52.35	0.974
4	315	364	10.3	537.67 ± 87.56	1.000
5	334	466	23.4	288.33 ± 33.68	0.987
6	391	556	24.2	293.33 ± 0.77	1.000

[a]: The ISO method results were obtained using the ISO method EN 16618:2015 E: Food analysis - Determination of acrylamide in food by liquid chromatography tandem mass spectrometry (LC-ESI-MS/MS)