Available online at www.sciencedirect.com
ScienceDirect
Food Chemistry 108 (2008) 760–767
Food Chemistry
www.elsevier.com/locate/foodchem

Analytical Methods

Quantitative analysis of acrylamide in tea by liquid chromatography coupled with electrospray ionization tandem mass spectrometry

Jie Liu [a], Ganghua Zhao [a], Yuan Yuan [b], Fang Chen [a,*], Xiaosong Hu [a]

[a] College of Food Science and Nutritional Engineering, China Agricultural University, Beijing 100083, China
[b] College of Quartermaster Technology, Jilin University, Changchun 130062, China

Received 14 August 2007; received in revised form 1 October 2007; accepted 8 November 2007

Abstract

An effective sample preparation procedure was optimized and a liquid chromatography–tandem mass spectrometry (LC–MS/MS) was developed for the quantitative analysis of acrylamide in tea. [$^{13}C_3$]-acrylamide was used as internal standard. Acrylamide was extracted at 25 °C for 20 min by 10 ml water followed by 10 ml acetonitrile, and then 4 g of magnesium sulfate and 0.5 g of sodium chloride were added to the above mixture under stirring thoroughly. In order to increase the response of acrylamide, 9 ml acetonitrile layer was taken and concentrated to 0.5 ml. Solid-phase extraction with an Oasis MCX cartridge was carried out for clean-up. The limit of detection (LOD) and limit of quantification (LOQ) were 1 and 5 ng/ml, respectively. The recovery efficiency of the extraction procedure ranged between 74% and 79%. The levels of acrylamide in 30 tea samples were less than 100 ng/g. Black, oolong, white and yellow tea samples had quite low acrylamide contents (<20 ng/g). Higher acrylamide levels occurred in baked, roasted, and one sun-dried green tea samples (46–94 ng/g).

Keywords: Acrylamide; Tea; Liquid chromatography–tandem mass spectrometry (LC–MS/MS); Quantitative analysis

1. Introduction

Acrylamide is a neurotoxic compound classified as a probable human carcinogen and genotoxicant (IARC, 1994). Since the high concentrations of acrylamide have been found in heated foodstuffs (Swedish National Food Administration, 2002; Tareke, Rydberg, Karlsson, Eriksson, & Tornqvist, 2002), much research on the analysis of acrylamide has been carried out by analytical chemists. Most of the methods have been developed to determine the acrylamide concentration in water, biological fluids, and non-cooked foods based on high performance liquid chromatography (HPLC) (Brown & Rhead, 1979) or gas chromatography (GC) (Bologna, Andrawes, Barvenik, Lentz, & Sojka, 1999; Castle, 1993; Pérez, Cheong, Yang, & Osterman-Golkar, 1999). However, the complexity and variability of food matrices still bring a challenge to the analysis of acrylamide.

The gas chromatography–mass spectrometry (GC–MS) method omitting derivatization step (Biedermann et al., 2002; Tateo & Bononi, 2003) appeared to be relatively simple, but its drawbacks were the lack of characteristic ions in the mass spectrum of underivatized acrylamide (Wenzl, Beatriz de la Calle, & Anklam, 2003) and the potential formation in the injection port if acrylamide precursors were present (Castle & Eriksson, 2005). The derivatization of acrylamide removed potentially interfering co-extractives and improved volatility, selectivity, and sensitivity in analysis (Taeymans et al., 2004). However, the bromination reaction was carried out from 1 h to overnight at the approximate freezing point of water (Castle, 1993; Nemoto, Takatsuki, Sasaki, & Maitani, 2002; Pittet, Périsset, & Oberson, 2004). The approaches based on GC–MS were laborious and time consuming.

* Corresponding author. Tel.: +86 10 62737645; fax: +86 10 62737434x11.
E-mail addresses: chenfangch@sina.com (F. Chen), huxiaos@263.net (X. Hu).

0308-8146/$ - see front matter © 2007 Published by Elsevier Ltd.
doi:10.1016/j.foodchem.2007.11.015

Nowadays, more attention was paid to employ liquid chromatography–tandem mass spectrometry (LC–MS/MS) techniques (Andrzejewski, Roach, Gay, & Musser, 2004; Govaert et al., 2006; Riediker & Stadler, 2003; Rosén & Hellenäs, 2002) having high sensitivity and saving time. HPLC with MS/MS detection working in multiple reaction-monitoring mode (MRM) had been demonstrated to be a powerful technique for the quantitative analysis of acrylamide (Andrzejewski et al., 2004; Govaert et al., 2006; Hoenicke, Gatermann, Harder, & Hartig, 2004; Zhang, Jiao, Ren, Wu, & Zhang, 2005). However, the presence of matrix effect becomes an important pitfall in MS/MS detection. Due to the low molecular weight of acrylamide, as well as its resulting low mass fragment ions, background interference in the characteristic acrylamide transitions may impede the analysis. Some studies discussed the matrix effect in the analysis of acrylamide (Aguas, Fitzhenry, Giannikopoulos, & Varelis, 2006; Mastovska & Lehotay, 2006; Petersson, Rosén, Turner, Danielsson, & Hellenäs, 2006). Co-eluting, undetected matrix components may reduce or enhance the ion intensity of the analytes and affect the reproducibility and accuracy of the assay (Matuszewski, Constanzer, & Chavez-Eng, 2003). Therefore, it is crucial to develop or explore different and specific methods for the preparation of samples due to complexity of the composition of different foodstuffs.

Tea is a kind of popular cosmopolitan beverage and about three million tons of teas were produced all over the world in 2006. China is famous for its tea culture and tea has been essential in Chinese life. The different categories of tea (black, oolong, green, white) are the result of variations in processing for the leaves after harvest. Especially, dehydration temperature plays an important role in formation of their attractive color, fragrance, and flavor. Unfortunately, different amounts of acrylamide in roasted green tea (247–1880 ng/g reported by Mizukami et al. (2006), 190–520 ng/g reported by Yoshida et al. (2005)), oolong and black tea (18–85 ng/g reported by Mizukami et al. (2006), 20–90 ng/g reported by Yoshida et al. (2005)) have been detected. Analysis methods of acrylamide in green tea have been reported by Nemoto et al. (2002) using GC–MS based on bromination and Takatsuki, Nemoto, Sasaki, and Maitani (2003) using LC–MS. This study developed a LC–MS/MS method to analyze acrylamide in tea by optimizing the preparation of sample, and the matrix effect on quantitative analysis was assessed to monitor the performance of the method. In addition, quantitative analysis was applied in detection of acrylamide in six classes of tea samples including green tea, oolong tea, black tea, white tea, yellow tea and Pu-erh tea in China.

2. Materials and methods

2.1. Samples

Fresh leaves of several cultivars of tea plants, such as 'Yingshuang', 'Fuding', 'Jiukeng', and 'Shubei' were picked in Hangzhou, Zhejiang province, in May 2006 and freeze-dried to the moisture content of 5.81% (wet basis), which were confirmed as blank sample. Six kinds of tea products, including green tea, oolong tea, black tea, white tea, yellow tea and Pu-erh tea were provided by Tea Institute of The Chinese Academy of Agricultural Sciences. All tea products were collected and processed from March to May 2006. All the samples were ground to a particle size of 1 mm or smaller, and kept at −18 °C until analysis.

2.2. Chemicals and materials

Acrylamide (2-propene amide) (purity >99.8%) and [$^{13}C_3$]-acrylamide (99% isotopic purity) were purchased from Sigma–Aldrich (St. Louis, MO, USA) and Cambridge Isotope Laboratories (Andover, MA, USA), respectively. Formic acid (⩾98%) and 2-propanol (HPLC-grade) was obtained from Sigma–Aldrich (Chemie GmbH, Germany), and methanol and acetonitrile (HPLC-grade) were from Honeywell Burdick and Jackson (SK Chemicals, Korea). Acetone (HPLC-grade) was purchased from Concord Tech. (Tianjin, China). HPLC-grade and 0.20 μm filtered water was prepared. Anhydrous magnesium sulfate and sodium chloride were of analytical grade and obtained from Beijing Chemicals Co. (Beijing, China).

The solid-phase extraction (SPE) cartridges Oasis MCX (3 ml, 60 mg) were supplied by Waters (Milford, MA, USA). Syringe filter units 13/0.22 and 13/0.45 MV were purchased from Hercules (Beijing, China).

2.3. Standard solutions

Stock solutions of 1 mg/ml for acrylamide and [$^{13}C_3$]-acrylamide were prepared with methanol. The stock solutions were diluted with water to give a series of standards. Linearity was established in both water and matrix. Seven concentration levels (1, 2, 5, 8, 10, 15, 20 ng/ml) were chosen and each level was performed in triplicate. The 90 ng/ml of [$^{13}C_3$]-acrylamide was used as internal standard in analysis.

2.4. Sample preparation

Acrylamide was analyzed according to the method described by Mastovska and Lehotay (2006) with the following modifications. A portion of the sample (1.0 g) was weighed into a 50 ml polypropylene centrifugal tube. Then, 1 ml of a 900 ng/ml [$^{13}C_3$]-acrylamide solution and 9 ml water were added into the tube. The samples were extracted in an incubated horizontal shaker at 25 °C for 20 min, then 10 ml acetonitrile, 4 g of anhydrous magnesium sulfate and 0.5 g of sodium chloride were added. The tube was sealed and shaken vigorously for 1 min immediately, then centrifuged at 5000 rpm for 5 min at 4 °C. The salt combination induced separation of water and acetonitrile layers and forced the majority of acrylamide into the acetonitrile layer. Three layers

were obtained as follows, an acetonitrile layer containing acrylamide at the top, the matrix layer in the middle and the water layer with the excessive salts in the bottom. The acetonitrile solution (9 ml) was transferred into a glass test tube and evaporated to dryness under a stream of nitrogen in a water bath at 40 °C. The residue on the wall of glass tube was re-dissolved in 0.5 ml of water under vortex, so some highly lipophilic co-extractives were excluded again. Aqueous extract was filtered through a 0.45 μm syringe filter for further clean-up.

Oasis MCX SPE cartridge was used for further clean-up. This kind of SPE sorbent is effective for extraction of basic compounds from complex matrices, while acrylamide exhibits weak basicity and does not interact strongly with the sorbent. The pass through strategy for the SPE clean-up was applied to retain the matrix interferences. SPE cartridge was conditioned consecutively with 2 ml of methanol and 2 ml water. Re-dissolved extract (0.5 ml) passed through the SPE cartridge and effluent was collected. Subsequently, a wash step was carried out to elute other acrylamide retained on the SPE column. Water, methanol, acetonitrile, acetone, and 2-propanol (0.5 ml of each solvent) were tested separately to find the suitable eluent. The final test solutions were filtered through a 0.22 μm syringe filter for quantification by LC–MS/MS.

2.5. LC–MS/MS analysis

The analysis of acrylamide in tea was performed by an Alliance 2695 Separations Module (Waters, Milford, MA, USA) coupled to a Micromass Quattro Micro triple-quadrupole mass spectrometer (Micromass, Manchester, UK) with MassLynx software. The final test solution (20 μl) after the preparation was injected onto a reversed ODS-C_{18} column (250 × 4.6 mm, 5 μm, Hypersil, Thermo, USA) maintained at 30 °C. The elution mode was isocratic using a mixture of 10% acetonitrile and 90% water containing 0.1% formic acid as mobile phase at a flow rate of 0.4 ml/min. Detection was performed by MS/MS after selection and optimization of relevant MRM traces from MS and daughter scans. Acrylamide was detected using electrospray ionization in the positive ion mode. The MRM mode of the degradation patterns m/z 72 → 55 for acrylamide and m/z 75 → 58 for [$^{13}C_3$]-acrylamide were used for quantification, respectively. The optimized MS instrument parameters obtained by the tuning were as follows: capillary voltage, 1 kV; cone voltage, 20 V; source temperature, 110 °C; desolvation temperature, 400 °C; desolvation gas flow, 600 l h^{-1} nitrogen; cone gas flow, 50 l h^{-1}; argon collision gas pressure to 2×10^{-3} mbar for MS/MS. The collision energy for each monitored transition was 13 eV in MRM mode. In the MRM transitions,

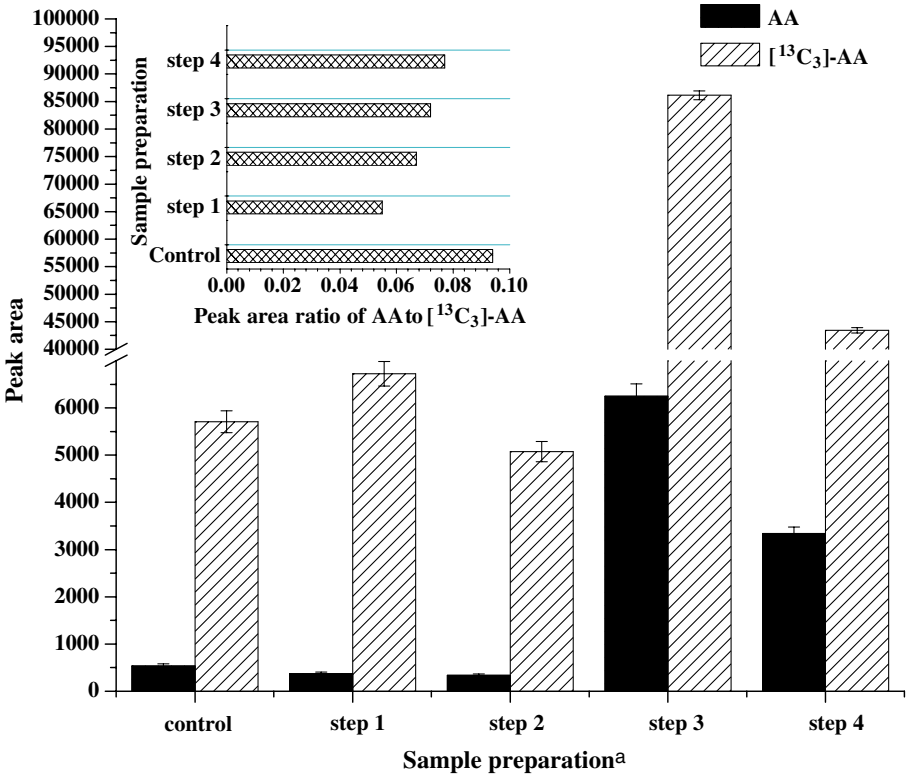

Fig. 1. Peak areas of acrylamide (AA) and [$^{13}C_3$]-acrylamide ([$^{13}C_3$]-AA) in critical steps of sample preparation. Inset: peak area ratio of AA to [$^{13}C_3$]-AA. Acrylamide (10 ng/ml) and [$^{13}C_3$]-acrylamide (90 ng/ml) were spiked into water as control. Acrylamide (10 ng/ml) and [$^{13}C_3$]-acrylamide (90 ng/ml) were spiked into blank sample before extraction and then were extracted through 4 critical steps. [a]Step 1, aqueous extraction; step 2, migration from aqueous extract to acetonitrile; step 3, enrichment from 9 ml acetonitrile extract to 0.5 ml aqueous solution; step 4, clean-up by SPE. Values are mean ± SD ($n = 3$).

the dwell and inter scan times were 0.4 and 0.1 s, respectively.

Acrylamide and [$^{13}C_3$]-acrylamide were spiked into the blank extracts *after* extraction. The concentrations of analytes were parallel to that in standard solutions. The matrix effect, recovery efficiency of the extraction procedure, and overall process efficiency of the quantitative LC–MS/MS method were assessed.

3. Results and discussion

The sample preparation is critical for analysis of acrylamide. In order to develop a suitable method to detect acrylamide at low levels in tea, some modifications are essential to increase response and decrease interference during LC–MS/MS analysis.

3.1. Optimization of sample preparation

The sample preparation was mainly based on the method described by Mastovska and Lehotay (2006). For the low levels of acrylamide in tea, it is necessary to find a way to improve its response for quantitative analysis. Therefore, enrichment and clean-up were important steps to increase the response and recovery in analysis. To assess sample preparation, the 10 ng/ml of acrylamide and 90 ng/ml of [$^{13}C_3$]-acrylamide were spiked into blank sample and extracted in the manner described as sample preparation. The same amounts of acrylamide and [$^{13}C_3$]-acrylamide were spiked into water as control. The extract obtained in some critical steps was separated for analysis.

Enrichment procedure consists of two stages, the migration of acrylamide from aqueous extract to acetonitrile (step 2) and re-dissolvation of residues dried from acetonitrile in 0.5 ml H_2O (step 3). In step 2, the responses of the analytes were declined to the least presumably because the significant loss of the analytes occurred in migration of analytes from water to acetonitrile. Unlike step 2, the responses of the analytes were greatly improved in step 3 due to the enrichment of analytes, namely, the responses of acrylamide and [$^{13}C_3$]-acrylamide improved 11.7 and 15.1 times to that of control (the enrichment factor should be 18), respectively (Fig. 1).

However, during the enrichment of analytes, the concentration of co-extractives also was enhanced. Consequently, interference of impurities in the response of analytes became more significant. Therefore, Oasis MCX SPE cartridge was used to remove the impurities left in extract of sample. Chromatograms (Fig. 2) indicated that 70% of analytes partitioned into the effluent when 0.5 ml of sample

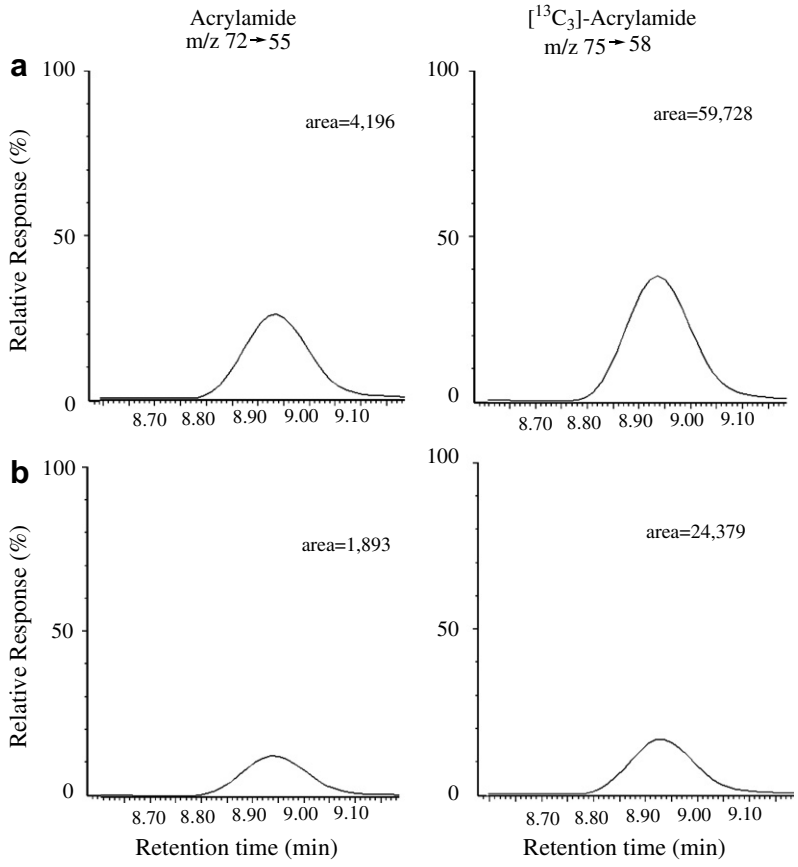

Fig. 2. Chromatograms of acrylamide and [$^{13}C_3$]-acrylamide upon SPE clean-up. (a) 0.5 ml of effluent obtained from load, (b) 0.5 ml of eluate obtained from water wash. Acrylamide (9 ng/ml) and [$^{13}C_3$]-acrylamide (90 ng/ml) were spiked into the blank sample. Values were mean (in triplicate).

extract loaded. The other analytes retained by the sorbents were eluted, using 0.5 ml of water, methanol, acetonitrile, acetone, and 2-propanol as eluent separately after loading. Results showed that water was the most apposite elution solvent as it gave the neat eluate and high response (Fig. 3). There was non-detects in eluate if further wash step was applied. Thus SPE clean-up caused no or minimum loss of analytes by collecting the effluent and eluate

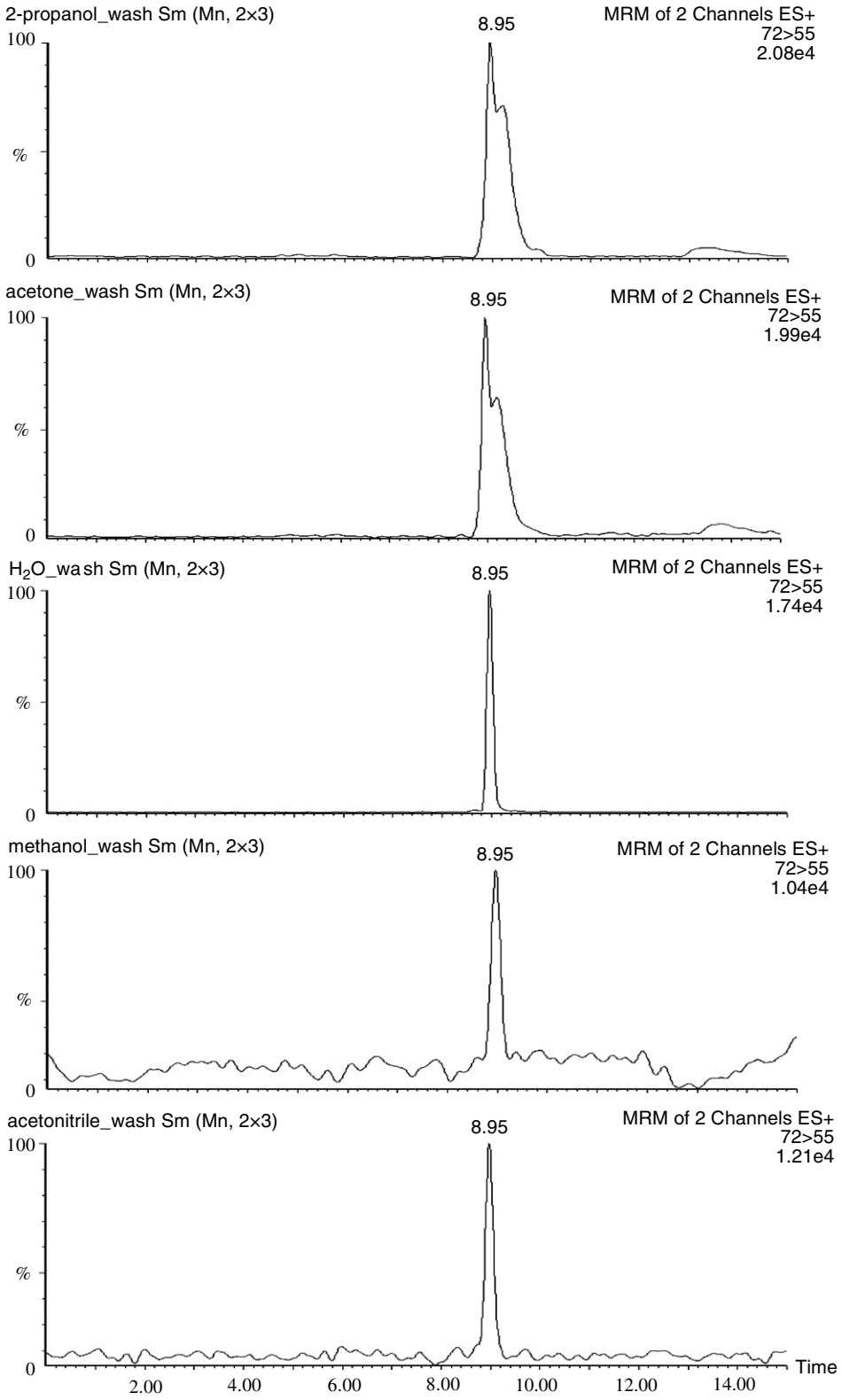

Fig. 3. Chromatograms of acrylamide in the eluates obtained from different eluents.

together. Based on the above SPE clean-up, the responses of acrylamide and [$^{13}C_3$]-acrylamide improved 6.4 and 7.6 times with respect to that of control, respectively (Fig. 1, step 4). The two increase factors of response were close to the enrichment 9 times, indicating that most impurities had been removed and the matrix effect was decreased.

In present study, [$^{13}C_3$]-acrylamide was added as internal standard for compensation of loss of analytes during sample preparation and improving the precision. Theoretically, a ratio of peak area of acrylamide to [$^{13}C_3$]-acrylamide would be constant upon preparation of each step for a certain sample given that matrix in the sample had no effect on analysis. In other words, the sample preparation would be ideal if the ratio of peak area of acrylamide to that of [$^{13}C_3$]-acrylamide obtained from the sample were identical to that in control. In practice, the ratios of different steps were still lower than that in control (inset of Fig. 1), resulting from interference of impurities in responses of analytes. The ratio in step 4 was the closest to that in control suggesting that there were the least effect of impurities on the analysis. Therefore, an effective clean-up had been achieved with the overall sample preparation.

The optimized sample preparation was applied in the analysis of acrylamide in tea products. The peak areas of the samples analyzed by non-enrichment and enrichment methods were compared. For enriched samples, both the peak areas of acrylamide and [$^{13}C_3$]-acrylamide were increased by 7.2–7.9 times and 7.3–8.1 times, respectively. Especially, acrylamide were not detected in Zhucha-2 and Zhucha-3 because of its much low concentration in the two samples when the non-enrichment step was used. In contrast, acrylamide in these samples can not only be detected but also quantified with the enrichment step (Table 1).

Acrylamide (1–20 ng/ml) and [$^{13}C_3$]-acrylamide (90 ng/ml) were separately spiked into water, blank tea extracts *after* extraction and blank samples *before* extraction to assess matrix effect, recovery efficiency of the extraction procedure, and overall process efficiency. They were evaluated by comparing the absolute peak areas for analytes (Table 2). Acrylamide and [$^{13}C_3$]-acrylamide had the average matrix effect of 92% and 102%, respectively. The average recovery efficiency of both the acrylamide and the [$^{13}C_3$]-acrylamide were 76%, and the average process efficiency containing matrix effect were 69% and 78%, respectively.

Table 1
Acrylamide content (ng/g) in tea products

Type of tea	Sample	Acrylamide (ng/g)
Green tea	Maofeng (Huangshan)	ND[a]
	Maojian (Xinyang)	46 ± 2[b]
	Longjing (Zhengjiang)	94 ± 1
	Biluochun (Jiangsu)	63 ± 1
	Zhucha-1 (Zhejiang)	ND
	Zhucha-2 (Zhejiang)	6 ± 1
	Zhucha-3 (Zhejiang)	3 ± 1
	Organic baked green tea (Zhejiang)	65 ± 1
	Yuya (Sichuan)	ND
	Longding (Zhejiang)	62 ± 2
Black tea	Fannings (Guangdong)	ND
	Congou (Guangdong)	9 ± 3
Oolong tea	Tieguanyin (Fujian)	ND
	Jasmine tea-1 (Fujian)	ND
	Jasmine tea-2 (Fujian)	18 ± 1
Raw Pu-erh	Sun-dried raw Pu-erh (Simao)	34 ± 1
	Raw cake (Lincang)	36 ± 1
	Raw cake (Simao)	24 ± 2
	Raw cake_Yinhao (Simao)	29 ± 1
	Raw brick (Yongde)	25 ± 1
	Sun-dried raw Pu-erh (third class)	20 ± 2
	Roasted raw Pu-erh (third class)	83 ± 2
	Sun-dried raw Pu-erh (first class)	ND
	Pu-erh Ziya (Lincang)	28 ± 1
	Canger nest (Xiaguan)	17 ± 1
Ripened Pu-erh	Royal cake (Xishuangbanna)	ND
	Qizi cake (Lincang)	ND
	Ripened Pu-erh (Simao)	50 ± 2
White tea	Yunmeng Baicha (Jiangsu)	ND
Yellow tea	Junshan silver needle (Hunan)	17 ± 2

[a] Not detected (below the detection limit of 1 ng/ml).
[b] Mean ± SD (standard deviation) ($n = 3$).

Table 2
Matrix effect (ME), recovery efficiency (RE), and process efficiency (PE) data for acrylamide (AA) and [$^{13}C_3$]-acrylamide (IS). Acrylamide (1–20 ng/ml) and [$^{13}C_3$]-acrylamide (90 ng/ml) were spiked into water, blank tea extracts *after* extraction and blank samples *before* extraction to assess these values

Nominal concentration (ng/ml)	ME (%)		RE (%)		PE (%)	
	AA	IS	AA	IS	AA	IS
1	88	108	75	76	66	82
2	92	99	75	78	69	77
5	95	102	77	76	73	78
8	90	96	79	75	71	72
10	93	105	74	77	69	81
15	93	105	74	76	69	80
20	92	101	75	77	69	78
Mean ± SD[a]	92 ± 2	102 ± 4	76 ± 2	76 ± 1	69 ± 2	78 ± 3

[a] Standard deviation ($n = 7$).

3.2. Method performance

To check performance of the developed analysis method of acrylamide, parameters such as limit of detection (LOD), limit of quantification (LOQ), linearity, repeatability or run-to-run precision were evaluated.

For the evaluation of instrumental performance in terms of LOD and LOQ, assays were performed in water. LOD and LOQ matrix dependent were calculated in blank tea sample. LOD and LOQ were determined as the amounts of acrylamide that produced a signal-to-noise ratio of 3:1 and 10:1, respectively. The values of LOD and LOQ were 0.8 and 4 ng/ml in water, respectively. In tea matrix, LOD and LOQ were higher (1 and 5 ng/ml) than those in

water due to chemical noise and matrix effect. The LOD and LOQ of other method adapted to analyze acrylamide in tea by LC/MS using column switch were reported as 9 and 30 ng/g, respectively (Takatsuki et al., 2003). It is clear that the LOD and LOQ of the method developed in this paper were reduced greatly. The linear range extended from 1 to 20 ng/ml and good coefficients of determination ($r > 0.99$) were achieved. Precision was calculated in terms of intra-day repeatability and inter-day reproducibility as RSD% at two concentration levels. The tested products were Ziya (a raw Pu-erh tea) and Biluochun (a green tea). The acrylamide concentrations ranged from 28 to 63 ng/g. The values of RSD ranged from 1.6% to 3.6% for the intra-day precision test ($n = 5$) and 2.9% to 8.3% for the inter-day precision tests ($n = 15$).

3.3. Acrylamide levels in teas

A total of 30 tea samples of six classes were analyzed for the acrylamide concentration (Table 1). The acrylamide levels of all samples were less than 100 ng/g. In most raw Pu-erh tea, the acrylamide levels ranged in 17–36 ng/g except that absence of acrylamide was in one and relatively high level (83 ng/g) in the other. Samples obtained from four classes of tea products including black tea, oolong tea, white tea and yellow tea had a quite low acrylamide concentration (<20 ng/g) probably because the fresh leaves were not dried at high temperature. High acrylamide levels occurred in baked, roasted, and one sun-dried green tea samples (46–94 ng/g), probably because 100–150 °C were usually used during process. The acrylamide levels of tested tea products in this study were much lower than the Japanese tea, especially in roasted green tea such as Houjicha (Mizukami et al. 2006). This might ascribe to the higher processed temperature (170–200 °C) used in Japanese roasted green tea than that in Chinese tea.

4. Conclusion

This work developed a quantitative method for the trace determination of acrylamide in tea. The critical steps including enrichment and SPE clean-up were optimized in sample preparation. This method was effective to decrease interference from sample and increase the response of acrylamide. Based on the above method, acrylamide in 30 Chinese tea products were analyzed, so the relatively low levels of acrylamide were found in these samples.

Acknowledgements

This work was carried out with support from the Yunnan Science and Technology Project, Specialized Research Fund for the Doctoral Program of Higher Education from the Ministry of Education of China (Grant No. 20050019002), and National Key Technology R&D Program (Grant No. 2006BAD05A06). Accordingly we gratefully acknowledge the fund supports.

References

Aguas, P. C., Fitzhenry, M. J., Giannikopoulos, G., & Varelis, P. (2006). Analysis of acrylamide in coffee and cocoa by isotope dilution liquid chromatography–tandem mass spectrometry. *Analytical and Bioanalytical Chemistry, 385*, 1526–1531.

Andrzejewski, D., Roach, J. A. G., Gay, M. L., & Musser, S. M. (2004). Analysis of coffee for the presence of acrylamide by LC–MS/MS. *Journal of Agriculture and Food Chemistry, 52*, 1996–2002.

Biedermann, M., Biedermann-Brem, S., Noti, A., Grob, K., Egli, P., & Mändli, H. (2002). Two GC–MS methods for the analysis of acrylamide in foods. *Mitteilungen aus Lebensmitteluntersuchung und Hygiene, 93*, 638–652.

Bologna, L. S., Andrawes, F. E., Barvenik, F. W., Lentz, R. D., & Sojka, R. E. J. (1999). Analysis of residual acrylamide in field crops. *Journal of Chromatographic Science, 37*, 240–244.

Brown, L., & Rhead, M. (1979). Liquid chromatographic determination of acrylamide monomer in natural and polluted aqueous environments. *Analyst, 104*, 391–399.

Castle, L. (1993). Determination of acrylamide monomer in mushrooms grown on polyacrylamide gel. *Journal of Agriculture and Food Chemistry, 41*, 1261–1263.

Castle, L., & Eriksson, S. (2005). Analytical methods used to measure acrylamide concentrations in foods. *Journal of AOAC International, 88*, 274–284.

Govaert, Y., Arisseto, A., Van Loco, J., Scheers, E., Fraselle, S., Weverbergh, E., et al. (2006). Optimisation of a liquid chromatography–tandem mass spectrometric method for the determination of acrylamide in foods. *Analytica Chimica Acta, 556*, 275–280.

Hoenicke, K., Gatermann, R., Harder, W., & Hartig, L. (2004). Analysis of acrylamide in different foodstuffs using liquid chromatography–tandem mass spectrometry and gas chromatography–tandem mass spectrometry. *Analytica Chimica Acta, 520*, 207–215.

IARC (1994). International Agency for Research on Cancer. Lyon, France.

Mastovska, K., & Lehotay, S. J. (2006). Rapid sample preparation method for LC–MS/MS or GC–MS analysis of acrylamide in various food matrices. *Journal of Agriculture and Food Chemistry, 54*, 7001–7008.

Matuszewski, B. K., Constanzer, M. L., & Chavez-Eng, C. M. (2003). Strategies for the assessment of matrix effect in quantitative bioanalytical methods based on HPLC–MS/MS. *Analytical Chemistry, 75*, 3019–3030.

Mizukami, Y., Kohata, K., Yamaguchi, Y., Hayashi, N., Sawai, Y., Chuda, Y., et al. (2006). Analysis of acrylamide in green tea by gas chromatography–mass spectrometry. *Journal of Agriculture and Food Chemistry, 54*, 7370–7377.

Nemoto, S., Takatsuki, S., Sasaki, K., & Maitani, T. (2002). Determination of acrylamide in foods by GC/MS using ^{13}C-labeled acrylamide as an internal standard. *Journal of the Food Hygienic Society of Japan, 43*, 371–376.

Pérez, H. L., Cheong, H.-K., Yang, J. S., & Osterman-Golkar, S. (1999). Simultaneous analysis of hemoglobin adducts of acrylamide and glycidamide by gas chromatography–mass spectrometry. *Analytical Biochemistry, 274*, 59–68.

Petersson, E. V., Rosén, J., Turner, C., Danielsson, R., & Hellenäs, K. E. (2006). Critical factors and pitfalls affecting the extraction of acrylamide from foods: An optimization study. *Analytica Chimica Acta, 557*, 287–295.

Pittet, A., Périsset, A., & Oberson, J.-M. (2004). Trace level determination of acrylamide in cereal-based foods by gas chromatography–mass spectrometry. *Journal of Chromatography A, 1035*, 123–130.

Riediker, S., & Stadler, R. H. (2003). Analysis of acrylamide in food by isotope-dilution liquid chromatography coupled with electrospray ionization tandem mass spectrometry. *Journal of Chromatography A, 1020*, 121–130.

Rosén, J., & Hellenäs, K. E. (2002). Analysis of acrylamide in cooked foods by liquid chromatography tandem mass spectrometery. *Analyst, 127*, 880–882.

Swedish National Food Administration (2002). Information about acrylamide in food, 24 April. ⟨http://www.slv.se⟩.

Taeymans, D., Wood, J., Ashby, P., Blank, I., Studer, A., Stadler, R. H., et al. (2004). A review of acrylamide: An industry perspective on research, analysis, formation, and control. *Critical Reviews in Food Science and Nutrition, 44*, 323–347.

Takatsuki, S., Nemoto, S., Sasaki, K., & Maitani, T. (2003). Determination of acrylamide in processed foods by LC–MS using column switching. *Journal of the Food Hygienic Society of Japan, 44*, 89–95.

Tareke, E., Rydberg, P., Karlsson, P., Eriksson, S., & Tornqvist, M. (2002). Analysis of acrylamide, a carcinogen formed in heated foodstuffs. *Journal of Agriculture and Food Chemistry, 50*, 4998–5006.

Tateo, E., & Bononi, M. (2003). A GC/MS method for the routine determination of acrylamide in food. *Italian Journal of Food Science, 15*, 149–151.

Wenzl, T., Beatriz de la Calle, M., & Anklam, E. (2003). Analytical methods for the determination of acrylamide in food products: A review. *Food Additives and Contaminants, 20*, 885–902.

Yoshida, M., Ono, H., Chuda, Y., Yada, H., Onhishi-Kameyama, M., Kobayashi, H., et al. (2005). Acrylamide in Japanese processed foods and factors affecting acrylamide level in potato chips and tea. *Advances in Experimental Medicine and Biology, 561*, 405–413.

Zhang, Y., Jiao, J., Ren, Y. P., Wu, X. Q., & Zhang, Y. (2005). Determination of acrylamide in infant cereal-based foods by isotope dilution liquid chromatography coupled with electrospray ionization tandem mass spectrometry. *Analytica Chimica Acta, 551*, 150–158.