Journal of Chromatography A, 1345 (2014) 17–28

Contents lists available at ScienceDirect

Journal of Chromatography A

journal homepage: www.elsevier.com/locate/chroma

Fast determination of 22 sulfonamides from chicken breast muscle using core–shell nanoring amino-functionalized superparamagnetic molecularly imprinted polymer followed by liquid chromatography-tandem mass spectrometry☆

Yong-Gang Zhao[a,b], Li-Xin Zhou[c], Sheng-Dong Pan[a,b], Ping-Ping Zhan[d], Xiao-Hong Chen[a,b], Mi-Cong Jin[a,b,*]

[a] Zhejiang Provincial Key Laboratory of Health Risk Appraisal for Trace Toxic Chemicals, Ningbo Municipal Center for Disease Control and Prevention, Ningbo, Zhejiang 315010, China
[b] Ningbo Key Laboratory of Poison Research and Control, Ningbo Municipal Center for Disease Control and Prevention, Ningbo, Zhejiang 315010, China
[c] Medical School, Ningbo University, Ningbo, Zhejiang 315211, China
[d] School of Marine Sciences, Ningbo University, Ningbo, Zhejiang 315211, China

ARTICLE INFO

Article history:
Received 9 January 2014
Received in revised form 6 April 2014
Accepted 9 April 2014
Available online 18 April 2014

Keywords:
Ultra-fast liquid chromatography-tandem quadrupole mass spectrometry (UFLC-MS/MS)
Core–shell nanoring amino-functionalized superparamagnetic molecularly imprinted polymer (CS-NR-Mag-MIP)
Dispersive micro-solid-phase extraction (d-μ-SPE)
Matrix effect
Sulfonamides
Chicken breast muscle

ABSTRACT

A novel, simple and sensitive method was developed for the simultaneous determination of 22 sulfonamides (SAs) in chicken breast muscle by using the dispersive micro-solid-phase extraction (d-μ-SPE) procedure combined with ultra-fast liquid chromatography-tandem quadrupole mass spectrometry (UFLC-MS/MS). The excellent core–shell nanoring amino-functionalized superparamagnetic molecularly imprinted polymer (CS-NR-Mag-MIP) was used as sorbent, and the main factors affecting the extraction efficiency were investigated in detail. All target compounds showed good linearities in the tested range with correlation coefficients (r) higher than 0.9980. The mean recoveries were in the range of 85.0–112.2% at low, medium and high concentration levels (0.1, 1.0 and 5.0 ng/g). The intra-day and inter-day relative standard deviations (RSDs) were lower than 6.0% and 8.9%, respectively. The limits of quantification for the 22 SAs were between 0.013 and 0.099 ng/g. The developed d-μ-SPE-UFLC-MS/MS method had been successfully applied to the chicken breast muscle samples for food-safety risk monitoring in Zhejiang Province, China. The results showed sulfamethazine, sulfamethoxazole and sulfaquinoxaline in five out of three hundred collected samples were detected with concentrations of 0.418–2.28, 16.4 and 2.93 ng/g, respectively. It was confirmed that the CS-NR-Mag-MIP was a kind of highly effective d-μ-SPE materials used for the SAs analyses.

1. Introduction

A large number of antibiotics are allowed to be used in food-producing animals, but sulfonamides (SAs) are the most widely employed [1,2]. SAs have played an important role as effective chemotherapeutics in veterinary medicine practice due to their inexpensiveness and wide-spectrum antimicrobial activity [3]. However, SAs have some side effects including emiction and hematopoiesis turbulence thus long-term use increases the risk of drug resistance and causes detrimental ecological effects even at low concentration [4,5]. To safeguard human health from this specific risk, the European Union (EU) has established a maximum residue limit (MRL) for SAs at the total level of 100 ng/g in foods of animal origin, such as meat, milk, and eggs [6]. Therefore, it is of great importance to develop a simple, rapid and efficient method for the determination of SAs residue in foods of animal origin.

The presence of SAs residue in chicken breast muscle is normally controlled by health authorities and by the food industry to preserve the product quality. Although many analytical methodologies have been proposed for the identification and quantification of these drugs, precision and cost-effectiveness are becoming an important issue for all laboratories involved in the residue

☆ Presented at the 40th International Symposium on High Performance Liquid Phase Separations and Related Techniques (HPLC 2013 Hobart), Hobart, Tasmania, Australia, 18–21 November 2013.

* Corresponding author at: Ningbo Key Laboratory of Poison Research and Control, Ningbo Municipal Center for Disease Control and Prevention, Ningbo, Zhejiang 315010, China. Tel.: +86 574 87274559; fax: +86 574 87277153.
E-mail address: jmcjc@163.com (M.-C. Jin).

http://dx.doi.org/10.1016/j.chroma.2014.04.028
0021-9673/

analyses [7–12]. Nowadays, liquid chromatography coupled with tandem quadrupole mass spectrometry (LC-MS/MS) is an attractive alternative due to its simplicity, separation efficiency and excellent sensitivity and selectivity for trace multiresidue analyses [6,7,13–18]. In addition to the advantages mentioned above, LC-MS/MS techniques allow to identify analogous compounds, i.e., SAs isomers, which have similar retention times (t_R) and similar fragment ions provided by the tandem mass spectra information.

In LC-MS/MS method, matrix effects need to be evaluated during its development because the trueness and precision of a LC-MS/MS method could be significantly affected by this phenomena [19,20]. Therefore, in addition to chromatographic analysis, the sample pretreatment is an important aspect of LC-MS/MS method. Several pretreatment methods have been described for the extraction of SAs, such as liquid–liquid extraction (LLE) [21], solid-phase extraction (SPE) [22], solid-phase micro extraction (SPME) [23], and so on. All of these protocols, however, are laborious and time consuming. Luckily, several other cleanup methods based on the traditional SPE technique, such as dispersive solid-phase extraction (dSPE), micro-solid-phase extraction (μ-SPE) and dispersive micro-solid-phase extraction (d-μ-SPE), have been widely used for multi-residue analyses [24–31]. Furthermore, different types of solid-phase sorbents such as hybrid silica monolith [32], cation exchange resin [17], C8 [15], C18 [33], Poly-MAA-EGDMA [23], florisil [34], Alumina N [35] and PDMS/DVB [36] have been reported for the determination of SAs in food samples. However, the use of these sorbents in d-μ-SPE involves difficulties for the quick separation of spent sorbent from the solution and low selectivity to SAs. Although magnetic multiwalled carbon nanotubes (m-MWCNTs) had been used for extraction of SAs from eggs, low selectivity to target molecules had been limited their further application [6].

Molecularly imprinted polymers (MIPs) exhibiting high selectivity and affinity to the predetermined molecule are now viewed as a fast-growing research. Given its application to a wide range of target molecules such as drugs, herbicides, carbohydrates, amino acids, and other biologically and environmentally important molecules, MIPs have become powerful tools for use in certain fields, for instance, enantiomer separation, catalysis, chemical sensors, solid-phase extraction, chromatography and drug delivery [37–43]. In our previous work, a novel core–shell nanoring amino-functionalized magnetic molecularly imprinted polymer (CS-NR-Mag-MIP) was synthesized by ultrasound-assisted suspension polymerization [44]. Therefore, we inspirited that CS-NR-Mag-MIP with SAs as target molecules can be a powerful sorbent to carry out d-μ-SPE procedure for the extraction of SAs from chicken breast muscle samples. To the best of our knowledge, the application of d-μ-SPE coupled with LC-MS/MS method for quantification of trace level SAs in chicken breast muscle sample has never been reported.

In this work, a novel, fast and simple method, which combines d-μ-SPE and magnetic separation technology together for the extraction and determination of 22 SAs in chicken breast muscle samples, is presented. The CS-NR-Mag-MIP does not need to be packed into the SPE cartridge but dispersed in the sample extraction solution instead. Extraction and enrichment, the two most time-consuming procedures in sample pretreatment, could be fulfilled simultaneously by simply blending and stirring the extraction solvent, and the use of CS-NR-Mag-MIP. When the extraction was completed, the CS-NR-Mag-MIP was rapidly separated from the sample matrix by the external magnetic field, and finally the SAs eluted from the sorbent was analyzed by UFLC-MS/MS without further enrichment. The excellent sensitivity and selectivity of the developed method for 22 SAs is investigated in laboratory batch tests, and it can be applied to the routine analyses for the determination of trace SAs residue in chicken breast muscle samples.

2. Experimental

2.1. Reagents and materials

Acetonitrile (ACN) and formic acid of HPLC grade were purchased from Merck (Darmstadt, Germany). Ammonium acetate used for mobile phase was of HPLC-grade obtained from Sigma (Steinheim, Germany). The standards of sulfaguanidine, sulfacetamide, sulfisomidine, sulfadiazine, sulfathiazole, sulfapyridine, sulfamerazine, sulfamethazine, sulfamethizole, sulfisozole, sulfamethoxypyridazine, sulfamethoxydiazine, sulfamonomethoxine, sulfachloropyridazine, sulfaclozine, sulfadoxine, sulfamethoxazole, sulfisoxazole, sulfabenzamide, sulfadimethoxine, sulfaquinoxaline, and sulfaphenazole were purchased from Dr. Ehrenstorfer GmbH (Augsburg, Germany). Ferric chloride, ferrous sulfate, oleic acid (OA), methyl methacrylate (MMA), tetraethylenepentamine (TEPA), glycidylmethacrylate (GMA), divinylbenzene (DVB), polyvinyl alcohol (PVA 217) and benzoyl peroxide (BPO) of analytical grade were purchased from Sinopharm Chemical Reagent Co., Ltd. (Shanghai, China). Ultrapure water was obtained using a MilliQ gradient ultrapure water system. Chicken breast muscle samples were collected from the local markets.

CS-NR-Mag-MIP used in the experiment was prepared with SAs as target molecules according to the reported procedure [44]. Control core–shell nanoring amino-functionalized magnetic non-imprinted polymer (CS-NR-Mag-NIP) nanoparticle was prepared using the same procedures described above but without addition of the SAs template. The CS-NR-Mag-MIP obtained was ringed and exhibited a well-defined core–shell configuration. The Fe_3O_4 nanoparticles exhibited a uniform morphology with an average particle size of about 20 nm. The inside and outside diameters of the ringed CS-NR-Mag-MIP was 70 and 150 nm, respectively, and the coated shell had an average thickness of approximately 10 nm. CS-NR-Mag-MIP showed a saturation magnetization value of 7.28 emu/g. The preparation procedure of CS-NR-Mag-MIP was illustrated in Fig. 1.

2.2. Equipment

Ultra-fast liquid chromatography-tandem quadrupole mass spectrometry (UFLC-MS/MS) analyses were performed using a Prominence UFLC XR system equipped with a DGU-20A3 degasser, a CTO-20AC column oven, a LC-20AD pump, a SIL-20AC autosampler (Shimadzu Corporation, Tokyo, Japan) and an AB SCIEX TRIPLE QUAD^{TM} 5500 mass spectrometer (Applied Biosystems, Foster City, CA, USA). The UFLC-MS/MS system was controlled and data were analyzed on a computer equipped with Applied Biosystems/MDS Sciex Analyst 1.5.1 (Applied Biosystems, Foster City, CA, USA).

2.3. Application of the CS-NR-Mag-MIP d-μ-SPE for extraction of SAs from sample

A 10.0 g of chicken breast muscle sample was transferred to a polypropylene centrifuge tube (50.0 mL) and homogenized for 2.0 min using an Ultra Turraxmixer. Then exactly 1.0 g of chicken breast muscle sample was transferred to another polypropylene centrifuge tube (50.0 mL) and then 5.0 mL of deionized water (pH 5.0) was added. And the mixture was immediately machine-shaken for 1.0 min and radiated by microwave for 1.0 min, then centrifuged at 15,000 rpm for 2.0 min. The residues were extracted with 5.0 mL of deionized water again. Afterwards, 1.0 mL of the supernatant was transferred to a polypropylene centrifuge tube (2.0 mL). Subsequently, 15 mg of CS-NR-Mag-MIP was added, and the mixture was immediately machine-shaken for 10.0 min, then separation under a magnetic field for 1 min. The adsorbed-CS-NR-Mag-MIP was washed sequentially with 2.0 mL of water and 2.0 mL of 10%