Phthalates leaching from plastic food and pharmaceutical contact materials by FTIR and GC-MS

Tatjana Andjelković[1] · Danica Bogdanović[1] · Ivana Kostić[1] · Gordana Kocić[2] · Goran Nikolić[3] · Radmila Pavlović[4]

Abstract

Phthalates are often used as plasticizers in the production of plastic food contact materials (FCMs) and pharmaceutical contact materials (PCMs), and having in mind that they are not bound to plastics, phthalates may easily leach from plastics under certain conditions. The aim of this research is determination of phthalates leaching potential from different plastic materials and quantitative determination of 5 phthalates (dimethyl phthalate (DMP), di-n-butyl phthalate (DnBP), benzyl butyl phthalate (BBP), diethyl hexyl phthalate (DEHP), and di-n-octyl phthalate (DOP)) in 44 different plastic articles of 7 different plastic polymers used as FCMs and PCMs by FTIR, GC-MS, and gravimetric methods. The FTIR technique is shown to be rapid method for determination of phthalate content in PVC articles. Comparing of FTIR method with GC-MS and gravimetric showed that separation and quantitative determination of each phthalate separately favor the GC-MS method, because FTIR method determines the total amount of phthalate content. However, the FTIR method is less expensive and demanding in terms of sample preparation, which is suited for use in pre-screening analysis. The results of GC-MS phthalates determination showed that PVC articles used as PCMs contain DEHP in significant amount, from 5.19 to 28.76% by weight and could be a potential risk to human health.

Keywords Phthalates determination · Food contact materials · Pharmaceutical contact materials · Leaching potential · GC-MS · FTIR

Introduction

Plastic is one of the most common used materials for packaging of food and pharmaceuticals due to its excellent properties like light weight and flexibility. Although packaging is the major application filed of plastic, providing preservation of the packed food and pharmaceuticals, it is also used during production, processing, preparation of food, and pharmaceuticals, and thus, plastic is considered as common food contact material (FCM) and pharmaceutical contact material (PCM).

Plastics are made up of polymers (70–99%), macromolecules composed of a large number of repeated units, and additives which provide functionality to plastics. Additives are added to the polymers to improve the properties of the plastics, to increase elasticity, softness, flexibility, and thus reduce their fragility. Phthalates are well known and the most commonly used plasticizers.

Phthalates used as plastic additives are diesters of 1,2-benzenedicarboxylic acid (phthalic acid), whose properties depend on an aliphatic or aromatic chain that substitutes carboxylic groups of phthalic acid. The most used phthalates are dimethyl phthalate (DMP), di-n-butyl phthalate (DnBP), benzyl butyl phthalate (BBP), diethyl hexyl phthalate (DEHP), di-n-octyl phthalate (DOP), diisononyl phthalate (DINP), and diisodecyl phthalate (DIDP). Annual production of all phthalates amounted to 6 million tons and that DEHP is the most frequently used plasticizer, with its production of up to

⚫ Springer

40% of the total phthalates (Rudel and Perovich 2009). The literature survey has shown that totally dominating as plasticizer is DEHP in medical devices and pharmaceutical contact materials. In 2008, DEHP was categorized as CMR 1B (carcinogenic, mutagenic, or toxic to reproduction) under the CLP Regulations (European Union Regulation 2008). This resulted in restriction of its use in plasticized PVC medical tubing in neonatology and maternity departments from July 1, 2015 (French law 2012; French law 2015). However, even there has been significant interest in non-DEHP alternatives for PVC medical devices, this investigation shows that the usage of DEHP as plasticizer in medical devices is still significant.

Phthalates as additives are usually added to polyvinyl chloride (PVC), since PVC is completely hard unless a plasticizer is added. But besides their occurrence as additives in PVC, phthalates are used during polymerization process for polypropylene (PP), polyethylene (PE), and polystyrene (PS) polymers as part of catalyst (Ziegler-Natta catalyst) (Kissin et al. 2008). Thus, phthalates have been found into PVC and non-PVC food and pharmaceutical contact materials (Bacha et al. 2012; Kostic et al. 2016).

Phthalates are colorless, viscous, lipophilic liquid, poorly soluble in water, and dissolvable in non-polar solvents. The physicochemical properties of 5 commercially significant phthalates that are the target analytes in this research are given in Table 1.

As seen in the Table 1, solubility in water is relatively low and decreases with increasing length of the alcohol chain or with higher molecular weight (MW) of phthalate. Usage of the various phthalates mainly depend on their MW; thus, low MW phthalates such as DMP, DnBP, and BBP are used for their emulsifying properties, as solvents in personal care products, insecticides, lacquers, and in coatings including those used to provide timed releases in some pharmaceuticals, while higher MW phthalates like DEHP, DnOP, DINP, and DIDP are principally incorporated in polymers (especially PVC) as plasticizers and are used in food packaging, medical devices, children's products, construction materials, and numerous PVC products including clothing, flooring, and wall coverings. Phthalate esters are not chemically bounded (covalent bonds) to the polymer and are therefore able to migrate to the surface of the polymer matrix. Here, they may be lost by

a variety of physical processes and can migrate from plastic into food or pharmaceuticals, especially by heat or solvents. This led to their ubiquitous spread in the environment. Their high hydrophobicity (high value for octanol-water partition coefficient-LogKow) makes them highly persistent and bio accumulating (Table 1).

Various studies have shown that phthalates impact human health because phthalate exposure may induce reproductive toxicity, liver damage, and carcinogenesis in humans (Rowdhwal and Chen 2018). Phthalates that show these effects on human health include DnBP, DEHP, BBP, DIDP, and DINP. Since phthalates represent endocrine disruptors, it is particularly important to do constant screening: to monitor their presence in various food and pharmaceutical staff and to monitor their leaching from the packaging to the surrounding environment (Benjamin et al. 2017). For this reason, European Food Safety Authority (EFSA) had set the tolerable daily intakes (TDI) for these phthalates: 0.01, 0.05, and 0.5 mg/kg body weight per day for DnBP, DEHP, and BBP, respectively, and 0.15 mg/kg body weight per day for DINP and DIDP (Silano et al. 2019). Due to occurrence of metabolites of DnBP, DEHP, and BBP in significant quantities in human blood and urine (Anderson et al. 2001), those three phthalates are investigated in this research together with DMP and DnOP. Although EFSA has not prohibited the use of DMP and DnOP, those two phthalates are included in the research being good representatives of low-MW and high-MW phthalates providing trend in leaching potential related to their MW, structure, polarity, and solubility.

The identification and quantification of phthalates require an analytical technique selective enough to distinguish each phthalate from other additives and from the plastic matrix. Depending on the matrix being tested and the physicochemical properties of the investigated phthalate, different techniques are applied. The most used methods for quantification are gas chromatography (GC) (Qian et al. 2018; Milojkovic et al. 2015) and high performance liquid chromatography (HPLC) (Dural 2020), and for sample preparation, liquid-liquid extraction (LLE) (Hadjmohammadi and Ranjbari 2012), solid-phase extraction (SPE) (Xin Gao et al. 2014), solid-phase micro extraction (SPME) (Amanzadeh et al. 2016), microwave extraction (Liang et al. 2010), Soxhlet

Table 1 The physicochemical properties of commercially significant phthalates

Phthalate	Chemical formula	Molar mass (g/mol)	Density (g/cm³)	Solubility in water at 20 °C (mg/L)	LogK$_{ow}$
DMP	$C_{10}H_{10}O_4$	194.18	1.19	5220	1.61
DnBP	$C_{16}H_{22}O_4$	278.35	1.05	9.9	4.27
BBP	$C_{19}H_{20}O_4$	312.37	1.10	3.8	4.70
DEHP	$C_{24}H_{38}O_4$	390.56	0.99	0.027	7.60
DnOP	$C_{24}H_{38}O_4$	390.56	0.98	0.022	8.10

extraction (Visser 2009), ultrasonic extraction (Zhou et al. 2012; Bogdanovic et al. 2019), and dispersive liquid-liquid micro extraction (DLLME) (Amin et al. 2018).

Besides those techniques, the Fourier-transform infrared spectroscopy (FTIR) is a technique that has many advantages in comparison with traditional analytical methods. This technique is reliable, rapid, non-destructive, and does not require special preparation of the samples; reagents are not typically required, far less costly than the GC-MS method, and therefore, it finds significance in the pre-screening for phthalates in plastic materials. In most cases, the FTIR analysis time is shorter than GC-MS analysis time, even when sample preparation time is included, allowing high sample throughput and quick feedback of analytical information.

In this study, the analysis of phthalate content and their leaching potential from 44 different plastic articles of 7 different plastic polymers used as FCMs and PCMs was performed. Considering the permitted and prohibited phthalates by EFSA and the expected high leaching potential the following 3 phthalates with low MW: DMP, DnBP, and BBP, and 2 phthalates with high MW: DEHP and DnOP have been determined in this study.

The analysis of phthalate content was performed by the FTIR technique as a rapid method for determination of phthalate content in PVC articles. The method was developed and compared with GC-MS and gravimetric determination of phthalates. For obtaining FTIR spectra, the samples were recorded by applying two techniques: polymer disk and KBr pellet. This rapid screening method for phthalates is simple and can be used as supportive of the regulated GC-MS method, because GC-MS screening is time-consuming and expensive.

Leaching potentials of phthalates from plastics were determined in contact experiments under "worst-case scenario" using *n*-hexane as recipient solvent with conditions such as ultrasound at room temp, prolonged contact time, and mixing, in order to have higher safety factor in health exposure calculations. The separation and quantification of leached phthalates from 44 different plastic articles was performed by GC-MS.

Experimental part

Reagents and materials

The tetrahydrofuran (THF) and methanol (HPLC grade) were purchased from Fischer scientific (USA). The *n*-hexane (HPLC grade) was purchased from Carlo Erba (France). DMP, DnBP, BBP, DEHP, and DnOP were purchased, in the highest available purity, from Sigma–Aldrich (USA). Dibutyl adipate (DBA) was purchased from Fluka (Switzerland) and used as an internal standard.

Apparatus and equipment

The Laser Fourier Transform Infrared Spectroscope (BOMEM Hartman and Braun—Michelson MB series 100) was used to record FTIR spectra. The manual hydraulic press (Graseby Specac), designed fundamentally to create pellet samples which can then be processed using analytical equipment such as FTIR and provide high optical throughput for precise and efficient analysis, was used. Amalgamator (Wig-L-Bug) was used for mixing a variety of materials including PVC precipitates from this analysis.

Gas chromatographic analysis was performed by Gas Chromatograph 6890 (Hewlett-Packard, USA) equipped with a mass selective detector (MSD) 5973 (Hewlett-Packard, USA), Auto sampler 7683 (Agilent, USA), and SGE 25QC2/BPX5 0.25 capillary column (25 m × 0.22 mm × 0.25 μm, non-polar).

The centrifuge Jouan C4I Benchtop (Termo Fisher) was used for separation and the analytical balance (Kern, CA) with accuracy of ±0.00001 g for gravimetric measurements.

Plastic materials

In this investigation, a total of 44 different plastic articles made of 7 different plastic polymers from 24 different manufacturers used as FCMs and PCMs were analyzed. The description of tested plastic articles is given in Table 2.

FCMs include products made by polyethylene terephthalate (PET), high-density polyethylene (HDPE), low-density polyethylene (LDPE), polypropylene (PP), cellophane, polystyrene (PS), and polycarbonate (PC), while PCM include articles made from polyvinyl chloride (PVC) and LDPE.

HDPE and LDPE granulate were purchased at a local factory to produce plastic packaging (Niš, Serbia). All other commercial plastic articles were purchased at local stores in Niš (Serbia). PCM for this analysis was taken from local Clinical Center Niš (Serbia).

Calibration standards

All stock, intermediate, and working solutions for GC-MS analysis were prepared in *n*-hexane. The stock solutions of DMP, DnBP, BBP, DEHP, DnOP, and dibutyl adipate (DBA) were prepared at a concentration of 1000 μg mL^{-1} of each. The stock standard was diluted stepwise with *n*-hexane to prepare calibration standard series in a concentration range 0.25–2.50 μg mL^{-1} of each phthalate with DBA at a concentration of 1 μg mL^{-1}.

Preparation of the calibration standards for FTIR analysis was done by spiking the PVC sample from which the total phthalate content was leached by the dissolution-precipitation method. Non-phthalate plastic precipitate (0.01000 g) was dissolved and spiked with DEHP so each spiked precipitate contained

Table 2 Description of investigated 44 plastic articles

Polymer type	Article purpose	Article name	Number of different commercial plastic articles analyzed
PVC	Peritoneal dialysis	Tubing, bag	2
	Transfusion	Tubing, transfer bag, quadrupole blood collection bag, Sag-M blood collection bag	5
PET	Food packaging and food handling equipment	Milk bottle, yogurt bottle, mineral water bottles, disposable plastic food container	15
PP	Food packaging and food handling equipment	Milk package bag, yogurt cups, sour milk cup, cutting board, freezer bag	12
PS	Food handling equipment	Disposable platter, egg box	2
PC	Food handling equipment	Packaging bag	1
Cellophane	Food handling equipment	Cellophane film	1
HDPE	Food handling equipment	Storage bag, granulate raw material	2
LDPE	Food handling equipment	Storage zip-bag, granulate raw material	2
	Infusion	Bottle for parenteral nutrition solution	2

0%, 5%, 10%, 20%, 30%, 40%, and 50% DEHP by weight. These calibration solutions were poured into evaporating dishes, and after THF evaporation, obtained calibration standards in the form of polymer disks were recorded on FTIR instrument.

Safety precautions and reduction of sample contamination by phthalates

Used solvents are volatile and flammable; therefore, care must be taken. THF is toxic by inhalation and ingestion and the evaporation must be done in the digester. To reduce laboratory phthalate contamination and since THF dissolves PVC, only glassware equipment was used. The glassware was cleaned using described procedure in the reference (Fankhauser-Noti and Grob 2007).

Determination of phthalates in PVC plastic materials by FTIR technique

The noted 7 PVC plastic articles in Table 2 were weighted (0.01000 g) and 4 mL of THF was added into vial with measured plastic. The vials were placed on the hot plate and covered with glass to prevent evaporation and loss of THF. After dissolving of plastic articles, THF solutions of plastics were poured into evaporation dishes. In this way, the polymer disks were created after evaporation of the THF for 30–40 min. The obtained polymer disks were mounted on an IR cell holder on FTIR instrument and FTIR spectrum of phthalates in polymer disks was obtained. The analysis was repeated three times.

Precipitation of PVC was done by adding 10 mL of *n*-hexane to THF solution of plastic (0.01000 g of PVC material dissolved in 4 mL THF), in order to obtain PVC precipitate without phthalates, since phthalates remained in the supernatant above the PVC precipitate. The PVC precipitate was separated from the supernatant by centrifugation (3500 rpm), dried in the oven at 80 °C, mixed in amalgamator with KBr,

and pellets were formed by manual hydraulic press. The obtained polymer pellets were mounted on an IR cell holder on FTIR instrument and FTIR spectrum was obtained. The analysis was repeated three times.

Determination of phthalates in PVC plastic materials by gravimetric technique

Precipitated PVC after adding 10 mL of *n*-hexane to THF solution of PVC plastic was separated from the supernatant by centrifugation at 3500 rpm and dried in the oven at 80 °C. Based on the difference of mass of PVC article and obtained PVC precipitate after phthalate removal, phthalate content was determined gravimetrically by the analytical balance with accuracy of ±0.00001 g.

Mass of the phthalates in PVC material was determined in the supernatant. After evaporation of THF from the supernatant, the obtained film with the plasticizers from the PVC sample was gravimetrically measured. The weight of the film represented mass of the phthalates in PVC.

Determination of phthalates in PVC plastic materials by GC-MS technique

The noted 7 PVC articles in Table 2 were weighted (0.01000 g) and dissolved in 4 mL THF. To obtain solutions of phthalate without plastic polymer, after dissolving of PVC articles in THF, 10 mL of *n*-hexane was added to the THF solution of each sample. Obtained turbid solutions were centrifuged at 3500 rpm and after that filtered through the 0.45 μm PTFE filter. A certain amount of THF-*n*-hexane solutions (10 μL) were diluted with *n*-hexane and after adding DBA as internal standard, samples were analyzed by GC-MS.

Beside usage of n-hexane as precipitating agent of PVC, methanol was also used and phthalate concentration in obtained THF-methanol solutions was also determined by GC-MS.

The gas chromatograph was operated in the split less injection mode. The oven temperature was programmed from initial temperature 90 °C (hold time 0 min) to 280 °C at a rate of 20 °C per min with hold time of 4 min, and post run 300 °C (2 min). Helium was the carrier gas (flow rate of 1.0 mL/min). The operating temperature of the MSD was 280 °C with the electron impact ionization (EI) voltage of 70 eV. The dwell time was 100 ms. The MSD was used in the single ion-monitoring mode (SIM), the quantification ion is m/z 149 for DnBP, BBP, DEHP, and DnOP, m/z 163 for DMP, and ion m/z 185 was chosen as representative ion of DBA. Analyte response was normalized to DBA as internal standard. The identification and quantification of target compound was based on the relative retention time, the presence of target ions, and its relative abundance. Both data acquisition and processing were accomplished by Agilent MSD ChemStation® D.02.00.275 software.

Migration testing of phthalates from plastic materials

The phthalate leaching potential from 44 plastic articles made from 7 plastic types into n-hexane was determined after 6, 15, and 30 days. This solvent was chosen as a worst-case scenario. Each of the 44 plastic articles was cut into pieces with area about 0.5 cm^2 and weighted on the analytical balance (the masses of a plastic pieces with indicated surface area were about 0.02000 g). n-Hexane (5 mL) was added to each plastic sample in glass vial and leaching of phthalates into this model solution under different conditions occurred. The following

conditions were applied: ultrasound at room temperature, prolonged contact time, and mixing within an appropriate time interval. Phthalates leaching was monitored within 6, 15, and 30 days. After the noted days, 900 μL of n-hexane was removed from n-hexane solution and DBA as internal standard was added (100 μg of DBA) to the removed aliquot. Concentration of phthalates was determined by GC-MS with the same method as previously described. The analysis of each sample was repeated three times.

Results and discussions

Determination of phthalates in PVC plastic materials by FTIR technique

The FTIR absorption spectrum of the polymer disk obtained after dissolution in THF of one PVC article (transfusion tubing) is given in Fig. 1a. It shows qualitative bands which identifies the phthalates. The useful band for infrared analysis of phthalates is strong absorbance band at 743 cm^{-1}, due to phthalate *ortho*-substituted aromatic ring. The doublet bands at 1579 cm^{-1} and 1599 cm^{-1}, appeared because of the aromatic ring quadrant stretching vibration, also identify the phthalates. The frequency and intensity of these bands in the infrared spectrum depend on the aromatic ring substitution. Other useful characteristic FTIR spectra absorption bands for phthalates are 2960 cm^{-1} (for -C-H bond), 1718 cm^{-1} (for the phthalate ester), 1450 cm^{-1} (for -CH2- and -CH3), 1278 cm^{-1} (for -CH2-), and 1073 cm^{-1} (for -CH3).

Two regions of the FTIR spectra of phthalates in PVC samples at 720–760 cm^{-1} and 1560–1600 cm^{-1} given in the

Fig. 1 The FTIR spectrum of transfusion tubing: **a** with identified characteristic bands for phthalate, **b** without characteristic bands for phthalate
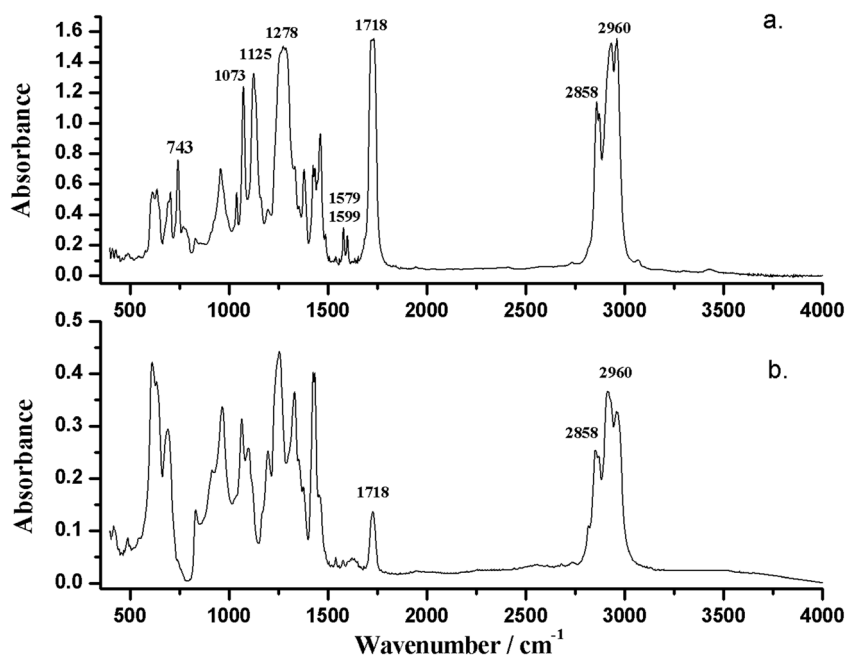
Fig. 2 show which PVC article contains the highest phthalate amount. The transfusion tubing contains the highest phthalate content, followed by tubing for infusion, quadrupole blood bag, transfer bag, tubing for dialysis bag, dialysis bag, and Sag-M bag.

This is a technique that does not give result for each phthalate separately but provide information about the total amount of phthalate in sample. The peak appearing in the spectrum is due to the absorption of all present phthalates at a given wavenumber. However, since the GC-MS method showed that only DEHP was present in all investigated PVC articles, the total phthalate content determined by the FTIR method corresponds to the total DEHP content. Therefore, further discussion will be about the FTIR determination of DEHP.

In order to quantify the DEHP in PVC articles, calibration standards of spiked polymer disks were recorded on the FTIR instrument. The calculated peak area in relation to concentration of DEHP was presented on the calibration graph in Fig. 3. The calibration curves obtained for DEHP within the concentration range from 0 to 50% are linear with correlation coefficient 0.997 and 0.996, for wavenumber ranges 720–760 cm^{-1} and 1560–1600 cm^{-1}, respectively. The data for calibration equations and determination coefficients are presented in Table 3. Although both linear relationships between DEHP concentration and peak area for both regions, 720–760 cm^{-1} and 1560–1600 cm^{-1}, show high value of determination coefficients, r^2, bearing in mind that the relationship for region 720–760 cm^{-1} has higher value of r^2 and value of slope of linear curve, this region was used for quantitive determination, while both regions were used to provide qualitative phthalate determination.
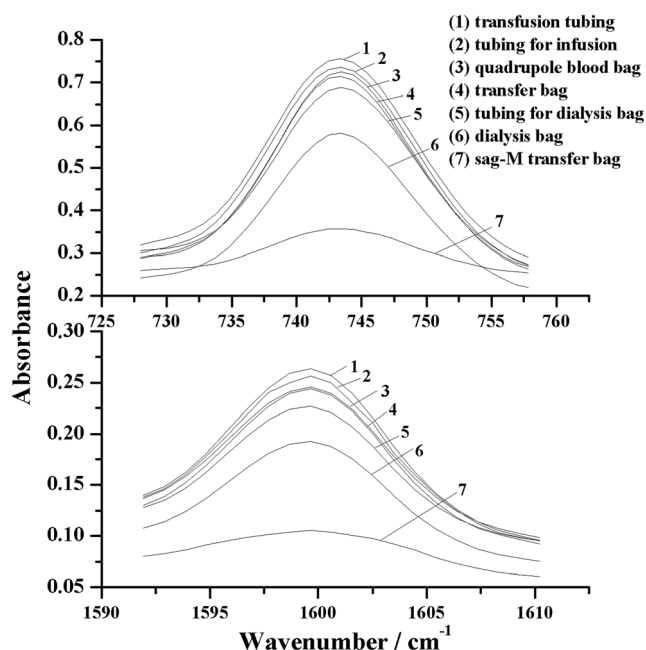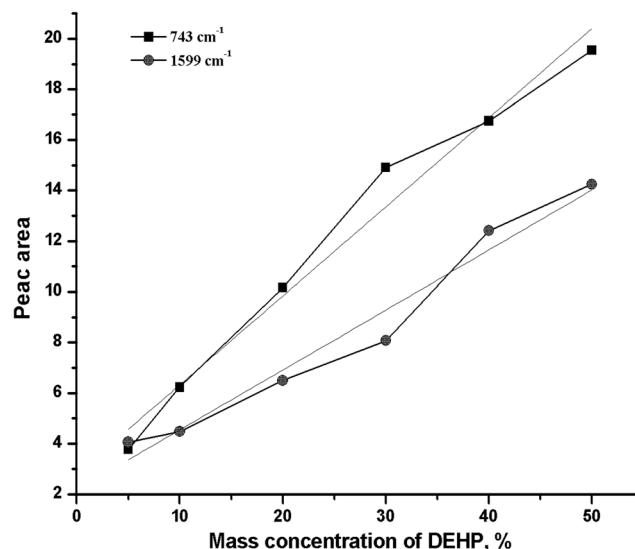

Fig. 3 Calibration curves for DEHP for different regions on FTIR spectrum: (a) 720–760 cm^{-1} and (b) 1560–1600 cm^{-1}

Based on the FTIR calibration curves for regions 720–760 cm^{-1} and 1560–1600 cm^{-1}, the mass of DEHP (expressed in mass percentages wt%) in each investigated PVC article was obtained and given in Table 4.

Obtained KBr pellets from all PVC reprecipitates were recorded by FTIR instrument and spectra show absence of characteristic absorbance bands for phthalates 743 cm^{-1}, 1579 cm^{-1}, and 1599 cm^{-1}. FTIR spectrum of transfusion tubing without phthalates is shown in Fig. 1b, as representative for all other spectra of PVC reprecipitates. Although both linear relationships between DEHP concentration and peak area for both regions, 720–760 cm^{-1} and 1560–1600 cm^{-1}, show high value of determination coefficients, r^2, bearing in mind that the relationship for region 720–760 cm^{-1} has higher value of r^2 and value of slope of linear curve, this region was used for quantitative determination, while both regions were used to provide qualitative phthalate determination.

Except the absence of specific absorbance bands, the spectrum also shows a reduced intensity of the absorbance of the other absorbance bands. These data indicate that the precipitation method of PVC by n-hexane completely releases phthalate from PVC samples. Identical FTIR spectra were obtained by recording a plastic disk obtained by evaporation of the dissolved precipitates in THF. Identical results were obtained even when methanol is used as a precipitating agent for PVC.

Determination of phthalates in PVC plastic materials by gravimetric technique

Mass of precipitated PVC after adding 10 mL of n-hexane to THF solution of PVC plastic was measured. Based on the difference of mass of PVC article and obtained PVC precipitate

Fig. 2 The FTIR spectrum of PVC samples: (a) region 720–760 cm^{-1} and (b) region 1560–1600 cm^{-1}

Table 3 Calibration equations and determination coefficients for FTIR determination of phthalates

Region	Calibration equation	Coefficients of determination, R^2
720–760 cm^{-1}	$y=(2.80\pm0.77)+(0.34\pm0.03)x$	0.997
1560–1600 cm^{-1}	$y=(2.17\pm0.65)+(0.24\pm0.02)x$	0.996

after phthalate removal, phthalate content was determined gravimetrically. The obtained phthalate-free PVC precipitate was non-flexible and soft confirming the absence of phthalates.

Mass of the phthalates in PVC article was determined in the supernatant after evaporation of THF/n-hexane from the supernatant. The obtained polymer film was elastic and highly flexible confirming the presence of phthalates. The obtained film with the phthalates from the PVC article was gravimetrically measured. The amount of phthalate determined gravimetrically is shown in Table 4.

Determination of phthalates in plastic materials by GC-MS technique

The chromatogram of a standard solution of the investigated phthalates and DBA, given in Fig. 4, shows good separation of five phthalates and DBA within a running time of 13.5 min.

For each phthalate, calibration curves were obtained by linear regression of the peak area against the concentration injected. The calibration curves were linear in the range from 0.25 to 2.5 µg mL^{-1} with correlation coefficient of calibration curves higher than 0.990. As an example of a calibration curve, the curve of DEHP is presented in Fig. 5.

The limit of determination (LOD) and limit of quantification (LOQ) were calculated from the signal/noise ratios which were multiplied with the factors 3 and 10, respectively (Wisconsin Department of Natural Resources Laboratory Certification Program 1996; Giupponi et al. 2018). The

retention times, correlation coefficients of calibration curves, LOD, LOQ, and relative standard deviation (RSD) ($n = 3$) of investigated phthalates are given in Table 4.

Based on the performed calibration, the concentration of phthalate in 37 non-PVC articles was determined, after leaching for 30 days from in n-hexane. The results show that non-PVC samples do not contain phthalate. Seven PVC articles are dissolved in THF and the phthalate was determined in the supernatant from PVC samples after the precipitation with n-hexane and methanol, separately. The results are also shown in Table 5. The results also indicate that the only phthalate that is in PVC articles was DEHP.

Comparison of FTIR, GC-MS, and gravimetric determination of phthalates in PVC articles

The analysis of variance (ANOVA) was used to evaluate differences in phthalate concentration determination between six methods used. These differences were found to be statistically non-significant ($p < 0.05$) since calculated F value (0.026) was lower than Ft$_{ab\ 5/36}$ at $p < 0.05$ (2.45), so the hypothesis that all means are equal is correct.

Leaching testing of phthalates from plastic articles and determination of phthalate leaching potential

The obtained chromatograms of n-hexane recipient solutions after leaching of phthalates from 44 plastic articles show the

Table 4 Comparison of FTIR, GC-MS, and gravimetric determination of phthalates in PVC articles

Plastic article name	Total phthalates weight % determined					
	By FTIR	By GC-MS			By gravimetric based on	
		In supernatant after precipitation by hexane	In supernatant after precipitation by methanol	After 30-day leaching	Phthalate mass in supernatant	Mass reduction before and after phthalate removal
Tubing for transfusion	27.5 ± 0.6	28.8 ± 0.6	29.8 ± 0.7	28.8 ± 0.7	29.3 ± 0.3	29.0 ± 0.4
Tubing for infusion	27.0 ± 0.3	26.1 ± 0.1	26.9 ± 0.6	26.4 ± 0.2	26.0 ± 0.5	26.1 ± 0.5
Quadrupole bag	24.1 ± 0.2	25.2 ± 0.1	25.9 ± 0.0	25.7 ± 0.1	25.5 ± 0.1	25.7 ± 0.2
Transfer bag	22.9 ± 0.2	24.8 ± 0.0	25.6 ± 0.4	25.2 ± 0.1	25.1 ± 0.1	25.2 ± 0.6
Dialysis tubing	21.8 ± 0.5	21.8 ± 0.4	22.4 ± 0.5	20.6 ± 1.1	21.0 ± 0.4	21.2 ± 0.4
Dialysis bag	17.9 ± 0.6	20.0 ± 0.6	20.6 ± 0.3	18.8 ± 1.1	19.2 ± 0.4	19.3 ± 0.4
Sag-M	4.98 ± 0.17	5.21 ± 0.14	5.38 ± 0.08	5.19 ± 0.01	5.50 ± 0.09	5.56 ± 0.19

Fig. 4 **a** GC-MS chromatogram of transfusion tubing made of PVC, after 30 days of leaching; **b** GC-MS chromatogram of a standard solution of investigated phthalates and DBA in a concentration of 1.5 μg mL^{-1} and 1.00 μg mL^{-1}, respectively
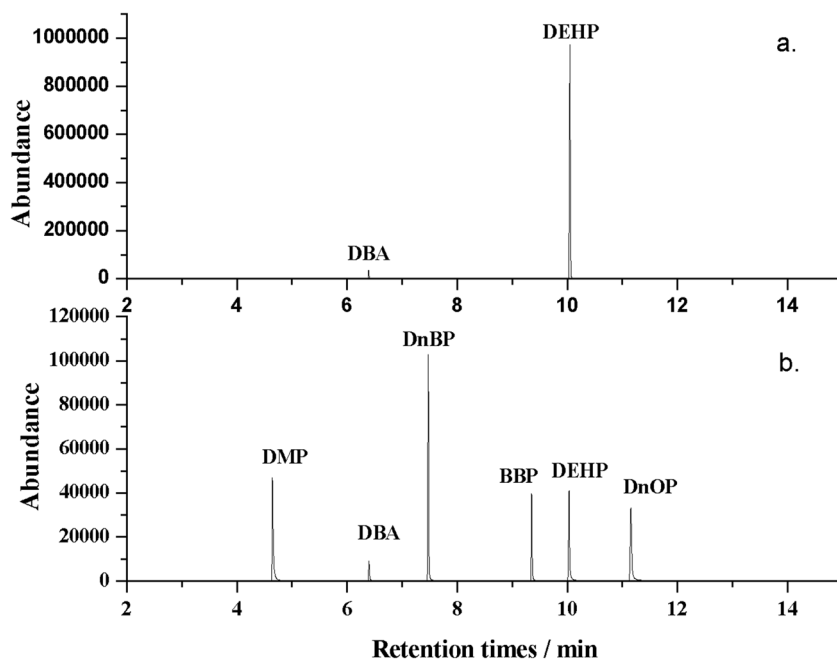
absence of DMP, DnBP, BBP, and DnOP in all 44 cases and the presence of DEHP in the 7 recipient solutions in which PVC articles were immersed and absence of DEHP in the rest of investigated articles, i.e., in 37 recipient solutions where non-PVC plastic articles were immersed.

The amounts of leached DEHP from the PVC articles in relation of leaching time are given in Table 6. DEHP was found in *n*-hexane recipient solutions in concentration levels of 5.19–28.76 wt% of PVC articles.

The leached concentrations of DEHP was compared using the Tukey's post hoc test, which determined whether

Fig. 5 GC-MS calibration curve for phthalates in the range from 0.25 to 2.5 μg mL^{-1}. P_A and P_{IS} presents the values for chromatogram peak of analyte and internal standard

there was a significant difference between the results obtained after different leaching times (6, 15, and 30 days). A significant difference is checked by comparing values of honest significant difference (HSD) for each pair of results to a critical value (4.34) (Hampton and Havel 2006). Values were computed by Origin© program, for $p < 0.05$. Obtained results indicate significant difference when leaching of DEHP lasted 6, 15, or 30 days for transfusion tubing, tubing for infusion set, quadruple blood bag, or transfer bag. In the cases of dialysis tubing, dialysis bag and SAG-M transfer bag results show no significant difference whether leaching lasted 6 days or 15 days.

In order to compare plastic articles, the DEHP leaching potential was determined for each article. The leaching potential is represented by partitioning coefficient plastic/hexane, $K_{H/P}$. This coefficient presents the ratio of phthalate mass in plastic material (mass obtained by GC-MS determination of phthalate in supernatant after precipitation by n-hexane) to dissolved phthalate mass in recipient solution after 6 and 15 days of phthalate leaching. The higher $K_{H/P}$ values indicate better chances that the phthalate will dissolve and the higher leaching potential. The leaching potentials of phthalates from plastics depend not only on a few parameters such as their molecular size, molecular weight, polarity, chemical bonds, solubility, boiling points, and initial concentration in the plastic polymer but also on properties of plastic material. The phthalate molecular size and boiling points increase with the increase of number of carbon atoms in the ester groups. Thus, small phthalate molecules have high leaching potential due to their low boiling points and small molecular sizes, which makes them easier to travel through polymeric plastic material. Since only DEHP was leached from the plastics samples, a
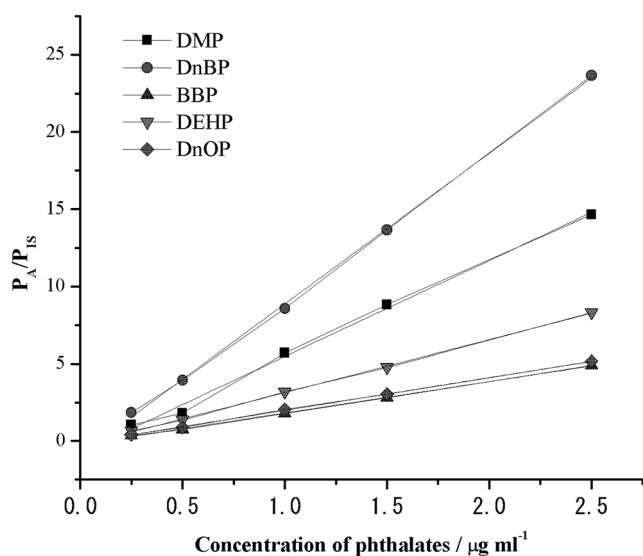
Table 5 Parameters for linearity and limits of detection and quantification for analyzed phthalates

	Calibration equations	Retention times (min)	R^2	LOD ($\mu g\ mL^{-1}$)	LOQ ($\mu g\ mL^{-1}$)	RSD (%) ($n = 3$)
DMP	$y=(-0.75\pm0.32)+(6.22\pm0.23)x$	4.64	0.995	0.03	0.10	7.14
DnBP	$y=(-0.88\pm0.21)+(9.75\pm0.15)x$	7.47	0.999	0.03	0.10	10.0
BBP	$y=(-0.22\pm0.04)+(2.04\pm0.03)x$	9.35	0.999	0.03	0.10	6.67
DEHP	$y=(-0.27\pm0.06)+(3.42\pm0.04)x$	10.0	0.999	0.03	0.10	6.67
DnOP	$y=(-0.13\pm0.03)+(2.12\pm0.02)x$	11.2	0.999	0.06	0.20	12.5

comparison of the type of plastic is made by comparing $K_{H/P}$ values of DEHP. Also, the influence of leaching time is shown by comparing these values. The $K_{H/P}$ values of DEHP are given in Table 7. Dialysis tubing, dialysis bag, and Sag-M have the strongest attachment of DEHP, in comparison with other PVC articles, but the differences are small. With the increase of the leaching time, the binding strength of phthalates to PVC articles decreases, so they are more easily released; therefore, $K_{H/P}$ is lower for leaching after 15 days compared to $K_{H/P}$ for leaching after 6 days.

The obtained results from GC-MS analysis of *n*-hexane receiving solutions for 37 non-PVC articles (PET, LDPE, HDPE, PP, PS, and PC) showed that migration of DMP, DnBP, BBP, DEHP, and DnOP did not occur even after 30 days, confirming that these types of polymers do not contain phthalates.

Conclusions

Since phthalates can pollute the environment by leaching from the most commonly used plastics (PET, LDPE, PVC, HDPE,

PP, PS, and PC) of great importance is testing of phthalate migration and determination of total amount of phthalates in plastic articles. The aim of this paper was to find the most optimal technique for determination of phthalates. Obtained results by GC-MS, FTIR, and gravimetric methods combined with dissolution/precipitation method are statistically non-significant ($p < 0.05$). In this study, it was found that the PVC PCMs contained DEHP in significant amount, from 5.19 to 28.75% in relation to mass of PVC article, which may endanger human health. The developed FTIR method is rapid and efficient, less expensive, and requires fewer steps in the preparation in relation to GC-MS analysis. The investigation also shown that quantitation of total amount of phthalate can be done using the FTIR method, but the deficiency of this method in relation to GC-MS is that phthalate separation cannot be performed as by GC-MS. Thus, FTIR can be used for pre-screening of phthalate content in PVC articles, and if the result is positive showing the presence of phthalate, the GC-MS analysis can be performed. It has been shown that dissolution/precipitation method (THF/*n*-hexane) is effective in the removal of the total amount of phthalate from PVC, thereby obtaining a stiff and rigid non-plasticized plastic. GC-MS analysis of leached phthalates from non-PVC plastic articles shows that these plastics do not contain any of the investigated phthalates and in view of these results, it can be concluded that the tested articles used as FCMs are safe.

Table 6 Concentration of leached DEHP (mg/g) from PVC articles determined for different leaching times (6, 15, and 30 days); *SD*, standard deviation ($n = 3$)

PVC article	Concentration of leached DEHP (mg/g)		
	Leaching time (days)		
	6	15	30
Transfusion tubing	139.4 ± 1.3^a	177.8 ± 3.4^b	287.5 ± 7.7^c
Tubing for infusion set	127.3 ± 1.2^a	161.4 ± 1.8^b	263.6 ± 6.4^c
Quadrupole blood bag	119.5 ± 1.5^a	152.6 ± 2.2^b	257.2 ± 6.0^c
Transfer bag	120.2 ± 0.6^a	154.6 ± 7.4^b	251.7 ± 3.8^c
Dialysis tubing	101.5 ± 1.3^a	109.9 ± 4.4^a	205.7 ± 7.2^b
Dialysis bag	91.50 ± 2.97^a	100.3 ± 5.0^a	188.1 ± 6.9^b
SAG-M transfer bag	23.60 ± 0.71^a	26.37 ± 2.16^a	51.90 ± 0.14^b

[a–c] Values with the same letter within a row are not statistically significant different at the $p < 0.05$ level (Tukey's HSD test)

Table 7 Partitioning coefficient hexane/plastic, $K_{H/P}$

PVC article	$K_{H/P}$		
	Leaching time (days)		
	6	15	30
Tubing for transfusion	0.48	0.62	1.00
Tubing for infusion	0.49	0.62	1.01
Quadrupole bag	0.47	0.61	1.02
Transfer bag	0.49	0.62	1.01
Dialysis tubing	0.47	0.51	0.94
Dialysis bag	0.46	0.50	0.94
Sag-M	0.45	0.51	1.00

Declarations

Ethics approval and consent to participate Not applicable.

Consent for publication Not applicable.

Competing interests The authors declare no competing interests.

References

Amanzadeh H, Yamini Y, Moradi M, Abdossalmi Asl Y (2016) Determination of phthalate esters in drinking water and edible vegetable oil samples by headspace solid phase microextraction using graphene/polyvinylchloride nanocomposite coated fiber coupled to gas chromatography-flame ionization detector. J Chromatogr A 1465:38–46. https://doi.org/10.1016/j.chroma.2016.08.068

Amin MM, Ebrahimpour K, Parastar S, Shoshtari-Yeganeh B, Hashemu M (2018) Method development of di-(2-ethylhexyl) phthalate metabolites detection by dispersive liquid–liquid microextraction gas chromatography-mass spectrometry from urine. Int J Env Health Eng. https://doi.org/10.4103/ijehe.ijehe_14_17

Anderson W, Castle L, Scotter M, Massey R, Springall C (2001) A biomarker approach to measuring human dietary exposure to certain phthalate diesters. Food Addit Contam A 18(12):1068–1074. https://doi.org/10.1080/02652030110050113

Bacha C, Dauchy X, Chagnon MC, Erienne S (2012) Chemical compounds and toxicological assessments of drinking water stored in polyethylene terephthalate (PET) bottles: a source of controversy reviewed. Water Res 46(3):571–583. https://doi.org/10.1016/j.watres.2011.11.062

Benjamin S, Masai E, Kamimura N, Takahashi K, Anderson R, Faisal PA (2017) Phthalates impact human health: epidemiological evidences and plausible mechanism of action. J Hazard Mater 340:360–383. https://doi.org/10.1016/j.jhazmat.2017.06.036

Bogdanovic D, Andjelkovic D, Kostic I, Kocic G, Anndjelkovic T (2019) The effects of temperature and ultrasound on the migration of di-(2-ethylhexyl) phthalate from plastic packaging into dairy products. Bulg Chem Commun 51(2):242–248. https://doi.org/10.34049/bcc.51.2.5027

Dural E, Determination of selected phthalates in some commercial cosmetic products by HPLC-UV. (2020) High throughput screening. Comb Chem https://doi.org/10.2174/1386207323666200630113850

European Union, Regulation (EC) No. 1272/2008 of the European Parliament and of the Council on classification, labelling and packaging of substances and mixtures, amending and repealing

Directives 67/548/EEC and 1999/45/EC, and amending Regulation (EC) No 1907/2006, 2008.

Fankhauser-Noti A, Grob K (2007) Blank problems in trace analysis of diethylhexyl and dibutyl phthalate: investigation of the sources, tips and tricks. Anal Chim Acta 582:353–360. https://doi.org/10.1016/j.aca.2006.09.012

French law Amendment n° 2012-1442 of december 24,2012 to the suspension of the manufacture, import, export and placing on the market of any specializing in food packaging containing bisphenol A (1) journal officiel de la république française n°0300, 26 décembre 2012 page 20395.

French law Amendment n° 2012-1442 of décembre 24, 2012, n°1610 to the French law voted 10 April, 10 2015.

Giupponi L, Pentimalli D, Manzo A, Panseri S, Giorgi A (2018) Effectiveness of fine root fingerprinting as a tool to identify plants of the Alps: results of a preliminary study. Plant Biosyst. 152(3):464–473. http://www.tandfonline.com/doi/full/10.1080/11263504.2017.1306003

Hadjmohammadi MR, Ranjbari E (2012) Utilization of homogeneous liquid–liquid extraction followed by HPLC-UV as a sensitive method for the extraction and determination of phthalate esters in environmental water samples. Int J Environ Anal Chem 92(11):1312–1324. https://doi.org/10.1080/03067319.2011.603049

Hampton R, Havel J (2006) Introductory biological statistics, 2nd. edn. Waveland Press, Long Grov, pp 99–120

Kissin Y, Liu X, David J, Pollick D, Brungard N, Chang M (2008) Ziegler-Natta catalysts for propylene polymerization: chemistry of reactions leading to the formation of active centers. J Mol Catal A: Chem 287(1-2):45–52. https://doi.org/10.1016/j.molcata.2008.02.026

Kostic I, Andjelkovic T, Andjelkovic D, Cvetkovic T, Pavlovic D (2016) Determination of di(2-ethylhexyl) phthalate in plastic medical devices. Hem Ind 70(2):159–164. https://doi.org/10.2298/HEMIND141129023K

Liang P, Linlin Zhang L, Peng L, Qian Li Q, Zhao E (2010) Determination of phthalate esters in soil samples by microwave assisted extraction and high performance liquid chromatography. Bull Environ Contam Toxicol 85(2):147–151. https://doi.org/10.1007/s00128-010-0078-x

Milojkovic D, Andjelkovic D, Kocic G, Andjelkovic T (2015) Evaluation of a method for phthalate extraction from milk related to the milk dilution ratio. J Serb Chem Soc 80(8):983–996. https://doi.org/10.2298/JSC141204028M

Qian S, Ji H, Wu XX, Li N, Yang Y, Bu J, Xiaoming Zhang X, Ling Qiao L, Yu H, Xu N, Zhang C (2018) Detection and quantification analysis of chemical migrants in plastic food contact products. Plos One. https://doi.org/10.1371/journal.pone.0208467

Rowdhwal SSS, Chen J (2018) Toxic effects of di-2-ethylhexyl phthalate: an overview. Biomed Res Int. https://doi.org/10.1155/2018/1750368

Rudel R, Perovich L (2009) Endocrine disrupting chemicals in indoor and outdoor air. Atmos Environ 43(1):170–181. https://doi.org/10.1016/j.atmosenv.2008.09.025

Silano V, Baviera JMB, Bolognesu C, Chesson A, Cocconcelli PS, Crebelli R, Gott DM, Grob K, Lampi E, Mortensen A, Riviere G, Steffensen IL, Tlustos C, Van Loveren H, Vernis L, Zorn H, Cravedi JP, Fortes C, Pocas MDFT, Waalkens-Berendsen I, Wolfle D, Arcella D, Cascio C, Castoldi AF, Volk K, Castle L (2019) Update of the risk assessment of di-butylphthalate (DBP), butyl-benzyl-phthalate (BBP), bis(2-ethylhexyl)phthalate (DEHP), di-isononylphthalate (DINP) and diisodecylphthalate (DIDP) for use in food contact materials. Efsa J 17(12):5838–5923. https://doi.org/10.2903/j.efsa.2019.5838

Visser R (2009) Reliability of proficiency test results for metals and phthalates in plastics. Accred Qual Assur 14:29–34. https://doi.org/10.1007/s00769-008-0465-8

Wisconsin Department of Natural Resources Laboratory Certification Program (1996) Analytical detection limit guidance & laboratory guide for determining method detection limits. Wisconsin Department of Natural Resources: Wisconsin, Madison, 1996. http://dnr.wi.gov/regulations/labcert/documents/guidance/-lodguide.pdf

Xin Gao X, Yang B, Tang Z, Xin Luo X, Wang F, Xu H, Cai X (2014) Determination of phthalates released from paper packaging materials by solid-phase extraction-high-performance liquid chromatography. J Chromatogr Sci 52(5):383–389. https://doi.org/10.1093/chromsci/bmt046

Zhou X, Shao X, Shu J, Liu M, Liu H, Feng X, Liu F (2012) Thermally stable ionic liquid-based sol–gel coating for ultrasonic extraction–solid-phase microextraction–gas chromatography determination of phthalate esters in agricultural plastic films. Talanta 89:129–135. https://doi.org/10.1016/j.talanta.2011.12.001

Ⓐ Springer

Terms and Conditions

Springer Nature journal content, brought to you courtesy of Springer Nature Customer Service Center GmbH ("Springer Nature").

Springer Nature supports a reasonable amount of sharing of research papers by authors, subscribers and authorised users ("Users"), for small-scale personal, non-commercial use provided that all copyright, trade and service marks and other proprietary notices are maintained. By accessing, sharing, receiving or otherwise using the Springer Nature journal content you agree to these terms of use ("Terms"). For these purposes, Springer Nature considers academic use (by researchers and students) to be non-commercial.

These Terms are supplementary and will apply in addition to any applicable website terms and conditions, a relevant site licence or a personal subscription. These Terms will prevail over any conflict or ambiguity with regards to the relevant terms, a site licence or a personal subscription (to the extent of the conflict or ambiguity only). For Creative Commons-licensed articles, the terms of the Creative Commons license used will apply.

We collect and use personal data to provide access to the Springer Nature journal content. We may also use these personal data internally within ResearchGate and Springer Nature and as agreed share it, in an anonymised way, for purposes of tracking, analysis and reporting. We will not otherwise disclose your personal data outside the ResearchGate or the Springer Nature group of companies unless we have your permission as detailed in the Privacy Policy.

While Users may use the Springer Nature journal content for small scale, personal non-commercial use, it is important to note that Users may not:

1. use such content for the purpose of providing other users with access on a regular or large scale basis or as a means to circumvent access control;

2. use such content where to do so would be considered a criminal or statutory offence in any jurisdiction, or gives rise to civil liability, or is otherwise unlawful;

3. falsely or misleadingly imply or suggest endorsement, approval , sponsorship, or association unless explicitly agreed to by Springer Nature in writing;

4. use bots or other automated methods to access the content or redirect messages

5. override any security feature or exclusionary protocol; or

6. share the content in order to create substitute for Springer Nature products or services or a systematic database of Springer Nature journal content.

In line with the restriction against commercial use, Springer Nature does not permit the creation of a product or service that creates revenue, royalties, rent or income from our content or its inclusion as part of a paid for service or for other commercial gain. Springer Nature journal content cannot be used for inter-library loans and librarians may not upload Springer Nature journal content on a large scale into their, or any other, institutional repository.

These terms of use are reviewed regularly and may be amended at any time. Springer Nature is not obligated to publish any information or content on this website and may remove it or features or functionality at our sole discretion, at any time with or without notice. Springer Nature may revoke this licence to you at any time and remove access to any copies of the Springer Nature journal content which have been saved.

To the fullest extent permitted by law, Springer Nature makes no warranties, representations or guarantees to Users, either express or implied with respect to the Springer nature journal content and all parties disclaim and waive any implied warranties or warranties imposed by law, including merchantability or fitness for any particular purpose.

Please note that these rights do not automatically extend to content, data or other material published by Springer Nature that may be licensed from third parties.

If you would like to use or distribute our Springer Nature journal content to a wider audience or on a regular basis or in any other manner not expressly permitted by these Terms, please contact Springer Nature at

onlineservice@springernature.com