Food Chemistry 197 (2016) 1200–1206

Contents lists available at ScienceDirect

Food Chemistry

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

Analytical Methods

Determination of phthalate esters in teas and tea infusions by gas chromatography–mass spectrometry

Liping Du *, Lijuan Ma, Yang Qiao, Yan Lu, Dongguang Xiao

Key Laboratory of Industrial Fermentation Microbiology Ministry of Education, Tianjin Industrial Microbiology Key Laboratory, College of Biotechnology, Tianjin University of Science and Technology, Tianjin 300457, People's Republic of China

ARTICLE INFO

Article history:
Received 6 July 2015
Received in revised form 12 November 2015
Accepted 14 November 2015
Available online 19 November 2015

Keywords:
Tea
Tea infusion
Phthalate esters
Simultaneous distillation extraction
Gas chromatography–mass spectrometry

ABSTRACT

Phthalate esters (PAEs), a group of environmental pollutants which are carcinogenic to human body, have been detected in teas. In this work, five PAEs in teas and tea infusions were quantitatively determined by a modified simultaneous distillation extraction (SDE) coupled with gas chromatography–mass spectrometry. After the optimization of SDE, the proposed method afforded a wide range of linearity and high linear regression coefficients with the limits of detection range of 0.24–3.72 μg/kg. The average recoveries were 79.83–116.67% for tea samples and 78.22–101.64% for tea infusions with all the relative standard deviations below 20%. The total content of five PAEs in teas was 1.135–3.734 mg/kg and the total dissolving ratio of five PAEs from tea to infusion was 19.05–28.07% for the selected tea samples. The risk assessment result of all the selected tea samples demonstrated that the population with the habit of drinking tea won't cause risk to human health.

1. Introduction

Phthalates esters (PAEs) are a group of important environmental pollutants, including dimethyl phthalate (DMP), diethyl phthalate (DEP), diisobutyl phthalate (DIBP), dibutyl phthalate (DBP), diethylexyl phthalate (DEHP), dioctyl phthalate (DOP) and so on. They were widely used as plasticizers and additives in many daily products such as plastics, pesticides, paints and cosmetics (Carlo et al., 2008; Shen, 2005). PAEs are not covalently bound to the polymer molecules by chemical, they can easily release and migrate from polymeric materials into the environment (Zhang et al., 2014) and food products (Wang et al., 2015). Studies have shown that PAEs had carcinogenic and estrogenic impact on human health (Luo, Yu, Yuan, & Feng, 2012; Matsumoto, Hirata-Koizumi, & Ema, 2008). So the US Environmental Protection Agency (EPA) and several other countries had established a maximum admissible concentration for specific PAEs (Gomez-Hens, & Aguilar-Caballos, 2003). For example, a definite acceptable daily intake (ADI), 0.01 mg/(kg·bw·d) for DBP and 0.05 mg/(kg·bw·d) for DEHP was set in the regulations which were made by US Food and Drug Administration (FDA) and European Union (EU).

Tea, as one of the most widely consumed functional beverages worldwide, has received more and more attentions for its health beneficial properties (Yang, Chung, Yang, Chhabra, & Lee, 2000). However, various studies have proved that tea leaves may contain contaminants which can be released into infusions and may be harmful to human health. PAEs have been detected in the aroma analysis of tea samples in recent studies (Joshi, Poonam & Gulati, 2011; Rawat et al., 2007; Sereshti, Samadi, & Jalali-Heravi, 2013; Yang, Hauser, & Goldman, 2013). Moreover, tea is consumed in the form of infusion by the consumers, so it is essential to determine the dissolving ratio of PAEs from dried tea to infusion.

Among the methods used for PAEs determination, such as gas chromatography (Li, Zeng, Chen, & Xu, 2004), high performance liquid chromatography (Fan, Liu, & Xie, 2014), gas chromatography–mass spectrometry (GC–MS) (Carlo et al., 2008; Farahani, Norouzi, Dinarvand, & Ganjali, 2007; Orecchio, Indelicato, & Barreca, 2014; Ye, Liu, Chen, & Hong, 2014) and liquid chromatography-mass spectrometry (Lu, Hashi, Wang, Ma, & Lin, 2011), GC–MS was the most commonly used. Before the GC–MS analysis, dispersive liquid–liquid micro extraction (Farahani et al., 2007), headspace solid-phase micro-extraction (HS-SPME) (Mous, Basheer, & Al-Arfaj, 2013; Rios, Morales, Márquez-Ruiz, 2010) and solid phase extraction (Osman, Özer, Beşirli, & Güçer, 2013; Zhao, Wang, Yuan, & Lin, 2008) are used most commonly for a needed prior extraction of PAEs.

As the boiling points of PAEs are exceed 280 °C and their contents in tea samples usually are trace level, SDE which has a good extraction capacity for compositions with high boiling point from

* Corresponding author.
E-mail address: dlp123@tust.edu.cn (L. Du).

http://dx.doi.org/10.1016/j.foodchem.2015.11.082
0308-8146/

the complex matrix is suitable for the extraction of PAEs. For example, Rawat et al. (2007) found that SDE was more efficient than other distillation processes for extracting volatile components in the analysis of volatile components in kangra orthodox black tea by GC–MS (Rawat et al., 2007). So far, there is no report about the utilization of SDE in the special extraction of PAEs from tea samples and their accurate quantification in tea samples. Recently, Yin, Liu, Chen, Pan, and Ma (2014) established a quantitative method for the determination of 16 PAEs in teas and QuEChERS method was used to extract the PAEs, and only five PAEs (DMP, DEP, DIBP, DBP and DEHP) were all detected in 105 tea samples (Yin et al., 2014). However, only the PAEs in tea samples were determined in their work, and tea is consumed in the form of infusion by the consumers, so it is essential to determine the dissolving ratio of PAEs from dried tea to infusion. Additionally, only these five kinds of PAEs (DMP, DEP, DIBP, DBP and DEHP) were detected in the volatile compounds of different teas in most of the previous studies. Therefore, it is of great significance to develop a proper extraction and quantitative analysis method for these five kinds of PAEs in tea samples and tea infusions.

In this work, we aimed to develop a method for the direct determination of the five PAEs (DMP、DEP、DIBP、DBP、DEHP) in teas and their infusions. SDE was employed to extract PAEs from different tea samples and tea infusions and the extraction conditions were optimized. GC–MS was used to determine the content of the five PAEs in different tea samples and their infusions. This study also assessed the transfer behavior of five PAEs from tea samples to their infusions. Moreover, we made a preliminary judgment of safety of drinking tea beverage through the risk assessment.

2. Materials and methods

2.1. Chemicals and reagents

Acetone, n-pentane, n-hexane, dichloromethane and ether were of HPLC grade and purchased from Sinopharm Chemical Reagent Co., Ltd (Tianjin, China). Dimethyl phthalate (DMP, ⩾99.7%), diethyl phthalate (DEP, ⩾99.5%) and dibutyl phthalate (DBP, ⩾99.5%) were purchased from Aladdin (shanghai, China), diisobutyl phthalate (DIBP, ⩾98%), diethylexyl phthalate (DEHP, ⩾98%) were purchased from TCI (shanghai, China). Anhydrous sodium sulfate was of analytical grade, purchased from BaiShi Chemical Industry Co., Ltd (Tianjin, China). Ultrapure water was obtained from a Milli-Q purification system (Millipore, Bedford, USA). To avoid PAEs contamination, all reagents should be used to detect the presence of PAEs before use.

2.2. Samples

A total of 25 tea samples including 5 green teas (No. 1–5), 5 oolong teas (No. 6–10), 10 black teas (No. 11–20) and 5 dark teas (No. 21–25) were used for PAEs determination. Among them, 21 tea samples (No. 1–16, No. 21–25) were purchased from China, and the other 4 tea samples (No. 17–20) were obtained from UK. No. 1 green tea sample was chosen for the following optimization and method validation process.

2.3. Samples and standard preparation

All of the tea samples were grinded to pass through 30–60 mesh and sealed for future use. The stock solution was prepared by dissolving the five standard compounds with n-hexane, which contained 11.90 g/L DMP, 11.10 g/L DEP, 10.20 g/L DIBP, 10.40 g/L DBP and 9.70 g/L DEHP. Standard solutions of these five PAEs with different concentration were prepared by diluting the stock solution with n-hexane. All the standard solutions were stored at 4 °C before use.

2.4. Simultaneous distillation extraction

10 g of the grinded tea sample was placed in a flask, and then 150 mL of ultrapure water was added into the flask. 30 mL of n-hexane was applied as extraction solvent in another flask. Both flasks were installed in a Likens-Nickerson apparatus (Anhui, China) and heated up to their boiling points. Then the extraction was refluxed for 3 h to collect the analytes. After the extraction, the extraction solvent containing PAEs was collected and dried over anhydrous sodium sulfate overnight. The extracts were then concentrated approximately to 0.4 mL with termovap sample concentrator (Orgnomation, USA) at 50 °C and volumed to 1 mL with n-hexane. The concentrate was stored at −20 °C before analysis.

2.5. Tea infusion preparation

The tea samples were subjected to infusion. According to the tea drinking habit, all the tea samples were infused three times.

10 g tea sample was immersed in 500 mL ultrapure water at 100 °C. After 5 min of infusion, the solution was filtered through a stainless steel filter. And the second infusion was prepared by adding 500 mL hot ultrapure water to the first tea residue for 5 min and filtered. Then the third tea infusion was prepared as before, all the infusions were cooled and examined for PAEs transfer from the tea samples to infusions.

2.6. GC–MS analysis

All analyses were performed on Agilent 7890A gas chromatograph coupled with an Agilent 5975C mass selective detector (MSD) (Agilent, Santa Clara, CA), equipped with HP-5MS capillary column (60 m × 0.32 mm inner diameter, 0.25 μm film thickness). The operating conditions of GC were: Helium (percentage purity > 99.999 %) was used as carrier gas at a flow rate of 1 mL/min, 1 μL concentrate was injected in splitless mode, and injector temperature was 300 °C. Oven temperature was programmed as follows: started from 60 °C, increase to 120 °C at the rate of 5 °C/min (hold for 2 min), then increase to 180 °C at the rate of 2 °C/min (hold for 5 min), and finally rose to 250 °C at the rate of 6 °C/min (hold for 12 min). The total GC runtime was 72.667 min. The mass spectrometer was operated in an electron-impact mode of 70 eV. The transmission line temperature was 280 °C; ion source temperature was 230 °C; quadrupole temperature was 150 °C. Based on the mass scan range of 35–500 atomic mass units (amu) with SCAN mode, retention times of PAEs were determined by comparing the MS fragmentation pattern of the standards and the National Institute of Standards and Technology (NIST) 08 GC–MS library.

2.7. Calculation of recovery

Known amounts of the standard solutions (1 mL) at three levels were added to the tea sample and infusion to produce the spiked samples. The recovery rate of each analyte in the tea sample and infusion were calculated as follows:

$$\text{Recovery rate} = (M_1 - M_2)/M_0 \times 100\%$$

where M_1 and M_2 are the concentrations of PAEs detected in the spiked sample and the tea samples (μg), and M_0 is the concentration of PAEs in the standard solutions added into the tea samples (μg), respectively.