Food Control 54 (2015) 374–382

Contents lists available at ScienceDirect

Food Control

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

Validation of QuEChERS method for organochlorine pesticides analysis in tamarind (*Tamarindus indica*) products: Peel, fruit and commercial pulp

Mário Paz [a, b], Luísa Correia-Sá [a], Helena Becker [b], Elisane Longhinotti [b], Valentina F. Domingues [a, *], Cristina Delerue-Matos [a]

[a] REQUIMTE, Instituto Superior de Engenharia do Porto, Instituto Politécnico do Porto, Rua Dr. António Bernardino de Almeida, 431, 4200-072 Porto, Portugal
[b] Departamento de Química Analítica e Físico-Química, Universidade Federal do Ceará (UFC), Cx. Postal 12200 CEP 60455-960 Fortaleza, CE, Brazil

ARTICLE INFO

Article history:
Received 27 December 2014
Received in revised form
4 February 2015
Accepted 5 February 2015
Available online 14 February 2015

Keywords:
QuEChERS
Tamarind
Organochlorine pesticides
GC-ECD
GC–MS/MS

ABSTRACT

In this study a citrate-buffered version of QuEChERS (Quick, Easy, Cheap, Effective, Rugged and Safe) method for determination of 14 organochlorine pesticides (OCPs) residues in tamarind peel, fruit and commercial pulp was optimized using gas chromatography (GC) coupled with electron-capture detector (ECD) and confirmation by GC tandem mass spectrometry (GC–MS/MS). Five procedures were tested based on the original QuEChERS method. The best one was achieved with increased time in ultrasonic bath. For the extract clean-up, primary secondary amine (PSA), octadecyl-bonded silica (C_{18}) and magnesium sulphate ($MgSO_4$) were used as sorbents for tamarind fruit and commercial pulp and for peel was also added graphitized carbon black (GCB). The samples mass was optimized according to the best recoveries (1.0 g for peel and fruit; 0.5 g for pulp). The method results showed the matrix-matched calibration curve linearity was $r^2 > 0.99$ for all target analytes in all samples. The overall average recoveries (spiked at 20, 40 and 60 μg kg^{-1}) have been considered satisfactory presenting values between 70 and 115% with RSD of 2–15 % (n = 3) for all analytes, with the exception of HCB (in peel sample). The ranges of limits of detection (LOD) and quantification (LOQ) for OCPs were for peel (LOD: 8.0–21 μg kg^{-1}; LOQ: 27–98 μg kg^{-1}); for fruit (LOD: 4–10 μg kg^{-1}; LOQ: 15–49 μg kg^{-1}) and for commercial pulp (LOD: 2–5 μg kg^{-1}; LOQ: 7–27 μg kg^{-1}). The method was successfully applied in tamarind samples being considered a rapid, sensitive and reliable procedure.

1. Introduction

The nutritional value of compounds with biological properties and health promoter effects has contributed significantly to the expansion and consumption of Brazilian exotic tropical fruits (UNICAMP, 2011; Vieira, Bezerra, Mancini-Filho, & Lima, 2011). Among these fruit stands out tamarind (*Tamarindus indica*) belonging to the Leguminosae family which is grown mostly in northeastern Brazil. The fruit is enclosed by a woody and brittle pod containing 3 to 8 seeds involved by an edible pulp.

The presence of bioactive compounds in various parts of tamarind (fruit and hull) is attractive for the use in the pharmaceutical industry, as laxative activity, expectorant, and in digestive and pulmonary problems (Matos, 2002). A recent study demonstrated the antimicrobial potential of commercial pulp of tamarind. The commercial pulp revealed growth inhibition of *Pseudomonas aeruginosa*, *Escherichia coli*, *Listeria monocytogenes*, Salmonella sp. and *Staphylococcus aureus* (Paz et al., 2015). Tamarind is essentially consumed in natural, processed frozen pulps, ice cream and juice concentrates (Gurjão, Bruno, Almeida, & Pereira, 2006).

Besides nutritional value, the fruit species can be a source of toxic substances due to the application of pesticides in the crop growing. In 2010, the application of these compounds in Brazil increased twice the world average (ca.190%) (ABRASCO, 2012). The

* Corresponding author. Tel.: +351 228340500; fax: +351 228321159.
E-mail address: vfd@isep.ipp.pt (V.F. Domingues).

http://dx.doi.org/10.1016/j.foodcont.2015.02.005
0956-7135/

Table 1
Retention time by GC-ECD and GC–MS/MS and identification ions for the GC–MS/MS analyses of selected OCPs.

Pesticides	GC-ECD	GC-MS/MS		
	Retention time (t_R)	Retention time (t_R)	Precursor ions (m/z)	Product ions (m/z)
α-HCH	9.7–9.8	8.05	183	179, 177
HCB	10.0	8.13	284	214, 249
β-HCH	10.4	8.30	183	179, 177
Lindane	11.2	8.44	183	179, 177
δ-HCH	11.9–12.0	8.75	183	109, 181
Aldrin	12.9–13.0	10.21	263	227, 193
α-Endosulfan	15.4	11.70	195	191, 170
p,p′-DDE	15.9	12.40	318	299, 281
Dieldrin	16.1–16.2	13.00	243	211, 176
Endrin	16.7–16.8	13.23	245	209, 173
DDT	17.0	13.60	235	165, 199
p,p′-DDD	17.3–17.4	14.04	235	165, 199
β-Endosulfan	17.5–17.6	14.10	195	191, 170
Methoxychlor	19.3–19.4	16.39	227	169, 197

deleterious action of these compounds can cause a range of clinical manifestations, such as nausea, dizziness, weakness, lack of appetite, headaches, allergies, kidney and liver damage, cancer, genetic alterations and neurological effects (Bakirci, Dilek, Bakirci, & Ötles, 2014).

There are over 800 compounds belonging to more than 100 different chemical classes, applied as pesticides in various cultures in Brazil (ANVISA, 2013). Among these classes, the main concern has been directed to the organochlorine pesticides (OCPs) due to bioaccumulation and potential toxic effects during decades (Bempah, Buah-Kwofie, Enimil, Blewu, & Agyei-Martey, 2012).

OCPs are semi-volatile organic compounds and can be transported by air over long distances, accumulating in matrices that did not have direct application and thereby be inserted into trophic levels through the food chain (Usman et al., 2014). These pesticides were banned for agricultural and domestic uses in Europe, North America and many countries in South America, according to the Stockholm Convention in 1980 based on their mutagenic potential, carcinogenic and teratogenic. Although these compounds prohibition and adverse effects on human health, they are still found in various food samples (Bakirci et al., 2014; Correia-Sá, Fernandes, Calhau, Domingues, & Delerue-Matos, 2013; Fernandes, Domingues, Mateus, & Delerue-Matos, 2012)

To ensure the safety of food for consumers, numerous legislations such as the EC directives (European Council Directives) have established maximum residue limits (MRLs), for pesticides in food. For non-usual fruits such as tamarind, the MRLs of same OCPs are correlated with limit of detection of method (European Commission, 2013). Studies involving the quantification of OCPs allow an estimation of human exposure and the presence in the environment of these substances, and contribute to the commercial regulatory decisions aimed at ensuring food security. Therefore, it is necessary to develop efficient and reliable analytical procedures for the determination of pesticide residues in food (Prestes, Friggi, Adaime, & Zanella, 2009).

Chromatographic methods are the most used for the determination of pesticides because they allow the separation of complex mixtures (Chiaradia, Collins, & Jardim, 2008). Gas chromatography (GC) combined with electron capture detector (ECD) is the most applied technique to the analysis of OCPs, since it has high selectivity and sensitivity for molecules containing electronegative functional groups (Fenik, Tankiewicz, & Biziuk, 2011). GC coupled with mass spectrometry (MS–MS) is also used because it provides a precise structural identification of analytes (Fernandes, Domingues, Mateus, & Delerue-Matos, 2011). These methods are recommended by the Environmental Protection Agency (EPA) and European standards for analysis of chlorinated pesticides (Chung & Chen, 2011).

As fruits are complex matrices and generally the concentration of pesticides found is low, it requires a step sample preparation prior to instrumental analysis. This step promotes the separation and enrichment of the analyte and the clean-up of the sample, if necessary (Prestes et al., 2009). Current trends prioritize the development of procedures for sample preparation that comply with principles of green chemistry (Anastas, 1999) e.g. decrease of organic solvent consumption. Accordingly, in 2003, Anastassiades et al. introduced a new method for the extraction of pesticides, mainly applied in fruits and vegetables seeking to overcome practical limitations of multiresidue extraction methods generally time-consuming and laborious. This method is described as quick, easy, cheap, effective, rugged and safe – QuEChERS (Anastassiades, Lehotay, Stajnbaher, & Schenck, 2003). When comparing with other techniques the QuEChERS method has proven to have several advantages due to giving excellent recoveries and involving less time and less solvent consumption (Lehotay, 2005).

Adaptions in the experimental procedure of the original QuEChERS method enabled the determination of different analytes in various food samples (Carneiro et al., 2013; Cieslik, Sadowska-Rociek, Ruiz, & Surma-Zadora, 2011; Fernandes et al., 2012; Restrepoa, Ortiza, Ossaa, & Mesaa, 2014). For these adaptions, an optimization of the procedure is required to maximize extraction efficiency, reducing errors and to obtain reliable results.

As far as the authors know there are not been reported an analytical methodology for determination of pesticide residues in tamarind which has unique characteristics. Thus, the purpose of the present work was to develop and validate a QuEChERS approach for the extraction of 14 OCPs in peel, fruit and commercial pulp of tamarind, applying GC-ECD analysis and GC–MS/MS confirmation.

Table 2
Tests performed for the analysis of OCPs in tamarind commercial pulp (m = 1 g; spiking level = 40 μg kg^{-1}).

Steps	Test 1	Test 2	Test 3	Test 4	Test 5
Extraction	Add 3 mL H_2O and 7 mL ACN	Add 3 mL H_2O and 7 mL ACN	Add 3 mL H_2O Vortex (1′) Ultrasonic bath (5′) Add 7 mL ACN	Add 2 mL H_2O Vortex (1′) Ultrasonic bath (5′) Add 7 mL ACN	Add 3 mL H_2O and 7 mL ACN
	Vortex (1′)	Vortex (1′)	Vortex (1′) Ultrasonic bath (5′)	Vortex (1′) Ultrasonic bath (5′)	Vortex (5′)
Partition	QuEChERS 1 Vortex (2′) Ultrasonic bath (10′) Centrifuge (10′)	QuEChERS 2 Vortex (2′) Ultrasonic bath (10′) Centrifuge (10′)	QuEChERS 2 Vortex (5′) Ultrasonic bath (10′) Centrifuge (10′)	QuEChERS 2 Vortex (5′) Ultrasonic bath (10′) Centrifuge (10′)	QuEChERS 2 Vortex (10′) Ultrasonic bath (30′) Centrifuge (10′)
Clean up		Add of Clean up with GCB			