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EU-Reference Laboratory for pesticides requiring Single Residue Methods (EURL-SRM)

hosted at

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Abstract

A quick multi-method for fumigants (QuMFu) allowing their simultaneous analysis in cereals and dried fruits was developed. The method involves a simple extraction step with n-hexane followed by centrifugation and GC-MS/MS analysis. The following fumigants were investigated in the study:

- 1,2-Dibromo-3-chloropropane
- 1,3-Dichloropropene
- Azobenzene
- Carbon tetrachloride
- Chloropicrin
- Ethylene chlorobromide
- Ethylene dibromide (1,2-dibromoethane)
- Naphthalene
- p-Nitrochlorobenzene
- p-Dichlorobenzene
- 1,1,2,2-Tetrachloroethane
- Tetrachloroethylene
- Trichloroethylene

Chlorobenzene D5 was used as internal standard. Most of the substances showed a linear concentration to signal-intensity relationship both in pure solvent and extracts in the range between 0.01 µg mL^{-1} and 2 µg mL^{-1}. All substances except trichloroethylene showed matrix-induced signal suppression effects. The method was validated in raisins and wheat via recovery experiments at spiking levels of 0.01 mg kg-1/0.05 mg kg^{-1} and 0.1 mg kg^{-1}. Average recoveries of the individual compounds (n = 5 at both levels) ranged between 79 % and 106 % (RSD 1.0 % – 10.5 %) in wheat and between 86 % and 109 % (RSD 0.9 % – 9.9 %) in raisins. A small number of dried fruit samples from the market were tested, but none of those contained any detectable residues of the tested fumigants.

Introduction

Fumigants are gaseous pesticides used for the prevention and eventual disinfestation of pests. Consisting of small molecules, they are typically gaseous at 20 °C and diffuse quickly [1], [2]. Fumigants are mainly used to counter two problems associated with globalized trade. Firstly, they protect goods from spoilage during long transports through different climatic zones. Secondly, they prevent the introduction of (harmful) organisms to importing countries [1].

With the ratification of the Montreal Protocol on substances that deplete the ozone layer in 1987, many halogenated fumigants included in this study as well as methyl bromide, that used to be the most widely used fumigant, are currently being phased out on a worldwide level [3]. Alternative fumigants such as sulfuryl fluoride and phosphine are thus increasingly employed [4].

Analytical Approaches

In studies that were carried out during the 80s, fumigants could be detected in both unprocessed (grain, cereals, fish) and processed foods (jellies, chocolate sauce, dairy products, butter). The Review of Daft et al 1991 gives a comprehensive overview of the findings [5]. After 1991, studies have been published on carbonyl sulfide mainly [6], [7]. At the moment only a few current data on residues of fumigants in food exist. Several

methods describe the simultaneous analysis of fumigants in food [8], [9], [10], [11], [12] and other commodities [13]. The most commonly used approaches of analysis include solvent extraction [10], [11], [12], headspace sampling [14], [15], [16], [13] extraction with organic solvents [17], [18], [9], [19] co-distillations with water [20] as well as purge-and-trap techniques [21]. Reviews of existing methods in food were published by Daft [5] and by Desmarchelier & Ren [22].

Both EU and the Codex Alimentarius Commission regulate residues of methyl bromide in food indirectly via the bromide ion. Laboratories thus focus on the analysis of bromide ion using, in most cases, procedures involving derivatization with propylene oxide and GC analysis of the derivative [23]. Sensitive methods for the detection of phosphine in food, one of the most commonly used, inexpensive, and fast acting fumigants were recently reported by Amrein et al. [24], Amstutz et al. [25], and Perz et al. [26].

Methods for the analysis of fumigants have also been reported for commodities other than food [13]. Fahrenholtz et al. developed a method for the determination of phosphine, volatile organic fumigants and industrial chemicals in the air of containers via thermodesorption-2-dimentional - gas chromatography - mass spectrometry/flame photometry [27]. EPA Method 8260b describes the determination of volatile organic compounds in soil using GC-MS. It entails various extraction, purification and measurement steps, such as direct purge and trap and headspace injections [28].

Legal Aspects and Enforcement

Due to health and environmental hazards [29] associated with the use of fumigants, maximum residue limits (MRLs) in food products have been established. An overview of the MRLs for fumigants included in this study is shown in Table 1. Some of the fumigants are not listed in Regulation EC No 396/2005 (see [d] in Table 1). However, if they are used for the protection of stored products, they are classified as pesticides with the default MRL of 0.01 mg kg^{-1} applying.

Although residues of fumigants in food are regulated in the EU, still very little is known about the residue situation in this compound group, since hardly any official controls take place in EU laboratories.

Table 1 EU-MRLs for fumigant residues in food (as of 30 September 2014)

Substance	Maximum Residue Level (mg kg^{-1})
1,2-Dibromo-3-chloropropane	0.01[d]
1,3-Dichloropropene	0.01* (products of animal origin); 0.05* in most products of plant origin 0.1* in certain products (e.g. brassica, garlic, celery)
Azobenzene (diphenyldiazene)	0.01[d]
Carbon tetrachloride	0.01[d]
Chloropicrin	0.02* (tea, spices) 0.01[d] (cereals)
Ethylene chlorobromide	0.01*
Ethylene dibromide	0.01[d] (fresh or frozen fruit, nuts, fresh or frozen vegetables, pulses (dry), cereals, sugar plants
Naphthalene	0.01[d]
p-Dichlorobenzene	0.01[d]
p-Nitrochlorobenzene	0.01[d]
1,1,2,2-Tetrachloroethane	0.01[d]
Tetrachloroethylene	0.01[d]
Trichloroethylene	0.01[d]

[d] Default MRL according to Article 18 (1B) Regulation (EC) No 396/2005 [30]

* MRL corresponds to LOQ

About the present work

The aim of this work was to develop a quick multi-method for fumigants (QuMFu) allowing the simultaneous analysis of the above mentioned compounds in cereals and dried food via GC-MS/MS following a simple extraction with a non-polar solvent. Furthermore a certain number of samples from the market were to be checked for the presence of residues and any potential interferences in analysis. In Part II of this paper we will present further validation data using GC-ECD instead of GC-MS/MS for analysis, as well as the results of additional samples from the market (paper in preparation). The present method was developed by the European Reference Laboratory for pesticides requiring Single Residue Methods (EURL-SRM) financed by DG-SANCO.

Experimental

Chemicals and Standards

The solvent n-hexane of EMPLURA® grade was purchased from Merck KGaA (Darmstadt, Germany). Helium 5.0, used as the carrier gas for gas chromatography, was supplied by Praxair. Argon 5.0 (Praxair) was used as the collision gas for GC-MS/MS analysis.

Table 2 List of substances and their sources of supply

Substance	Purity	Company
Carbon tetrachloride	≥ 99.5 %	Dr. Ehrenstorfer GmbH (Augsburg, Germany)
Trichloroethylene	≥ 99.6 %	
Ethylene chlorobromide	≥ 99.5 %	
1,3-Dichloropropene (cis+trans)	≥ 92 %	
Tetrachloroethylene	≥ 99 %	
1,1,2,2-Tetrachloroethane	≥ 98.5 %	
p-Dichlorobenzene	≥ 99.5 %	
Naphthalene	≥ 99.5 %	
1,2-Dibromo-3-chloropropane	≥ 98.5 %	
p-Nitrochlorobenzene	≥ 99.5 %	
Azobenzene	≥ 98.5 %	
Chloropicrin	≥ 99 %	Sigma-Aldrich Chemie GmbH (Munich, Germany)
Ethylene dibromide	≥ 99.6 %	
Chlorobenzene D5	≥ 99 %	

A stock solution of 1 mg mL^{-1} in n-hexane was prepared for each fumigant and the internal standard. The stock solutions were diluted to 10 µg mL^{-1} and 1 µg mL^{-1} (working solutions) with n-hexane. All solutions were stored in a fridge.

Samples and commodities

The method development focused on cereals and dried fruits. The organic raisins and wheat grain samples used for validation experiments were purchased at a local market and found not to contain any measureable residues of the compounds included in this study. The samples tested for residues of fumigants (see Table 6) were all sampled from local markets. After arriving at the lab they were stored at room temperature and were tested immediately after opening their packages.

Apparatus

The automatic shaking machine Geno Grinder 2010 (SPEX Sample Prep, Metuchen, USA) was used for automated extraction. The centrifuge Rotanta 460 by Hettich (Tuttlingen, Germany) was appropriate for the centrifuge tubes employed and was capable of achieving 4000 rpm. Electronic pipettes applicable for volumes of 10 – 100 µL and 100 – 1000 µL, respectively and manual pipettes applicable for volumes of 1 – 10 mL were from Eppendorf (Hamburg, Germany).

An analytical balance capable of weighing substances from 0.01 g to 205 g was from Mettler-Toledo (Greifensee, Switzerland) and had a minimum indication of 0.1 mg.

A volumetric pipette (10 mL; DIN B Ex 20 °C; Hirschmann Laborgeräte, Eberstadt, Germany) was used for preparation and dilution of the stock and working solutions.

The 50 mL PP (114 × 28 mm) single-use centrifuge tubes with screw caps used for sample extraction were from Sarstedt (Nümbrecht, Germany). The 1.5 mL GC autosampler vials were from Klaus Ziemer GmbH (Langerwehe, Germany). The 6 mL single-use syringes from Henke Sass Wolf (Tuttlingen, Germany) and disposable polyester syringe filters (0.45 µm pore size, 15 mm diameter) from Machery-Nagel (Düren, Germany) were used to filter the fumigant extracts.

A ThermoScientific Trace 1310 GC system (ThermoScientific, Waltham, USA) combined with the mass spectrometer ThermoScientific TSQ 8000 (ThermoScientific, Waltham, USA), run in EI positive mode was used for the analysis of the fumigant extracts. The GC system was connected to a TriPlus RSH autosampler (ThermoScientific, Waltham, USA).

For GC-MS/MS analysis the samples were injected onto a 30 m, 0.20 mm, 1.12 µm Agilent HP VOC column (Agilent, Waldbronn, Germany) equipped with a 10 m, 0.25 mm, deactivated Fused-Silica pre-column (Agilent, Waldbronn, Germany).

Sample Extraction

The samples are directly used for analysis, without any milling or addition of water. 5 g ± 0.1 g of the sample material is weighed into a 50 mL centrifuge tube. Then 5 mL of n-hexane is added followed by 50 µL of the internal standard working solution (10 µg mL^{-1} chlorobenzene-D5). The tube is closed and shaken by a mechanical shaker for one minute. Afterwards the tube is centrifuged for 5 min at 4000 rpm. If necessary, the extract is filtered through a syringe filter (0.45 µm) into a 50 mL tube. Finally, 1 mL of the extract is transferred into a vial for measurement. In case subsampling variability of analytical portions is expected or shown to be a problem, the method can be scaled up 2–4-fold.

MS/MS Measurement conditions used

The extracts were measured by GC-MS/MS using a split-mode injection with a split ratio of 1:5 (split flow: 5 mL min^{-1}). The temperature program of the injector is shown in Table 3 and Figure 1. The initial injection temperature is set at 120 °C and the injection volume was 2 µL.

Table 3 Temperature program of the injector

	Rate (°C s^{-1})	Temperature (°C)	Time (min)
Injection		120	0.1
Transfer	14	250	5
Cleaning	10	300	10

The helium carrier gas, had a constant flow rate of 1 mL min^{-1}. An oven temperature gradient program was applied, starting at a temperature of 45 °C, which was held for 2 min. The temperature was then gradually increased at a rate of 12 °C min^{-1} to 80 °C and held for 5 min. From there the temperature was first slowly increased at 8 °C min^{-1} to 200 °C and then faster at 50 °C min^{-1} to 260 °C. The temperature program of the oven is shown in Table 4 and Figure 1B.

Table 4 Temperature program of the oven

Ramp	Rate (°C min^{-1})	Temperature (°C)	Hold Time (min)
Initial		45	2
1	12	80	5
2	8	200	0
3	50	260	-

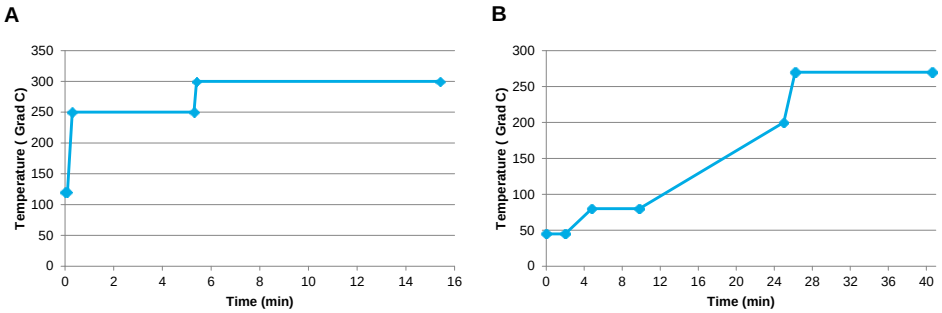

Figure 1 A: Injector temperature program and **B:** Oven temperature program

EI ionization in positive mode was employed. The MS/MS detection was performed in the selected reaction monitoring (SRM) mode. The transfer line was kept at 350 °C and the source temperature was set at 280 °C. The mass transitions for each compound are shown in Table 7.

Method validation

The method was validated at 0.01 mg kg^{-1} and 0.1 mg kg^{-1} (n = 5 each) using blank wheat grain and raisins. The blank samples were spiked with 50 µL of the appropriate fumigant working solution in hexane. Matrix-matched calibration standards at concentrations representing 60 % and 120 % of the validation levels for each matrix were employed. Quantification was carried out by calculating the ratio between analyte peak area and internal standard peak area for both, calibration standards and the extracts of the recovery experiment. The concentration of the fumigants in the extracts of the recovery experiments were then calculated using the calibration curve. Further results of on-going validations can be extracted online from the EURL-DataPool, a database jointly run by the EU Reference Laboratories for residues of pesticides [31].

Results and Discussion

Selection of extraction solvent

Initially the intention was to use isooctane, the solvent used in the method for carbon disulfide analysis following cleavage with HCl/$SnCl_2$ [32]. However, as isooctane interfered with some of the most volatile compounds, it

was decided to switch to n-hexane. In his publication JL Daft also describes the choice of n-hexane as a suitable solvent for multiple fumigants [33]. n-Hexane was also used in methods employing co-distillation [34]. Results for the stability of the stock solutions and mixed working solutions are not yet available, but are in progress. Daft demonstrated in his publication, that the fumigants influence each other.

Chromatographic behavior

In GC-MS/MS analysis we used a special column for volatile compounds (Agilent HP VOC) for chromatographic separation. This column is 30 m long, has a film thickness of 1.12 µm and contains a special coating, the composition of which was not disclosed by the manufacturer. All fumigants showed well-shaped peaks (see Figure 2) and well-repeatable retention times both in pure n-hexane and in extracts.

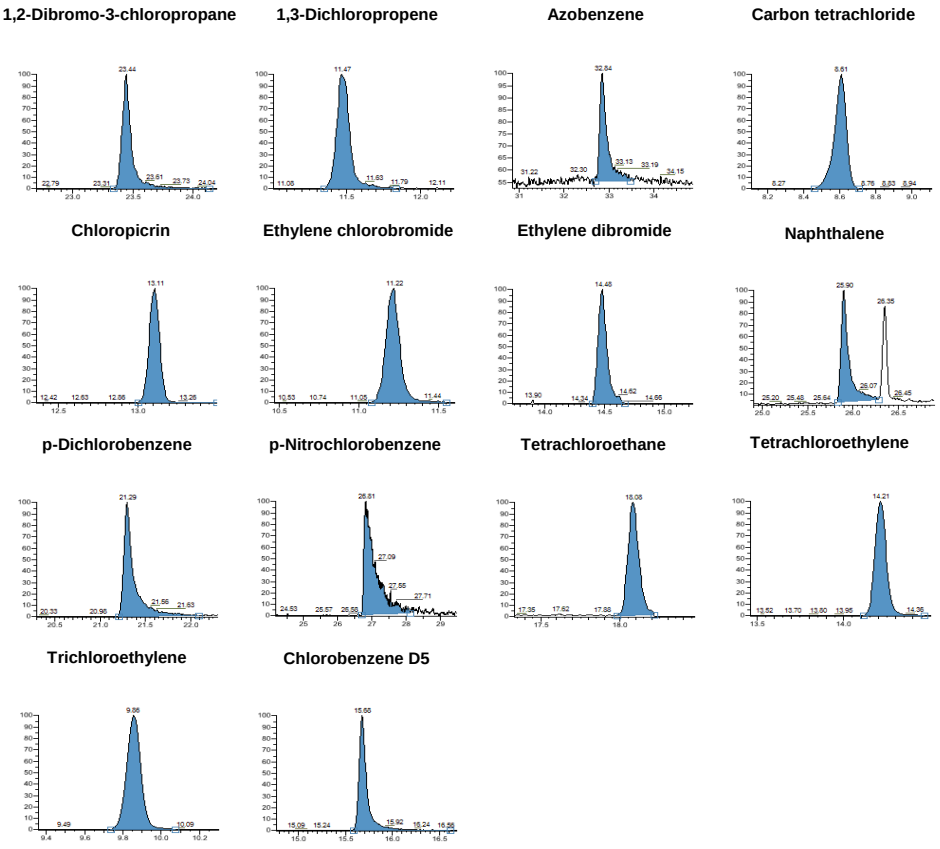

Figure 2 Chromatograms of fumigants derived from extracts obtained from wheat extract spiked at 0.12 mg kg^{-1}

Volatility of the analytes

When working with volatile solvents or compounds it is necessary to check if there are losses of solvent or analytes at different stages of the analytical procedure. As discussed in previous studies there are some important facts to consider [33].

In our experiments, the samples (wheat, raisin) were weighed directly for the analysis of fumigants. Any losses that would have occurred during comminution were thus avoided. The related aspects of homogeneity and extractability will be studied at a later stage using samples with incurred residues (if available) or using samples that are spiked in the lab and aged for a certain period of time.

For the analysis of fresh fruit and vegetables, further tests will have to be conducted in order to avoid losses of fumigant during comminution, as discussed by Daft [33]. If a vial is stored at room temperature for some time before being measured, a volatilization of solvents may occur. In an experiment we checked how much n-hexane escapes from punctured vials within 4 days compared to an unpunctured vial (see Figure 3). The results from the 5-fold determination are shown in the following graph. After 4 days, the weight of the punctured vials was about 3 % lower compared to the unpunctered vial. In further studies we will continue stability experiments with the substances.

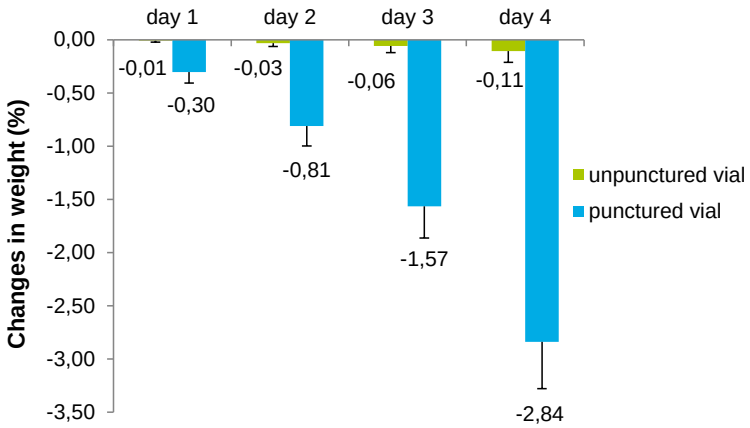

Figure 3 Losses of n-hexane in punctured vials within 4 days in comparison to unpunctured vials

Linearity of detection and matrix effects

For the determination of linearity a fumigant mixture was spiked on aliquots of a blank matrix extract at different levels (n = 3, triplicate determination). Figure 2 shows typical chromatograms of fumigants (0.12 mg kg^{-1}) and the internal standard chlorobenzene-D5 (0.1 µg mL^{-1}) in an n-hexane extract from wheat.

The majority of fumigants showed a linear detection range at concentrations corresponding to spiking levels between 0.005 mg $kg^{-1}$and 2 mg kg^{-1}. In our experiments, the slopes of the calibration curves in presence of matrix were in most cases significantly lower compared to those obtained from standards in pure solvent (see Figure 4). An exception was trichloroethylene. Azobenzene showed the most pronounced matrix-induced suppression effects with its signal declining by ca. 50 % in the presence of matrix.

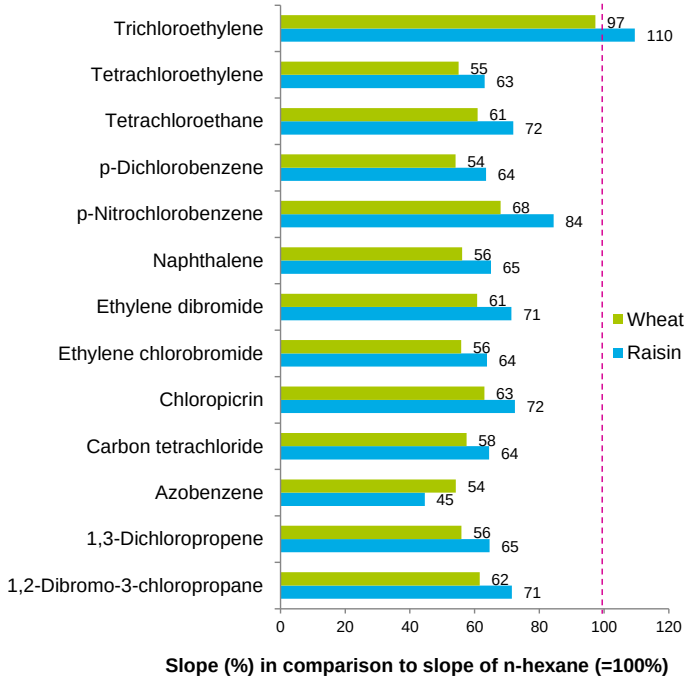

Figure 4 Matrix effects. The graph shows the slope of the calibration curve obtained from solutions in pure solvent compared to those obtained from equally concentrated solutions in extracts of wheat and raisins

An overview of the limits of quantification (LOQs) and the minimum linear ranges achieved for the tested substances can be found in Table 5. The

LOQs were roughly determined based on a signal-to-noise ratio of at least 10. In most cases, spiked pure solvents showed less notable interferences resulting in lower LOQs compared to the spiked matrix extracts. Nevertheless the matrix had a positive impact on the peak shapes. Thus, despite the suppression effect by the matrix the LOQs in n-hexane and extracts were still comparable.

Table 5 Approximate LOQs and linear ranges of the fumigants in solvent and extracts of wheat and raisins

Fumigant	Matrix	LOQ (mg kg^{-1})	Linear Range (µg mL^{-1})
1,2-Dibromo-3-chloropropane	n-hexane	0.005	0.005–1.5
	Raisins		0.005–2
	Wheat		
1,3-Dichloropropene	n-hexane	0.01	0.01–2
	Raisins	0.05	0.05–2
	Wheat		
Azobenzene	n-hexane	0.01	0.01–2
	Raisins	0.05	0.05–2
	Wheat		
Carbon tetrachloride	n-hexane	0.001	0.001–2
	Raisins	0.005	0.005–2
	Wheat		
Chloropicrin	n-hexane	0.005	0.005–2
	Raisins		
	Wheat		
Ethylene chlorobromide	n-hexane	0.05	0.05–1.5
	Raisins		0.05–2
	Wheat		
Ethylene dibromide	n-hexane	0.01	0.01–2
	Raisins	0.05	0.05–2
	Wheat		
Naphthalene	n-hexane	0.01	0.01–1.5
	Raisins		0.01–2
	Wheat		
p-Nitrochlorobenzene	n-hexane	0.05	0.05–2
	Raisins		
	Wheat		
p-Dichlorobenzene	n-hexane	0.005	0.005–2
	Raisins		
	Wheat		
Tetrachloroethane	n-hexane	0.005	0.005–2
	Raisins	0.01	0.01–2
	Wheat		

Fumigant	Matrix	LOQ (mg kg^{-1})	Linear Range (µg mL^{-1})
Tetrachloroethylene	n-hexane	0.001	0.001–2
	Raisins		
	Wheat		
Trichloroethylene	n-hexane	0.005	0.005–2
	Raisins		
	Wheat		

In raisins a peak from the matrix interfered the signal of azobenzene (see Figure 5). Despite its generally good detection sensitivity, the LOQ of azobenzene is therefore higher than that of the other compounds. In further analyses we found a similar interference in dried apricots. The lowest successfully validated level was at 0.01 mg kg^{-1} in case of 1,2-dibromo-3-chloropropane, carbon tetrachloride, chloropicrin, ethylene dibromide, naphthalene, p-dichlorbenzene, tetrachlorethane, tetrachlorethylene and trichlorethylene and at 0.05 mg kg^{-1} in all other cases. These are considered as the reporting limits (RL).

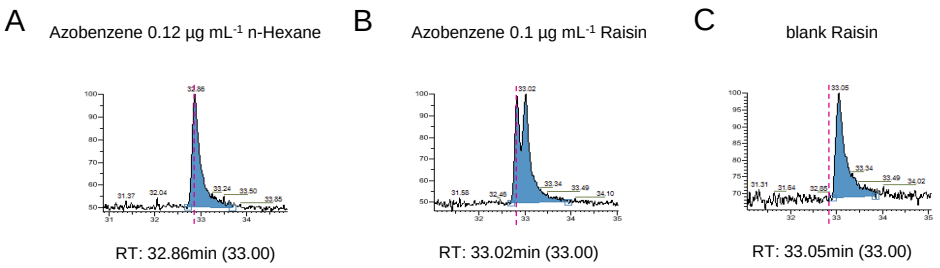

Figure 5 Chromatographic interferences in the case of azobenzene. The peaks obtained in the mz 105 → 77.100 mass transition trace of azobenzene when injecting a) calibration solution at 0.12 µg mL^{-1} in pure solvent, b) a calibration solution of azobenzene in blank raisin extract corresponding to 0.10 µg mL^{-1} and c) azobenzene blank raisin extract

Method validation

The method was validated on raisins (commodity with high sugar and low water content and wheat (high starch content and low water and fat content). Blank samples were spiked with the fumigant mixture at 0.01 mg kg^{-1} or 0.1 mg kg^{-1} (n = 5 each), extracted and analyzed as described above. All blank samples were proven not to contain fumigants in relevant amounts right before validation. To eliminate matrix effects, matrix-

matched calibration standards were employed. The use of an internal standard (chlorobenzene-D5) further reduced the influence of volume fluctuations and to some extent of evaporations and suppressions. Two calibration levels were prepared at concentrations representing 60 % and 120 % of the respective spiking level. Satisfying recoveries and variabilities were achieved for all compounds. For wheat, the average recoveries were between 79 % and 106 % at both validation levels and the RSDs were between 1.0 % and 10 %. For raisins, the average recoveries were between 86 % and 109 % at both validation levels and the RSDs were between 1 % and 9.9 %.

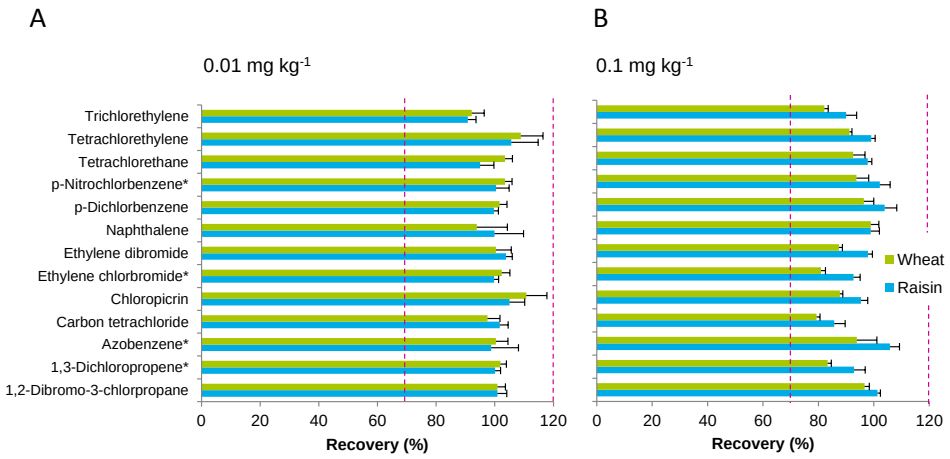

Figure 6 Recoveries achieved in the validation experiments on wheat grains and raisins, 0.1 mg kg^{-1} and 0.01 mg kg^{-1}, except substances marked with * = 0.05 mg kg^{-1}

The next figure shows the validation data, calculated either with or without the use of an internal standard. The results show that the use of an internal standard compensates losses in the extraction and/or injection step. This is especially evident at low concentrations. In the case of naphthalene the absolute recoveries obtained in raisin using solvent based calibration were at 193 % ± 17 % without ISTD and at 94 % ± 10 % with ISTD. This demonstrates the positive impact of the ISTD in this procedure.

Please note that the present work mainly focused on the measurement step. The extraction conditions (e.g. time, temperature) are not yet properly optimized due to the lack of samples with incurred and aged residues of fumigants [22]. Such experiments are planned for the future and may lead to an alteration of the extraction conditions to optimize the yields.

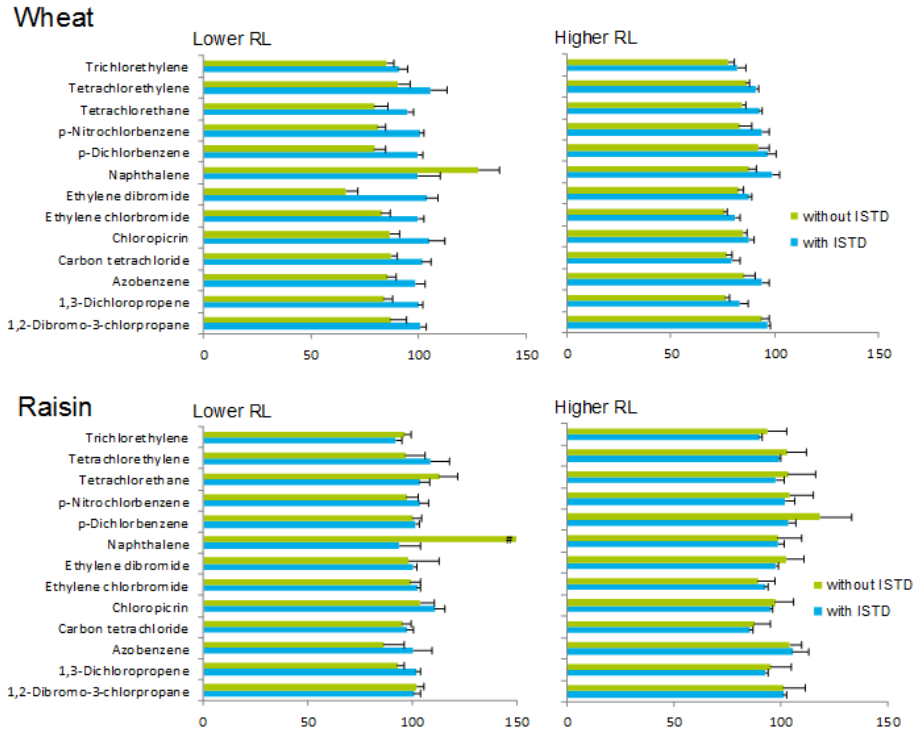

Figure 7 Comparison of the validation data with and without the use of an ISTD

Analysis of real samples

To start with 12 dry samples were analyzed for the fumigants included in this study. No fumigant residues could be detected in any of these samples (see Table 6). The investigations will continue.

Table 6 Fumigant analyses in different commodities and country of origin

Commodity	Country of origin	Fumigant levels in mg kg^{-1}
Candied ginger	Thailand	< 0.01
Dates	Tunisia (2x)	
Dates	unknown	
Dried apricots	unknown	
Dried apricots	Turkey	
Dried cranberries	unknown	
Dried figs	Turkey	
Dried papaya	unknown	
Dried physalis	South America	
Dried pineapple	Ghana	
Dried mango	Peru	
Raisins	unknown	

Conclusions and outlook

Our study demonstrates that several fumigants can be analyzed simultaneously applying extraction with n-hexane and determinative analysis by GC-MS/MS. Very satisfying recoveries and RSDs were achieved using this method on spiked wheat grain and raisins. Chlorobenzene-D5 was used as internal standard. The analysis of further samples from the market is in progress. Following treatment of commodities in the laboratory, it is also planned to investigate how to further optimize extraction yields of aged residues. The impact of comminution on the residue levels will be studies. Our further plans include the adaptation of the method for other types of commodities including fresh fruits (kiwi, bananas, …). In this case an up-scaling of the procedure would be advantageous to reduce sub-sampling variability. The impact of salt-addition during the extraction step should be investigated here. It is also desirable to routinely check fruit and vegetable samples for the presence of fumigants.

Appendix:

Table 7 Mass transitions of compounds

Compounds	Retention Time (min)	Precursor Mass (u)	Product Mass (u)	Collision Energy (eV)
1,2-Dibromo-3-chloropropane	22	154.9	75	5
		154.9	92.9	25
		156.8	75	5
		234	155	5
1,3-Dichloropropene	12	110	75	5
		111.9	77.2	5
Azobenzene	29.7	105	77.1	5
		182.1	105.1	5
Carbon tetrachloride	8.2	116.9	81.9	28
		118.9	83.9	28
		120.9	83.9	28
Ethylene chlorobromide	10.6	65	65	0
		143.9	63	5
		63	63	0
Ethylene-dibromide	13.8	187.8	107	5
		106.9	106.9	0
		108.9	108.9	0
Naphthalene	24.4	127.9	77.7	20
		128	128	0
p-Nitrochlorobenzene	25.5	111.1	75.1	10
		156.9	99	15
p-Dichlorobenzene	20	111	75.1	10
		112.8	75	15
		146	111	15
		147.9	113	15
Tetrachloroethane	17.2	132.6	97	15
		165.8	83	5
		82.8	82.8	0
Tetrachloroethylene	13.5	128.9	93.9	20
		130.9	95.9	15
		163.9	128.9	15
		165.8	130.9	10
Trichloroethylene	9.3	94.9	60	25
		129.9	95	10
		131.8	96.9	10
Chlorobenzene-D5	15.8	82	54	15
		117	82.1	15
		118.8	82.1	15

Table 8 Recovery validation for raisins

Raisin														
Compound	**Lower RL**							**Higher RL**						
	Recovery (%)					Average (%)	RSD (%)	Recovery (%)					Average (%)	RSD (%)
1,2-Dibromo-3-chloropropane	101	101	99	106	97	101	3,2	103	98	102	102	101	101	1,7
1,3-Dichloropropene*	105	102	100	102	101	102	1,8	95	93	92	92	93	93	1,3
Azobenzene*	115	102	94	100	91	100	9,3	94	104	111	112	108	106	7,3
Carbon tetrachloride	100	98	93	101	96	98	2,9	86	85	85	85	88	86	1,2
Chloropicrin	119	110	109	112	104	111	5,2	96	95	94	95	97	95	1,1
Ethylene chlorobromide*	105	103	102	102	101	102	1,5	92	91	95	92	93	93	1,6
Ethylene-dibromide	97	102	102		101	100	2,2	96	97	99	98	99	98	1,3
Naphthalene	83	105	97	100	84	94	9,9	102	97	101	99	95	99	2,9
p-Nitrochlorobenzene*	102	103	100	100	103	102	1,5	107	99	101	106	107	104	3,6
p-Dichlorobenzene	108	107	100	105	98	103	4,4	108	104	102	98	98	102	4,4
1,1,2,2-Tetrachloroethane	111	100	106	99	102	104	4,7	103	92	98	95	101	98	4,2
Tetrachloroethylene	94	110	114	108	118	109	9,2	100	98	98	100	100	99	1,0
Trichloroethylene	93	90	90	97	91	92	2,8	91	90	88	90	91	90	1,4

Table 9 Recovery validation for wheat

Wheat														
Compound	**Lower RL**							**Higher RL**						
	Recovery (%)					Average (%)	RSD (%)	Recovery (%)					Average (%)	RSD (%)
1,2-Dibromo-3-chloropropane	100	104	101	102	97	101	2,7	97	96	98	95	98	97	1,0
1,3-Dichloropropene*	102	97	102	100	100	100	2,1	79	83	83	90	82	83	4,0
Azobenzene*	104	94	102	97	97	99	4,1	97	95	97	89	92	94	3,5
Carbon tetrachloride	105	94	104	102	104	102	4,3	77	81	79	85	75	79	3,9
Chloropicrin	99	100	116	102	109	105	7,0	87	87	89	91	85	88	2,5
Ethylene chlorobromide*	98	98	105	100	99	100	2,8	82	78	80	85	80	81	2,4
Ethylene-dibromide	113	100	103	102	101	104	5,3	85	87	89	87	89	87	1,6
Naphthalene	114	93	88	97	107	100	10,5	94	103	98	101	99	99	3,2
p-Nitrochlorobenzene*	101	99	101	101	95	100	2,5	97	99	101	90	95	96	4,3
p-Dichlorobenzene	102	97	103	101	100	100	2,4	90	98	92	91	97	94	3,7
1,1,2,2-Tetrachloroethane	93	97	96	97	92	95	2,5	91	91	93	94	94	93	1,5
Tetrachloroethylene	102	99	113	115	100	106	7,6	90	91	91	93	91	91	1,4
Trichloroethylene	88	87	98	91	90	91	4,2	79	84	82	87	79	82	3,8

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