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Simplified method for the determination of chlorinated fungicides and insecticides in fruits by gas chromatography

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Abstract

A fast, reliable method for the determination of more than twenty chlorinated fungicides and insecticides in a variety of fruit samples is presented. The pesticides are extracted from chopped samples with magnetic stirring, after adding 13 ml of acetone–phosphate buffer–brine solution (12:1, v/v) with 5 ml of *n*-hexane. The continuous module employed allows sequential decolourization of the organic phase, solvent changeover and solid-phase extraction for clean-up and preconcentration purposes. A 1-μl aliquot of the pesticides in ethyl acetate (eluent) is finally injected into the gas chromatograph for separation and identification. The method provides excellent clean-up despite the complexity of the matrices involved. Fruit samples (5–20 g) containing 0.1–1250 ng/g pesticides were analysed with a high precision (4–6%). After contamination of the fruit samples for 12 h, average recoveries >90% at fortification levels of 5–25 ng/g were obtained for most of the pesticides. Positive findings of these pesticides in fruits purchased at local markets were confirmed by GC–MS.

Keywords: Fruits; Food analysis; Pesticides; Organochlorine compounds

1. Introduction

The number of pesticides that are routinely applied to agricultural commodities to both lessen the detrimental effect of weeds, insects or diseases, and boost crop production, has dramatically increased in recent years. As a consequence, the determination of pesticide residues in crops has been strictly regulated by governments in all countries, with two basic aims, namely to detect the presence of forbidden pesticides on a particular commodity and to determine whether the concentrations of the pesticides used exceed their maximum residue limits (MRLs) [1,2]. The determination of pesticide residues in fresh fruits requires rapid, robust, efficient methods because of the short time between harvest and sale in markets. Undoubtedly, chlorinated fungicides and insecticides constitute one of the most important groups of hazardous organic contaminants owing to their high persistence and impact on human health [3].

A literature survey of available methods for the determination of chlorinated residues in fruits leads to the conclusion that these matrices are among the most troublesome. The procedures used are tedious and time-consuming, and involve several extraction and clean-up steps to remove the huge amount of potentially interfering compounds (mainly plant pigments and endogenous acids), which are generally present at higher concentrations than the pesticide residues themselves. The large volumes of organic solvents employed in the first step (typical-

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ly acetone [4–7], acetonitrile [8–11] and, more recently, ethyl acetate [12,13]), 50–200 ml, pose major hazards for both the environment and the operator. Moreover, conventional liquid–liquid partition sometimes results in emulsification, which can lower the accuracy and reproducibility of the analytical results [14]. In this regard, solid-phase extraction (SPE) is an effective tool for clean-up purposes. However, liquid–liquid extraction (LLE) cannot be replaced by SPE, although it can be used at a later stage. The typically low concentrations of pesticides in fruits and their also low MRLs (in some cases a few ng/kg) call for an additional preconcentration step by evaporation under a nitrogen stream; this is done by redissolving the dry residue in the most appropriate solvent for chromatographic separation. The separation, detection and identification of chlorinated pesticides has usually been carried out by gas chromatography (GC), using highly selective detectors such as electron-capture [4,5,11–13,15,16], or atomic emission [11,17] models for screening, and mass spectrometric ones for confirmation [5,6,8,9,13,18]. Recently, a semi-automatic method for the determination of organochlorine pesticides in vegetables was proposed [16]. The method uses a single LLE step to separate the analytes from the matrix; after simultaneous evaporation/redissolution of the residue in distilled water, analytes are preconcentrated/cleaned-up on an RP-C_{18} column, eluted with ethyl acetate and injected into a gas chromatograph for analysis.

This work was aimed at developing a method as simple as possible, involving minimal sample manipulation, for the screening and the sensitive and selective determination of chlorinated fungicides and insecticides in fruits. LLE of the analytes from the chopped sample is unavoidable, and is followed by automated clean-up. For this purpose, a continuous-flow system similar to one described elsewhere [16] was suited to the proposed application. The method is highly sensitive: it allows the identification and quantitation of up to 23 residual chlorinated fungicides and insecticides residues in amounts below their MRLs. In addition, the high efficiency of the clean-up procedure provides good blanks and avoids the interference of coextractives.

2. Experimental

2.1. Standards and sampling

Stock standard solutions of aldrin, captan, captafol, chlorbensid, chlordane, dichlofuanid, dicloran, dicofol, dieldrin, α- and β-endosulfan (3:1, w/w), endosulfan sulphate, endrin, hexachlorobenzene (HCB), heptachlor, iprodione, α-, β-, δ- and γ-hexachlorohexane (α-, β-, δ- and γ-HCH, 1:1:1:1, w/w), lindane (γ-HCH), methoxychlor, procymidone, and vinclozolin [all obtained from Riedel-de Haën (Seelze, Germany)], were prepared in acetone (by exception, HCB was dissolved in dichloromethane), at concentrations of 5 mg/ml, and stored in glass stoppered bottles at 4°C in the dark. A 2 *M* potassium phosphate buffer–brine solution was prepared by dissolving 87 g of K_2HPO_4 and 68 g of KH_2PO_4 in ~250 ml of distilled water. The pH was adjusted to 7.0 with dilute HCl or KOH and the solution made up to volume (500 ml). Finally, sodium chloride was added to the buffer solution up to saturation under constant stirring. 2,4-Dichlorophenol (internal standard), activated carbon and RP-C_{18} HPLC sorbent were supplied by Sigma (Madrid, Spain). All other reagents and solvents, in analytical grade or better, were purchased from Merck (Darmstadt, Germany).

Fruits were purchased at local markets in Córdoba, Spain. Because the legally tolerated limits of pesticide residues have been set for raw materials, samples were analysed unwashed, in a raw state [19]. Sampling was done according to the legally established protocol [20]. Thus, a raw laboratory sample consisting of 10–50 units (depending on the fruit) was reduced to 3–20 units following the most appropriate procedure (mainly quartering). For melon, five units were cut into slices of ~200 g and then reduced similarly to the other fruits. These units were cut into slices and some of them were chopped into smaller pieces to obtain the 5–20 g fractions required by the proposed method.

2.2. Instruments and apparatus

Chlorinated fungicides and insecticides were chromatographed on a fused-silica capillary column (30

m×0.25 mm I.D.) coated with 5% phenylmethylpolysiloxane (film thickness 0.25 μm) (Supelco, Madrid, Spain). Analyses were performed on a Hewlett-Packard 5890A gas chromatograph equipped with a ^{63}Ni electron-capture detection (ECD) system. Nitrogen, at a flow-rate of 1.0 ml/min, was used as carrier gas. The injector port and detector temperatures were kept at 225 and 325°C throughout. The oven temperature program was as follows: 120°C, held for 2 min; 8°C/min ramp to 180°C (hold 4 min); 8°C/min ramp to 255°C (hold 10 min) and 10°C/min ramp to 270°C (hold 2 min). Peak areas were measured with a Hewlett-Packard 3392A integrator. Confirmatory analyses were carried out on a Fisons 8000 GC instrument interfaced to a Fisons MD800 mass spectrometer and controlled by a computer running MASSLAB software (Thermo, Madrid, Spain); the chromatographic column and temperature program were both similar to those used with ECD. Ultrapure helium (6.0 Air Liquide), at a flow-rate of 1.0 ml/min, was employed as carrier gas. The injection port and transfer line temperatures were maintained at 225 and 300°C, respectively. The ion source temperature was 200°C for the 70 eV electron impact mode, with scanning from m/z 50 to 500. In all analyses, 1 μl of the organic extract was injected in the split mode (1:25 ratio).

The flow system comprised two peristaltic pumps (Gilson Minipuls-2) and two Rheodyne 5041 injection valves. Poly(vinyl chloride) and Solvaflex pumping tubes for aqueous solutions and *n*-hexane–methanol, respectively, PTFE tubing (0.5 mm I.D.) and commercially available connectors were also employed. A sorbent column was prepared by packing a commercial Omnifit glass column (2 cm×2.5 mm I.D.) with ~40 mg of RP-C_{18} sorbent material; small cotton beads were used on the ends to prevent material losses. A laboratory-made glass column (5 cm×4 mm I.D.) packed with ~100 mg of activated carbon was also constructed to decolourize the organic extracts from the fruit samples. A magnetic stirrer (Selecta, Barcelona, Spain) was also employed.

2.3. Procedure

The flow system used to extract the chlorinated fungicides and insecticides from the fruits is depicted in Fig. 1. A 5–20 g amount of chopped sample was weighed inside a 50-ml glass bottle and 13 ml of a solution containing the acetone–phosphate buffer–brine (12:1, v/v) was added, followed by 5 ml of extractant (*n*-hexane); once stoppered, the slurry was magnetically stirred for 5 min. After 1 min of standing, a volume of 2 ml of the upper organic phase, containing the pesticides, was aspirated at 1

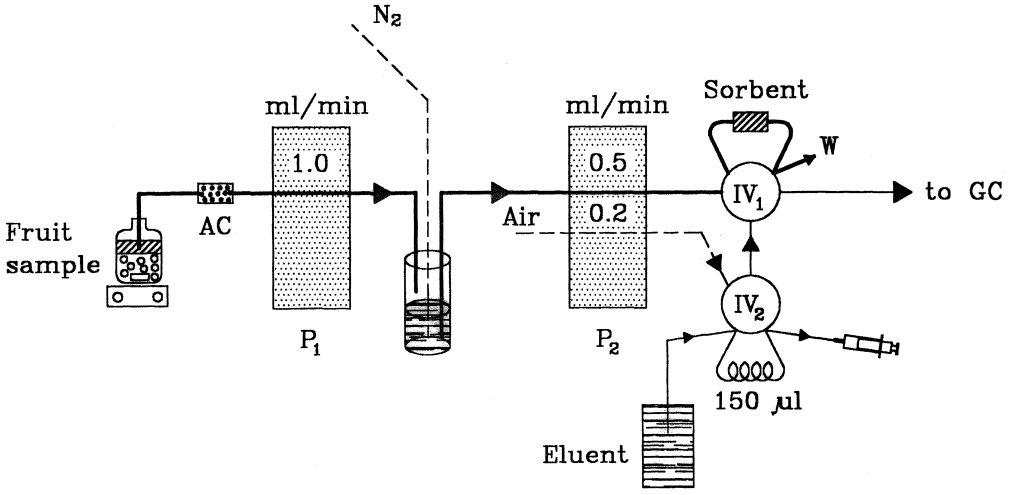

Fig. 1. Scheme of the clean-up/preconcentration steps used for the screening and determination of chlorinated fungicides and insecticides in fruits. P=peristaltic pump; IV=injection valve; W=waste; AC=activated carbon; GC=gas chromatograph with ECD or MS detector.

ml/min (sampling time 2 min) and passed through an activated carbon column for decolourization (0% adsorption of pesticides) and also to prevent suspended particles from reaching the continuous unit. The eluate was evaporated under a N_2 stream as it dropped into 5 ml of distilled water held in a glass tube. The nitrogen also helps to homogenize the solution. Once evaporation/redissolution was complete, the second pump was started, which allowed the analytes, dissolved in the 5 ml of water, to be instantaneously retained on the sorbent column, located in the loop of IV_1, at a flow-rate of 0.5 ml/min (bold line in Fig. 1). The loop of IV_2 (150 μl) was simultaneously filled with ethyl acetate (eluent) containing the internal standard (2,4-dichlorophenol, 1 μg/ml) by means of a syringe. In the elution step, both valves were switched simultaneously and the analytes eluted at 0.2 ml/min, using air as carrier. The whole organic extract (150 μl) was collected in a glass vial containing anhydrous sodium sulphate and a 1-μl aliquot injected into the gas chromatograph for analysis. Between samples, the activated carbon and the sample aspiration channel were flushed with 3 ml of methanol; the RP-C_{18} sorbent column and connectors were flushed with 3 ml of 0.2 *M* ammonium hydroxide to remove potentially retained interferents and then conditioned with 3 ml of water. Under these conditions, the activated carbon column was serviceable for about 25 analysis while the sorbent column remained active for 3 months.

3. Results and discussion

3.1. Selection and optimisation of the clean-up/preconcentration process

Pesticide residue analyses usually involve the extraction of the analytes from the fruit by using an organic solvent. Preliminary experiments were thus focused on examining the retention of the analytes, in an organic medium, onto a sorbent column, in order to minimize sample pretreatment (simultaneous preconcentration and clean-up). Various sorbents [RP-C_{18}, Florisil, silica gel, activated carbon, Serdolit, alumina, LiChrolut-EN and Amberlites (XAD-2 and XAD-7)] were assayed by preparing solutions of the analytes in acetone, acetonitrile, *n*-hexane and ethyl acetate. In the most favourable situation (1 g of silica-gel as sorbent and 4 ml of *n*-hexane as sample medium) only a preconcentration factor of four was achieved. In addition, the retention of the analytes spiked to a strawberry sample was found to be lower than that with the standards; also, the chromatograms were very dirty, hindering identification of some analytes and poor reproducibility was obtained.

From these results, the chemical conditions found in previous experiments for the automatic clean-up and preconcentration of organochlorine pesticides from vegetables [16], were assayed in the present work. Thus, aqueous medium for the extraction of organochlorine pesticides prior to direct insertion into the manifold for their adsorption on RP-C_{18}, was discarded as it provided low recoveries (~40%). Therefore, organic extractants are required, which involves a solvent changeover step to aqueous medium before preconcentration on RP-C_{18} sorbent. In the proposed method, a flow system similar to that depicted Fig. 1 was used, and the first variable studied was the organic extractant. Thus, the chopped test fruit sample was placed together with the extractant in a glass bottle and, after magnetic stirring, aliquots of the organic extract were continuously aspirated into the clean-up/preconcentration system (Fig. 1). Acetone and acetonitrile are commonly used extractants for this purpose on account of their miscibility with plant material, so they were tested with the strawberry samples. However, both extractants provided poor results (polar coextractives and pigments were also extracted, redissolved and eluted, leading to dirty chromatograms). Phase separation and pesticide re-extraction can also be accomplished by salting out with saturated NaCl in phosphate buffer [11], which prevents partitioning of endogenous organic acids (viz., citric, oxalic and fannic acids) into the aqueous–acetone layer. This method was assayed and the effect of the ratio acetone–phosphate buffer–brine solution (v/v) optimised. For this purpose, different samples of 5.0 g of strawberries were spiked with 100 ng per gram of fruit (1 ml of acetone containing 500 ng/ml of the pesticides to 5 g of fruit) and, after 12 h of standing (to simulate native analytes), the mixed solution was added, with a fixed acetone volume of 12 ml (for quantitative extraction) and a variable buffer volume

(1–10 ml); this slurry was extracted with 5 ml of *n*-hexane. The cleanest extracts were obtained by using 12 ml of acetone and 1 ml of phosphate buffer–brine solution, so this combination was selected for further experiments. The need of the buffer solution can be inferred from Fig. 2, which shows

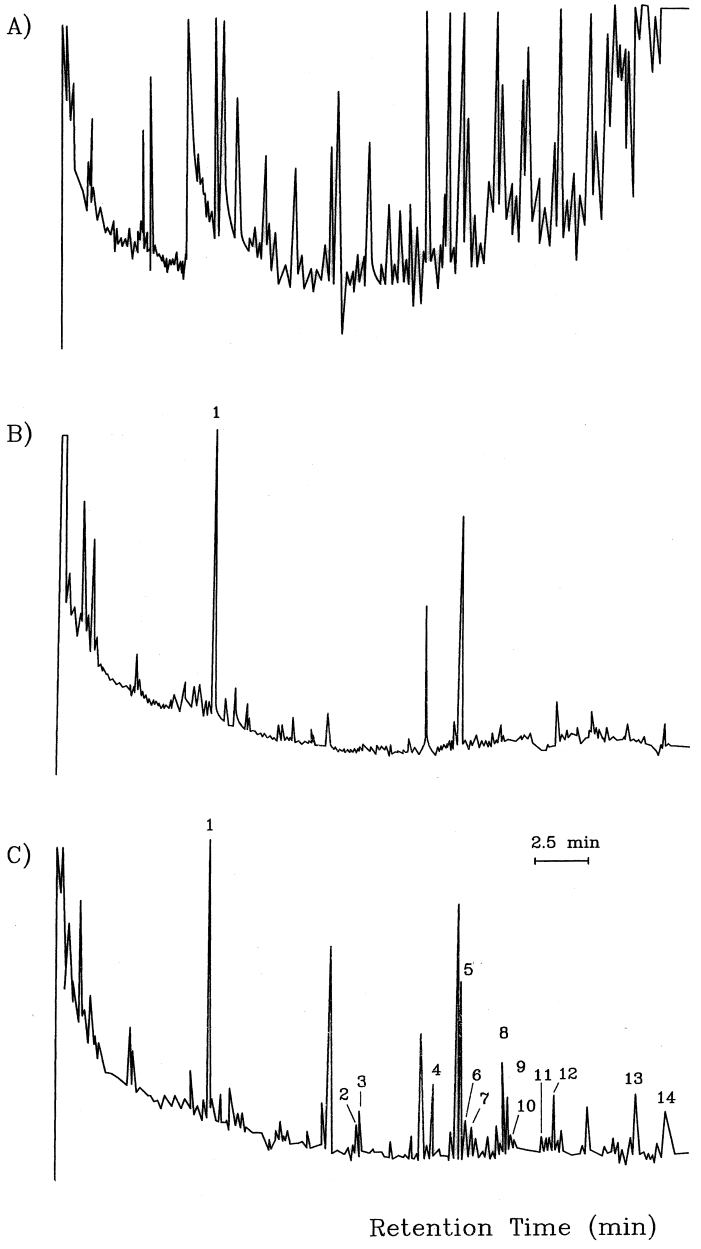

Fig. 2. Gas chromatograms for 5.0 g of unfortified strawberry sample extracted without (A) and with (B) a phosphate buffer–brine solution; same as (B) but fortified (C) with the 13 chlorinated fungicides and insecticides listed in Table 3. Peaks: 1=Internal standard, 2=HCB, 3=dicloran, 4=vinclozolin, 5=dichlofuanid, 6=aldrin, 7=dicofol, 8=captan, 9=chlorbenside, 10=chlordane, 11=dieldrin, 12=endrin, 13=captafol, 14=methoxychlor.

the chromatograms for an uncontaminated strawberry blank, without (A) and with buffer addition (B). Finally, the *n*-hexane volume was varied between 5 and 20 ml, taking into account that the final concentration of the pesticides in the organic phase remained constant, by spiking variable concentrations of the pesticides to the strawberry test sample. No significant differences were observed over the interval studied, so a volume of 5 ml of *n*-hexane was selected in order to increase the sensitivity of the proposed method; aliquots of 2 ml were inserted into the solid-phase separation unit as shown in Fig. 1.

As the *n*-hexane phase was yellowish, a glass column packed with 100 mg of activated carbon was placed at the entry to the sample aspiration channel in order to remove natural pigments present in the fruits. This resulted in cleaner chromatograms and an increased RP-C_{18} column lifetime; moreover, activated carbon did not retain pesticides (0% adsorption) when they are passed in a low polarity solvent as *n*-hexane; so this column was only used for clean-up purposes. The *n*-hexane phase containing the analytes was then evaporated and redissolved in 5 ml of distilled water as in previous work [16]. In this aqueous medium, several sorbent materials (RP-C_{18}, Amberlite XAD-2, silica gel and activated carbon) were assayed, and RP-C_{18} exhibited the best sorption properties (ca. 90%). Different eluents (ethyl acetate, *n*-hexane and light petroleum) were evaluated using RP-C_{18} as sorbent. Ethyl acetate was the best eluent (~100%). Taking into account that the sorbent and eluent were coincident with those of the elsewhere described method [16], the optimal values found for the variables (amount of sorbent material, sample and eluent flow-rate and eluent volume) were also adopted here (see Fig. 1).

3.2. Analytical GC performance

All the pesticides studied exhibited good gas chromatographic properties and could be identified in a direct manner without overlap. The strawberry blank was spiked with the 23 pesticides. The addition procedure was as follows: 1 ml of standard solutions in acetone containing all pesticides at variable concentrations was spiked to individual amounts of 5 g of fruit blanks, in the interval 0.4–1250 ng/g, held in 50-ml glass bottles; the strawberry sample was then extracted with 13 ml of acetone–buffer solution (12:1, v/v)+5 ml of *n*-hexane in the manifold of Fig. 1. This addition procedure ensured that the analytes in the strawberry sample would be present primarily in a deposited state; as a result, the recoveries for the 23 pesticides in these spiked strawberries were higher than 95%, relative to aqueous standards directly inserted into the solid-phase extraction unit (100%). Based on these results, the fruit blank was used to run calibration graphs for the 23 pesticides. The regression equations were obtained by plotting the analyte-to-internal standard peak area ratio against the analyte concentration, using eight points per curve. The sensitivity (slope of the calibration graph) and the linear ranges are listed in Table 1. The limits of detection were calculated as the minimum concentrations providing chromatographic signals three times higher than background noise; iprodione exhibited the highest limit of detection (25 ng/g) by virtue of its slightly broadened peak. Repeatabilities (obtained from 11 injections of the same sample containing 5–25 ng/g of each pesticide except iprodione, which was used at 80 ng/g), expressed as relative standard deviation, are also listed in Table 1. In any case, the sensitivity of the method can be increased 4 times by using greater amount of fruit (20 g), with negligible changes in precision. The *m/z* values used for identification in the electron impact ionization mode are shown in Table 1.

3.3. Application to fruit samples

Various types of fruit were used to examine the effect of the matrix on recoveries, separation and interfering peaks. For this purpose, 40 fruit samples of 10 different types (namely, nectarine, melon, peach, pear, apple, orange, red plum, cherry, strawberry and banana), purchased at various local markets were analysed. Sampling was done as described in Section 2.1 and laboratory samples were analysed as soon as received, using the procedure described in Section 2.3. Initially, 5.0 g of each fruit was weighed and analysed; when negative results were obtained, the sample amount was increased to 20 g. In all instances, quantitation was done by ECD and positive findings were confirmed by MS. Only six samples were found to contain chlorinated residues

Table 1
Figures of merit for the determination of chlorinated fungicides and insecticides using an SPE system

Compound	Sensitivity[a] (10^{-2})	Linear range (ng/g)	Limit of detection (ng/g)	RSD (%)	m/z[b] (ng/g)
α-HCH	2.3	2.5–500	0.7	5.4	111, **181**, 219
HCB	1.5	5.0–500	1.0	4.9	249, *282*, **284**
Dicloran	9.2	0.4–250	0.1	4.6	**124**, 176, *206*
β-HCH	2.7	5.0–500	1.0	4.6	**109**, 181, 219
Lindane	3.8	2.5–500	0.5	5.6	111, **181**, 219
δ-HCH	4.8	2.5–500	0.5	4.0	111, **181**, 219
Vinclozolin	4.8	2.5–500	0.5	5.7	**187**, 212, *285*
Heptachlor	1.3	5.0–500	1.0	5.5	**100**, 272, *370*
Dichlofuanid	2.5	5.0–500	1.2	5.8	**123**, 224, *332*
Aldrin	2.4	2.5–500	0.5	5.4	**66**, 263, 298
Dicofol	0.9	6.2–500	1.2	4.8	111, **139**, 250
Captan	5.6	2.5–500	0.7	5.0	**79**, 264, *299*
Procymidone	2.5	5.0–500	1.0	5.9	67, **96**, *283*
Chlorbensid	0.7	12.5–1000	2.5	6.0	**125**, 127, *268*
Chlordane	0.2	25.0–1250	5.0	5.1	202, **373**, 408
α-Endosulfan	1.4	5.0–500	1.0	5.8	**195**, 241, 339
Dieldrin	0.9	5.0–500	1.2	6.0	**79**, *378*, 380
Endrin	1.0	5.0–500	1.0	5.9	**67**, 317, 319
β-Endosulfan	1.1	5.0–500	1.0	5.7	**195**, 241, 339
Endosulfan sulphate	2.7	5.0–500	1.0	5.4	**272**, 387, *420*
Iprodione	2.1	63.0–1250	25.0	5.3	**56**, 314, *329*
Captafol	3.6	5.0–500	1.2	5.7	**79**, *347*, 349
Methoxychlor	0.4	12.5–1000	2.5	5.9	**227**, 228, *344*

[a] Relative area (analyte/internal standard peak area ratio)/ng/g.
[b] Values of m/z in italics are M^+ values; those in bold face correspond to the base peak.

at detectable levels. The results thus obtained, for 5 g of sample are listed in Table 2. As can be seen, the concentrations found were lower than the EU maximum residue limits (see Table 2) for all compounds examined. The presence of the residues was confirmed by GC–MS, using 20 g of sample as the sensitivity was ten times lower than with ECD. By way of example, Fig. 3A shows the chromatogram for the peach sample containing only vinclozolin, obtained with electron capture detection. Fig. 3B illustrates the identification of vinclozolin by comparing the mass spectrum for the corresponding peak with that in the library. Spectral comparison resulted in coincidence above 85%.

Recoveries of the proposed method were assessed by adding, to each previously analysed uncontaminated fruit, the pesticides according to use and occurrence/appearance [19,21,22]. In order to ensure

Table 2
Summary of the chlorinated fungicides and insecticides found in fruit samples (±SD, $n=6$) analysed by using the proposed method and GC–ECD

Sample	Pesticide found	Concentration (ng/g)	MRL[a] (μg/g)
Peach	Vinclozolin	6.0±0.3	2
Nectarine	Captan	5.0±0.2	2
Banana	Dicofol	7.3±0.4	0.5
Strawberry	Dichlofuanid	6.4±0.4	10
Apple	Lindane	4.5±0.3	1
Strawberry	Vinclozolin	4.8±0.3	5

[a] Established by the European Union.

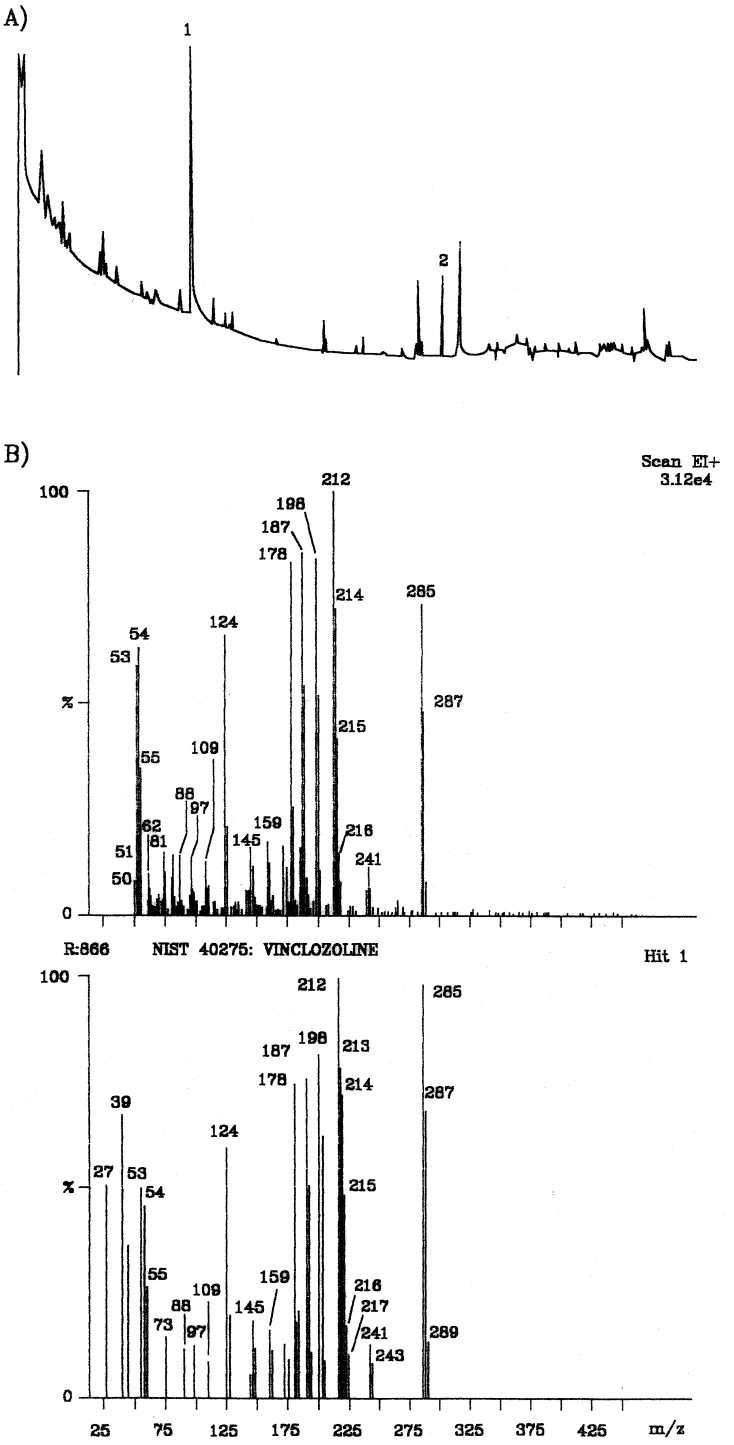

Fig. 3. Chromatogram for a chlorinated pesticide found in a peach sample (A): 1=internal standard, 2=vinclozolin. Full-scan EI mass spectra for vinclozolin in the fruit sample and from NIST library (B).

reliable simulation of the actual leaf applications [22], 5.0 g of chopped sample was spiked with 1 ml of acetone containing variable concentrations of chlorinated pesticides and fungicides between 25 and 125 ng/ml, except for iprodione (400 ng/ml), and allowed to stand overnight at room temperature in a

Table 3
Percent recovery (mean of six determinations±SD) of chlorinated fungicides and insecticides added to fruit samples at 5–25 ng/g

Commodity	Compound	Recovery (%)	Commodity	Compound	Recovery (%)
Nectarine	Aldrin	101±6	Orange	Aldrin	99±6
	Captan	75±4		Captafol	100±5
	Chlorbensid	97±6		Captan	79±4
	Dichlofuanid	107±7		Chlorbensid	99±6
	Dieldrin	96±5		Chlordane	92±5
	α-Endosulfan	91±5		Dichlofuanid	102±6
	β-Endosulfan	109±5		Dichloran	88±4
	Endosulfan sulphate	83±6		Dicofol	109±4
	Endrin	103±5		Dieldrin	99±7
	HCB	97±7		Endrin	96±4
	α-HCH	87±5		HCB	96±5
	β-HCH	89±5		Heptachlor	88±6
	δ-HCH	101±6		Lindane	103±5
	Lindane	103±5		Procymidone	84± 5
	Procymidone	83±7		Methoxychlor	90±5
				Vinclozolin	81±4
Melon	Aldrin	100±6			
	Chlorbensid	98±6	Red plum	Aldrin	89±5
	Dichlofuanid	99±6		Captafol	89±6
	Dichloran	83±5		Chlorbensid	93±6
	Dicofol	102±4		Chlordane	91±6
	Dieldrin	105±7		Dichloran	84±5
	Endrin	104±6		Dieldrin	100±5
	HCB	92±5		Endrin	108±7
	α-HCH	91±6		HCB	99±6
	β-HCH	96±5		Heptachlor	94±6
	δ-HCH	103±4		Methoxychlor	92±4
	Heptachlor	88±5		Procymidone	70±5
	Lindane	91±6		Vinclozolin	71±5
	Methoxychlor	102±6			
	Procymidone	72±4	Cherry	Aldrin	90±6
	Vinclozolin	81±5		Chlorbensid	99±6
				Chlordane	85±5
Peach	Aldrin	95±5		Dichlofuanid	90±6
	Dichloran	95±6		Dichloran	82±6
	Dieldrin	101±6		Dicofol	90±7
	α-Endosulfan	92±6		Dieldrin	91±6
	β-Endosulfan	106±7		α-Endosulfan	103±6
	Endosulfan sulphate	91±6		β-Endosulfan	101±7
	Endrin	101±6		Endrin	96±5
	HCB	91±4		HCB	84±6
	Methoxychlor	100±4		α-HCH	95±7
	Procymidone	78±5		β-HCH	96±7
	Vinclozolin	80±5		δ-HCH	97±5
				Lindane	99±5
				Methoxychlor	98±6
				Vinclozolin	71±4

closed fume hood to avoid contamination. Each sample was spiked three times and then analysed in duplicate ($n=6$), using the proposed method; the results obtained are listed in Table 3. All compounds were correctly identified and average recoveries (90–101%) were acceptable for all matrices. The lower values obtained for captan, procymidone and vinclozolin (77–80%) can be ascribed to their being either partially irreversibly bound to the matrices or degraded during contamination time. Matrix interferences were reduced during the clean-up steps. Fig. 2 shows the chromatograms for a strawberry sample, both unfortified (B) and after fortifying with the 13 chlorinated fungicides and insecticides listed in Table 3 (C).

4. Conclusions

The aim of this work was satisfactorily fulfilled, especially with regard to obtaining clean extracts from the fruits. These matrices are among the most troublesome to analyse, not only because of the bright colour of the natural pigments they contain (e.g. strawberries), but also because of the many major and minor compounds that are initially co-extracted with the analytes. Liquid extraction of the pesticides from the sample could not be avoided with any of the approaches tested. The sensitivity of the method complies with the limits established by legislation; the method uses amounts of a few grams of sample that can be increased if required. This is

Table 3 (*continued*)

Commodity	Compound	Recovery (%)	Commodity	Compound	Recovery (%)
Pear	Aldrin	105±6	Strawberry	Aldrin	98±6
	Captafol	94±4		Captafol	95±5
	Chlorbensid	103±6		Captan	87±5
	Dichlofuanid	90±5		Chlorbensid	95±6
	Dichloran	95±4		Chlordane	81±5
	Dicofol	105±5		Dichlofuanid	94±6
	Dieldrin	92±5		Dichloran	91±5
	α-Endosulfan	93±6		Dicofol	98±5
	β-Endosulfan	97±6		Dieldrin	93±6
	Endosulfan sulphate	92±5		Endrin	101±5
	Endrin	93±5		HCB	87±5
	Methoxychlor	104±6		Methoxychlor	92±5
	Procymidone	78±6		Vinclozolin	81±5
	Vinclozolin	70±4			
			Banana	Aldrin	90±5
Apple	Aldrin	90±6		Captafol	90±6
	Chlorbensid	91±6		Chlorbensid	102±7
	Chlordane	93±6		Dicofol	90±4
	Dichlofuanid	102±6		Dieldrin	95±6
	Dichloran	98±7		α-Endosulfan	106±5
	Dicofol	90±5		β-Endosulfan	96±6
	Dieldrin	94±5		Endrin	102±7
	α-Endosulfan	95±6		HCB	98±4
	β-Endosulfan	98±6		α-HCH	92±5
	Endosulfan sulphate	90±5		β-HCH	98±5
	Endrin	90±6		δ-HCH	102±5
	HCB	88±4		Heptachlor	99±5
	Heptachlor	82±5		Lindane	106±7
	Lindane	93±7		Vinclozolin	78±5
	Vinclozolin	82±6			
	Iprodione	86±5			

therefore a competitive, simple, fast and inexpensive method.

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