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Residues of plant protection product co-formulants in food

Sub-project I

Substance selection and method development

Project report on behalf of the FSVO
Ulrich Schaller and Marianne Balmer

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Authors:

Ulrich Schaller and Marianne Balmer

Agroscope
Plants and Plant Products Competence Division
Plant Protection Chemistry
Müller-Thurgau-Strasse 29
CH-8820 Wädenswil

Sponsor

Federal Food Safety and Veterinary Office (FSVO)
Risk Assessment Department
Toxicology and Biology
Schwarzenburgstrasse 155
CH-3003 Bern

Contact: Christoph Geiser

Photo on title page:
peppers and apples after treatment with a plant protection product (U. Schaller, Agroscope)

Contents

1 Executive summary

In this sub-project, analytical methods were developed for selected co-formulants of plant protection products so that their residues in and/or on foods of plant origin can be determined. Various co-formulants from the class of solvents, namely N,N-dimethyldecanamide (DMDA), cyclohexanone and components of solvent naphtha (including 2-methylnaphthalene), as well as the surfactant dioctyl sulfosuccinate (docusate), can be quantified specifically and sensitively with LC-MS/MS or GC-MS on apples and a variety of vegetables.

In laboratory studies, apples, peppers and tomatoes were treated with selected plant protection products and stored for up to 3 days before the co-formulant residues were determined as a function of time. In these laboratory tests, residues of all four co-formulants were detected at levels greater than 0.01 mg/kg. The residues of the relatively volatile co-formulants 2-methylnaphthalene and cyclohexanone were low from the outset and decreased very rapidly. The residues of dimethyldecanamide and dioctyl sulfosuccinate decreased much more slowly, or hardly at all, and were still readily measureable after three days. The data shows that both the initial amount of residue and the rate of its decline depend significantly on the volatility of the substance.

Both the solvent naphtha components and cyclohexanone were also found in small quantities in blank and untreated control samples. This shows that plant protection product co-formulants, which are often also used in a wide range of everyday consumer products and industrial processes, are ubiquitous and their source cannot always be easily identified. The co-formulant dioctyl sulfosuccinate was found on several types of vegetable purchased in retail trade (a major Swiss distributor) at concentrations in the range from 0.002 to 0.1 mg/kg. It is plausible that these residues originated in the application of plant protection products.

It will only be possible to gain a sense of the actual quantity and the behaviour of co-formulant residues under practical conditions in field tests, which are to be conducted in the next sub-project.

2 Summary

2.1 Background and objectives

The Plant Protection Products Ordinance (PlantPPO)[1] specifies that residues of plant protection products must not produce any harmful effects on human or animal health or on groundwater. A plant protection product (PPP formulation) generally consists of one or more active substances, and co-formulants. In practice, the risk assessment and the regulation of residues are limited exclusively to the active substances in the plant protection products and their metabolites and breakdown or reaction products. Co-formulants of plant protection products are not considered. Since, when using plant protection products, the co-formulants are applied to the crop in the same way as the active substances, it must be assumed that they will also be found on the food products.

Together with future work of a more in-depth nature, this sub-project aims to establish a basis for risk assessment and to enable risks arising from co-formulant residues in food products to be identified and, where necessary, measures to be defined for the protection of consumers. The first step is to gather information on the potential exposure, in other words the type and amount of co-formulant residues on food products.

In this first sub-project, therefore, co-formulants or components thereof are to be selected and examined in order to determine their suitability for further residue investigations. This includes the development of analytical methods for these substances, as well as simple laboratory tests that can provide some indication of the behaviour of the co-formulants on harvested produce. The findings obtained in this way will form the basis for planning further sub-projects, including field tests.

2.2 Literature on co-formulant residues

To date, few scientific studies have been published that look specifically at the incidence and behaviour of plant protection product co-formulants in the environment or on food products. Since most co-formulants are also used in industrial processes, or in industrial and household chemicals, it is not surprising that they are also detected in the environment. This applies, for example, to the large group of surfactants (see e.g. [1] and the studies cited there). Concerning co-formulant residues on food, Adrian et al. [2] present a proposal for assessing their presence on products of plant origin, based on the concept of the Residue Unit Dose (RUD), as proposed by EFSA for assessing the risk posed by active substances of plant protection products to birds and mammals [3]. However, no reference is made either to the effective concentrations of co-formulants in plant protection products or to actually measured residues. Klatyik et al. [1] and Mullin [4] suggest that co-formulants (in particular surfactants) can influence the toxicity of the active substances of plant protection products. Takacs et al. [5] concluded that formulations with active substances from the group of neonicotinoids have a greater toxic effect on daphnia than would be expected based on the toxicological endpoints of the active substances alone. We are not aware of any studies on the actual incidence of co-formulants and their impacts under real environmental conditions. However, all

[1] Ordinance on the Placing on the Market of Plant Protection Products (Plant Protection Products Ordinance, PlantPPO) of 12 May 2010 (version: 1 January 2019)

authors agree that the available data on co-formulants, both with regard to exposure and in terms of toxicological endpoints, is not sufficient to assess the potential risks.

In 2003, the Danish environmental protection authority published a report on selected plant protection product co-formulants [6]. The aim of the report was to compile data on co-formulants and evaluate the toxicological effects. The authors concluded that the availability of data on the 18 selected co-formulants was limited. Nonetheless, the available data showed that co-formulants cannot be considered inert from a toxicological standpoint. This study, however, only looked at the risk and explicitly factored out exposure. One of the substances evaluated was cyclohexanone, which was also examined in this sub-project. However, the authors were unable to draw any clear conclusions about the possible risk posed by cyclohexanone.

An informal survey of authorities in EU Member States and industry representatives also indicated that, at present, there are no studies available or ongoing that look at co-formulants and their presence and behaviour in/on food products or in the environment.

Possible reasons for the low level of research on co-formulant residues to date could include the analytical challenges (see 3.2) or the fact that publicly available data on the composition of plant protection products is limited. In general, the focus tends to be on exposure and on the effects of the biologically active substances, with the co-formulants being studied only rarely.

2.3 Selection of co-formulants

The co-formulants investigated were selected on the basis of the results of the preliminary study for this project [7]. In this study, product compositions and sales figures were analysed in order to identify frequently used co-formulants that could potentially lead to residues on fruits or vegetables. The preliminary study showed that solvents in particular are important co-formulants. Firstly, they make up a significant proportion of some plant protection products (70% on average), namely in emulsion concentrates; and secondly, individual representatives of this category are among the leading co-formulants in terms of quantity. For this reason, several substances were selected from the group of solvents that cover a range of substance properties and differ in particular in terms of volatility.

- N,N-Dimethyldecanamide (DMDA): According to our projections, some 9 tonnes of DMDA were sold in plant protection products in Switzerland in 2015. In comparison with other solvents, this substance is somewhat less volatile, but it can still be considered a volatile substance[2] (also see Table 1). A frequently cited and typical use of DMDA is its inclusion in plant protection product formulations. This substance is also used in detergents and cleaning agents, polishes and waxes, both in industrial processes and in consumer products[3]. According to the ECHA, the quantity of DMDA that is produced and imported in Europe is in the range from 10,000 to 100,000 tonnes per year.
- Components of solvent naphtha: 2-Methylnaphthalene, 1-methylnaphthalene and biphenyl: Solvent naphtha is one of the most widely used co-formulants (with a projected use in plant protection products of >50 tonnes). It is an umbrella term that is used for various crude oil

[2] According to the classification of vapour pressures in FOCUS Air [8]
[3] ECHA dossier submission on N,N-dimethyldecan-1-amide: https://echa.europa.eu/de/registration-dossier/-/registered-dossier/15021/1

fractions, including petroleum (heavy aromatic) and petroleum (light aromatic), in other words mixtures of different hydrocarbons. Solvent naphtha can be found in a wide range of products for end consumers. The heavy aromatic fractions, for instance, are used in lubricants and sealants, greases, propellants and antifreeze[4]. In Europe, 0.1 to 1 million tonnes of solvent naphtha (heavy aromatic) and 1 to 10 million tonnes of the lighter fraction (solvent naphtha light aromatic) are produced or imported every year (according to figures from ECHA).

Individual components of solvent naphtha, namely 2-methylnaphthalene and 1-methylnaphthalene, were selected for the studies. According to our measurements, they make up approx. 8 to 13% and 4 to 6% respectively in the mixture. Another component studied was biphenyl, which is generally present in a proportion of less than 1%.

Solvent naphtha is an example of a co-formulant which itself is an imprecisely defined mixture and whose components are ubiquitous due to their broad and diverse applications.

- Cyclohexanone is another solvent commonly used in plant protection product formulations, with a projected usage of some 20 tonnes (for Switzerland). Compared to DMDA and the solvent naphtha components studied, cyclohexanone is slightly volatile. It is used widely in everyday products, such as inks and toners, paints, cleaning agents and disinfectants[5]. The ECHA puts the production and import quantity for Europe at 1 to 10 million tonnes a year.

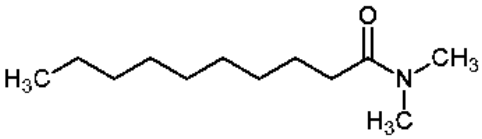
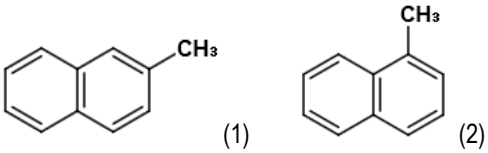
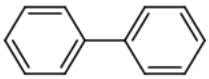
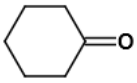
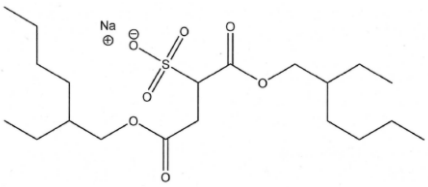
As wetting agents, surfactants play an important role and are the second major class of plant protection product co-formulants after solvents. There are particular challenges associated with the analysis of surfactants, since they tend to be a mixture of substances. However, dioctyl sulfosuccinate is a representative of this category that is used as a single substance and can be readily analysed.

- Dioctyl sulfosuccinate sodium salt (docusate): The quantity of dioctyl sulfosuccinate sold as a plant protection product co-formulant in Switzerland is comparatively low, coming in at 1 tonne for 2016 according to our projections. However, we believe that the compound is a suitable representative of the category of anionic surfactants for further studies. Docusate is also used in cleaning agents, modelling clay and finger paint, as well as fertilisers[6]. According to ECHA, >10,000 tonnes of this substance are produced and imported in Europe every year.

The co-formulant propylene glycol was also considered for further studies. Propylene glycol is included in a large number of liquid formulations as antifreeze. According to projections, 27 tonnes of propylene glycol were sold in plant protection products in Switzerland in 2015. However, owing to its high volatility and low molar mass, it has not been possible to develop robust and sensitive analytical methods for this compound. The compound was not studied any further in this project.

[4] ECHA substance information for solvent naphtha (petroleum), heavy arom. https://echa.europa.eu/de/substance-information/-/substanceinfo/100.059.253
[5] ECHA substance information for cyclohexanone: https://echa.europa.eu/de/substance-information/-/substanceinfo/100.003.302
[6] ECHA substance information for docusate sodium: https://echa.europa.eu/de/substance-information/-/substanceinfo/100.008.553

Table 1: Chemical structure and selected physicochemical properties of the co-formulants studied.

Co-formulant	**Substance properties** [a]			
Name and CAS RN Chemical structure	Molar mass [g/mol]	Partition coeff. octanol/water log POW	Water solubility at 20 °C [mg/L]	Vapour pressure at 25 °C [Pa]
N,N-Dimethyldecanamide (DMDA) CAS RN: 14433-76-2 	199.3	3.44	340	0.11
Components of solvent naphtha: (1) 2-Methylnaphthalene CAS RN: 91-57-6 (2) 1-Methylnaphtahlene CAS RN: 90-12-0 (3) Biphenyl CAS RN: 92-52-4 	(1) 142.2 (2) 142.2	3.86 3.87	24.6 (25 °C) 25.8 (25 °C)	7.3 8.9
	(3) 154.2	4.01	7.5 (25 °C)	1.19
Cyclohexanone CAS RN: 108-94-1 	98.2	0.86	8.6×10^4	7×10^2
Dioctyl sulfosuccinate sodium salt (docusate) CAS RN: 577-11-7 	444.6	2.00	8.2×10^3	1.6×10^{-12}

[a] Information for DMDA, cyclohexanone and docusate according to the applicable ECHA registration dossier
Information for methylnaphthalene and biphenyl according to the US National Library of Medicine

More details on the co-formulants studied can be found in section 4.

2.4 Analytical methods for residue determination

The plant samples were prepared using the QuEChERS technique [10], an established method for the analysis of pesticide residues in food products. According to this method, the samples are frozen and ground into a powder, before being extracted with solvent (acetonitrile or ethyl acetate) (extract I). The extract is usually then purified by means of solid/liquid separation (extract II, Fig. 1). In order to verify and quantity the residues, gas chromatography or liquid chromatography was used, depending on the substance properties, coupled with (tandem) mass spectrometry (GC-MS/MS or LC-MS/MS). The table below provides an overview of the performance of the methods. See sections 4 and 6 for details of the analytics.

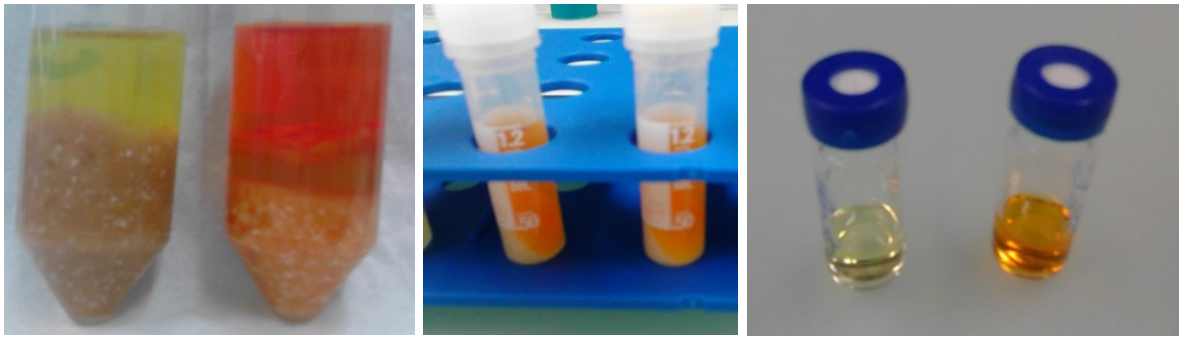


Figure 1: Extracts of apple and pepper (left, extract I), purification with QuEChERS tube II (centre, extract II), and extracts in the HPLC vial ready for measurement (right).

Table 2: Overview of the methods developed for the analysis of residues in products of plant origin

Co-formulant	QuEChERS sample preparation	Analysis	Matrices	Instrument limit of quantification (mg/kg)	Recovery (%)
N,N-Dimethyl-decanamide	Acetonitrile / Extract II	LC-MS/MS m/z 200.2→ m/z 102.2	Apples, peppers	0.002 (for standard)	102 (apples) 99 (peppers)
2-Methylnaph-thalene	Ethyl acetate / Extract II	GC-MS m/z 142.1	Apples, tomatoes	0.002 (for standard)*	103 (apples) 93 (tomatoes)
1-Methylnaph-thalene	Ethyl acetate / Extract II	GC-MS m/z 142.1	Apples, tomatoes	0.002 (for standard)*	108 (apples) 97 (tomatoes)
Biphenyl	Ethyl acetate / Extract II	GC-MS m/z 154.1	Apples, tomatoes	0.002 (for standard)	104 (apples) 92 (tomatoes)
Cyclohexa-none	Ethyl ace-tate/Extract I	GC-MS m/z 98.0	Apples	0.002 (for standard)*	97 (apples)
Dioctyl sulfo-succinate	Acetonitrile/ Extract I	LC-MS/MS m/z 423.5→ m/z 199.1	Apples, aubergi-nes, cucumbers, peppers, toma-toes, courgettes	0.002 (for standard)	91-100 (Apples and misc. vegetables)

* The actual limits of quantification in the samples were higher for these compounds and were determined using the blank values.

For all co-formulants, specific verification methods were developed in at least one plant matrix. The limits of quantification were well below 0.01 mg/kg, the legally prescribed minimum limit of quantification for plant protection product active substances; the recovery rates were almost 100% for all analytes in the matrices examined.

Cyclohexanone and both methylnaphthalenes were also confirmed in blank samples at concentrations of above 0.002 mg/kg, namely at concentrations of around 0.02 mg/kg for cyclohexanone and around 0.006 and 0.003 mg/kg for 2-methylnaphthalene and 1-methylnaphthalene respectively. As a result, the actual limit of quantification in the samples increased significantly but was not determined precisely. These substances are used very widely and in a diverse array of applications, and are therefore present in virtually every environment. This is reflected in the elevated blank values.

2.5 Laboratory tests

2.5.1 Conducting the laboratory tests

Simple laboratory tests were conducted to determine whether the selected co-formulants could potentially lead to verifiable residues when fruits or vegetables are treated with plant protection products containing these co-formulants.

The treated products were apples, peppers and tomatoes. These products were selected because they are available all year round, are easy to handle in the laboratory, and are relatively unproblematic for analysis (few interfering signals, little matrix in the extract and correspondingly lower limits of quantification).

For the treatment step, selected plant protection products were diluted with water so that the concentration corresponded to those found in the spray mixture according to the respective

product authorisation. Plant protection products were used that had a sufficiently high content of one of the co-formulants being investigated. Where possible, the products were also authorised for use in at least one of the cultures studied.

Table 3: Overview of the plant protection products used in the laboratory tests

Trade name (Formulation) Function	Fed. reg. no.	(Selected) cultures or culture group with authorisation	**Active substance** in the PPP	Declared content	**Co-formulant** in the PPP	Measured content% (w/w)
Input (EC) Fungicide	W-6392	Cereals	Spiroxamine	300 g/L	DMDA	38.6%
Slick (EC) Fungicide	W-5056	Misc. berries, fruit, vegetables, potatoes, rapeseed, wheat, ornamental plants	Difenoconazole	250 g/L	Solvent naphtha 2-Methylnaphthalene 1-Methylnaphtahlene Biphenyl	 6.2% 3.1% 0.16%
Milbeknock (EC) Insecticide	W-6526	Berries, apples/pears, ornamental plants	Milbemectin	9.3 g/L	Cyclohexanone	20%
Armicarb (SP) Fungicide	W-6432	Misc. berries, fruit, vegetables, ornamental plants	Potassium bicarbonate	850 g/kg	Docusate	8.9%

In order to simulate treatment, the apples, peppers or tomatoes were immersed in the “spray mixture” and then stored in an open tray. Shortly after treatment, once the spray mixture had dried on the produce, one sample per food item was taken (1 hour after treatment, or 10 minutes in the case of cyclohexanone). Further samples were taken 24 and 72 hours after treatment.

For the co-formulant DMDA and the components of solvent naphtha, the influence of varying environmental conditions (light, heat, air circulation) was also investigated. For each of these co-formulants, a portion of the treated fruits/vegetables was stored in open trays outdoors (on the roof).

Table 4: Overview of the laboratory tests conducted

Product	Product in the spray mixture	Co-formulant(s)	Co-formulant in the spray mixture	Products tested	Tests
Input	0.1% (w/v)	DMDA	386 mg/L	Apple	Laboratory: 1, 24, 72 hours Roof: 24, 72 hours
		(Spiroxamine) [a]	306 mg/L	Pepper	Laboratory: 1, 24, 72 hours Roof: 24, 72 hours
Slick	0.05% (w/v)	2-Methylnaphtha-lene	31 mg/L	Apple	Laboratory: 1, 24, 72 hours Roof: 24, 72 hours
		1-Methylnaphtah-lene	16 mg/L	Tomato	Laboratory: 1, 24, 72 hours Roof: 24, 72 hours
		Biphenyl	0.8 mg/L		
Milbeknock	0.125% (w/v)	Cyclohexanone	250 mg/L	Apple	Laboratory: 10 min, 1, 24, 72 h
Armicarb	0.3% (w/v)	Docusate	267 mg/kg	Apple	Laboratory: 1 hour
				Pepper	Laboratory: 1, 24, 72 hours

* Spiroxamine, active ingredient in the product Input, was also analysed in these tests

See section 4 for more details of the test procedure.

2.5.2 Evaluation of the laboratory tests

Dimethyldecanamide (DMDA)

In the tests with the product Input, the active substance spiroxamine was analysed in addition to the co-formulant DMDA. In both types of produce investigated (peppers and apples), the concentration of DMDA decreased significantly both when stored in the laboratory and outdoors. In contrast, spiroxamine decreased only slowly (apples) or not at all (peppers) during the test period. The more rapid decrease in DMDA residues can primarily be attributed to a much higher vapour pressure.

The type of storage had a low influence. For DMDA, however, a weak trend towards a more rapid decrease outdoors was observed, where the samples were exposed to significantly higher temperatures[7].

[7] At the neighbouring weather station, a mean temperature of 25 °C and a maximum temperature of 31 °C were recorded during the test period; however, the temperatures at the storage site on the roof would have been higher overall. In comparison, the samples in the laboratory were stored at around 20 °C.

Table 5: Residues of the co-formulant DMDA and the active substance spiroxamine on apples and peppers after treatment with a "spray mixture" of the product Input and after storage in the laboratory or outdoors (on the roof).

	DMDA residues (mg/kg)				**Spiroxamine residues (mg/kg)**			
Time (hours)	Peppers Labora-tory	Peppers Roof	Apples Labora-tory	Apples Roof	Peppers Labora-tory	Peppers Roof	Apples Labora-tory	Apples Roof
1	1.29	-	1.39	-	0.52	-	0.78	-
24	0.72	0.66	1.37	0.55	0.49	0.52	1.01	0.57
72	0.46	0.49	0.48	0.25	0.53	0.53	0.67	0.50

The visualisation of the DMDA residues in comparison with the spiroxamine residues in each sample provides an especially clear overview of the comparatively faster decrease in DMDA, since it enables the variability due to the sample treatment or processing to be compensated for (Fig. 2).

At the start of the tests, the ratio of DMDA to spiroxamine was much higher than would be expected based on the (calculated) concentrations of both substances in the spray mixture. A slightly higher octanol/water partition coefficient and the slightly lower water solubility of DMDA compared with spiroxamine may have resulted in greater accumulation of the former on the surface of the produce. However, this assumption would have to be investigated in further tests.

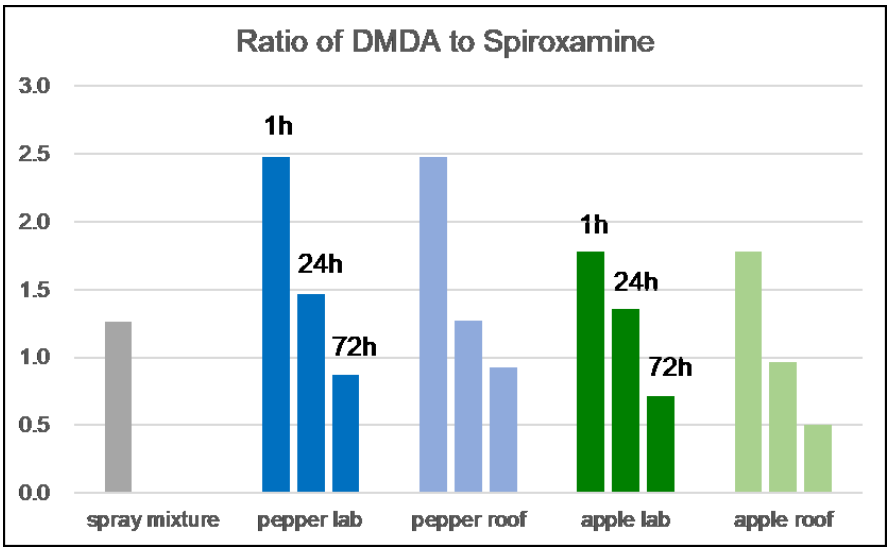


Figure 2: Ratio of the concentrations of the co-formulant DMDA and the active substance spiroxamine in peppers (blue) or apples (green), 1, 24 and 72 hours after treatment in the laboratory. The 24-hour and 72-hour samples were stored in the laboratory or outdoors, in warm and sunny weather on the roof in an open tray.

Components of solvent naphtha and cyclohexanone

The levels of the co-formulants or co-formulant components 2-methylnaphthalene, 1-methylnaphthalene, biphenyl and cyclohexanone were already very low in the samples tested 1 hours or 10 minutes (cyclohexanone) after treatment. All these compounds can be described

as volatile. A rapid decrease in residues after treatment was expected accordingly. The methylnaphthalenes and biphenyl were also already present in the “spray mixture” at low concentrations as individual components of solvent naphtha (see Table 4). After 1 hour, the concentrations of 2-methylnaphthalene and 1-methylnaphthalene were in the range of 0.02 mg/kg and < 0.01 mg/kg, while biphenyl was always below or in the range of the limit of quantification of 0.002 mg/kg. 10 minutes after treatment, the cyclohexanone residues were at 0.06 mg/kg.

Solvent naphtha and cyclohexanone are both widely used substances in everyday products and industrial processes, and are therefore ubiquitous. This is reflected in the fact that the examined co-formulants were found in readily verifiable quantities in untreated control samples and in blank samples. The concentrations in the control and blank samples were on the same order of magnitude as those in the treated products. Laboratory materials are the most prominent possible source for the blank values.

Table 6: Concentrations of cyclohexanone and components of solvent naphtha on various types of harvested produce after treatment with plant protection products in the laboratory, and in blank samples and untreated control samples. See section 4 for further measured values.

	2-Methylnaphthalene		1-Methylnaphthalene		Biphenyl		Cyclohexanone
	Apples (mg/kg)	Tomato (mg/kg)	Apples (mg/kg)	Tomato (mg/kg)	Apples (mg/kg)	Tomato (mg/kg)	Apples (mg/kg)
Blank sample	0.006	0.006	0.003	0.003	< 0.002	< 0.002	0.022
Untreated control	0.006	0.003	0.003	0.002	< 0.002	< 0.002	0.013
10 minutes	-	-	-	-	-	-	0.058
1 hour	0.023	0.017	0.009	0.008	0.002	< 0.002	0.036

Owing to the low concentrations and the blank values, a more detailed evaluation of the results is not appropriate here. However, the tests confirm the following points mentioned above:

- The verification and quantification of co-formulants, which are themselves a mixture of various compounds (such as solvent naphtha), is difficult because individual components have to be examined that may make up only a small proportion of the mixture and therefore have low concentrations (such as 1- and 2-methylnaphthalene and biphenyl).
- In the case of co-formulants that are widely used for other purposes, a background presence is to be expected. It is then virtually impossible to assess whether the concentrations found are from residues of plant protection products or from other sources.
- Minimising blank values in the laboratory can be a very time-consuming task and it must be carefully determined whether this effort is proportionate.

Dioctyl sulfosuccinate (docusate)

After peppers and apples had been treated with the product Armicarb, docusate was measured on the harvested produce in readily verifiable concentrations (approximately 0.3 and

0.5 mg/kg respectively). The residues remained roughly the same for 72 hours in the laboratory. In view of the comparatively high concentration of docusate in the spray mixture and the low vapour pressure of the compound, this was in line with expectations. The individual measured values can be found in section 4.

In blank samples, docusate was not found at concentrations above the limit of quantification (< 0.002 mg/kg). In untreated control samples, however, the surfactant was found in readily verifiable concentrations. We assume that this is from residues of plant protection products (see section 2.6). According to these laboratory tests, docusate appears to be a suitable candidate for investigating the behaviour of surfactants on harvested produce.

Influence of vapour pressure

It can be assumed that the volatility of a co-formulant has a significant impact on the amount of residue after application. On the whole, the comparison of (normalised) residues shortly after application with the respective vapour pressures confirms this.

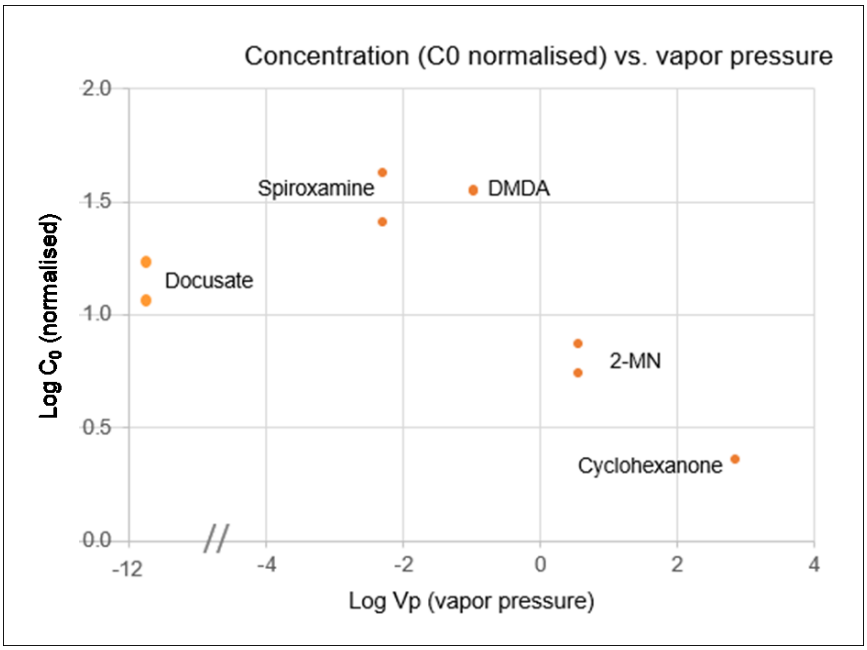


Figure 3: Vapour pressure [Pa] at at 25 °C vs. concentration measured on the fruits, normalised for an application concentration in the spray mixture of 1 kg co-formulant per hL. The concentrations were measured 1 hour after treatment, or 10 minutes in the case of cyclohexanone.

2.6 Residues of co-formulants on market samples

In the laboratory tests, it was established that a control sample (in other words, a pepper sample not treated in the laboratory test) exhibited readily verifiable residues of the surfactant dioctyl sulfosuccinate (docusate). For this reason, further vegetables from retail trade and control samples from the laboratory tests with other co-formulants (i.e. untreated apples, peppers and tomatoes) were investigated for the presence of this substance.

Residues of dioctyl sulfosuccinate were found in six vegetable samples (aubergines, peppers, cucumbers and courgettes, all originating from Spain) which were purchased in December 2018. In three samples, these concentrations were ≥ 0.01 mg/kg, in other words above the default limit of quantification for plant protection product active substances. The

highest concentration was around 0.1 mg/kg. In three samples, the residues were in the range of the limit of quantification (0.002 to 0.005 mg/kg). In the other samples, which were taken at a different time, homogenised and frozen, no residues of dioctyl sulfosuccinate were found (< 0.002 mg/kg).

Table 7: Residues of the co-formulant docusate in market samples from retail trade

Product	No. of samples	Measured docusate residues [mg/kg]
Apples	3	0.002; < 0.002; < 0.002
Aubergines	1	**0.110**
Cucumbers	1	**0.010**
Peppers	4	**0.020**; 0.005*; 0.003; < 0.002
Tomatoes	2	< 0.002; < 0.002
Courgettes	1	0.005

Limit of quantification: 0.002 mg/kg
bold: Samples with residues above the default maximum residue content of 0.01 mg/kg for plant protection product active substances
* The samples were from organic farming according to the declaration. Products with the active substance potassium bicarbonate (such as Armicarb) are approved for organic farming.

For the other co-formulants investigated (DMDA, components of solvent naphtha and cyclohexanone), no residues above the respective limit of quantification or above the blank value concentrations were found in the control samples from the laboratory tests. Therefore, no further market samples were tested.

2.7 Expected residues under practical conditions

Although the laboratory tests gave initial indications of the behaviour and the possible formation of residues on harvested produce, the test setup gave rise to certain limitations: (1) Immersing the fruit in the spray mixture might not adequately simulate the application of the plant protection product in the field, and (2) The impact of weather conditions as well as differing surface properties of fruits in the field compared to fruits that have already been harvested and stored may affect the behaviour of the co-formulants.

Therefore, it is only possible to obtain data on actual concentrations under field conditions in field tests. However, estimating the expected concentrations can help to organise the laboratory findings and plan field tests.

The concept of the RUD (residue unit dose) was developed for the risk assessment of plant protection product applications for birds and mammals [8]. The RUD values used for various crop groups indicate the amount of expected residues for a normalised active substance concentration directly after application. The values are largely based on a publication by Baril et al. [10], in which the authors evaluated data from around 13,000 residue studies. In a refinement of the RUD concept, Maclachlan and Hamilton [11] provided normalised day-zero values ($C_{0,norm}$) for a wide range of individual crops. These are based on numerous measured residue values for plant protection product active substances, with the median, mean and 90th percentile being stated in each case. The $C_{0,norm}$ values (median) and the estimated

concentrations for the co-formulants investigated and spiroxamine are compiled in Table 8 for selected crops.

Table 8: Estimation of the expected residues of the investigated co-formulants and the active substance spiroxamine on selected harvested produce according to [11]. The concentrations shortly after application (day 0 values) are stated, based on the median $C_{0,norm}$ value.

Co-formulant		**DMDA**	**2-MN**	**Cyclohexa-none**	**Docusate**	**Spiroxa-mine**
$C_{SprayMixture}$ (kg/hL)		0.0386	0.0031	0.025	0.0267	0.0306
Harvested produce	$C_{0,norm}$ [mg/kg]	Estimated concentration shortly after application $C_{0,calc}$ [mg/kg]				
Apples	10	0.386	0.031	0.250	0.267	0.306
Peppers	12	0.463	0.037	0.300	0.320	0.367
Tomatoes	3.6	0.139	0.011	0.090	0.096	0.110
Salad	180	6.9	0.558	4.500	4.806	5.508

$C_{SprayMixture}$: Concentration in kg/hL of the co-formulant (or active substance) in the "spray mixture" in the laboratory tests.

$C_{0,norm}$: Estimate of the expected residues in mg/kg on the corresponding harvested produce, normalised for an application concentration of 1 kg co-formulant (or active substance) per hL spray mixture. The median is given here.

$C_{0,calc}$: Estimated concentration on the corresponding harvested produce (at time 0) based on $C_{0,norm}$ and the concentration of the co-formulant (or active substance) in the spray mixture ($C_{SprayMixture}$).

The following remarks can be made about the estimated values for the expected residues:

a) They are based on the median from a large quantity of measured residue data. However, the values can deviate significantly in individual cases; the stated range extends over two orders of magnitude in some cases.

b) They estimate the residues shortly after application, and therefore do not take any degradation or other decrease in the concentration into consideration. Somewhat lower concentrations would therefore be expected for volatile compounds. This is also evident in the comparison of $C_{0,calc}$ and measured residues (see Fig. 4).

c) The amount of estimated residues is heavily dependent on the crop. In the fruits examined (apples and fruiting vegetables), they are comparatively low. By contrast, they are much higher in leafy vegetables at the same application quantity (see the calculated values in Table 8). This is related to the fact that a smaller amount of the plant protection product applied ends up on the harvested produce in the case of fruit and vegetables cultivated for their fruit.

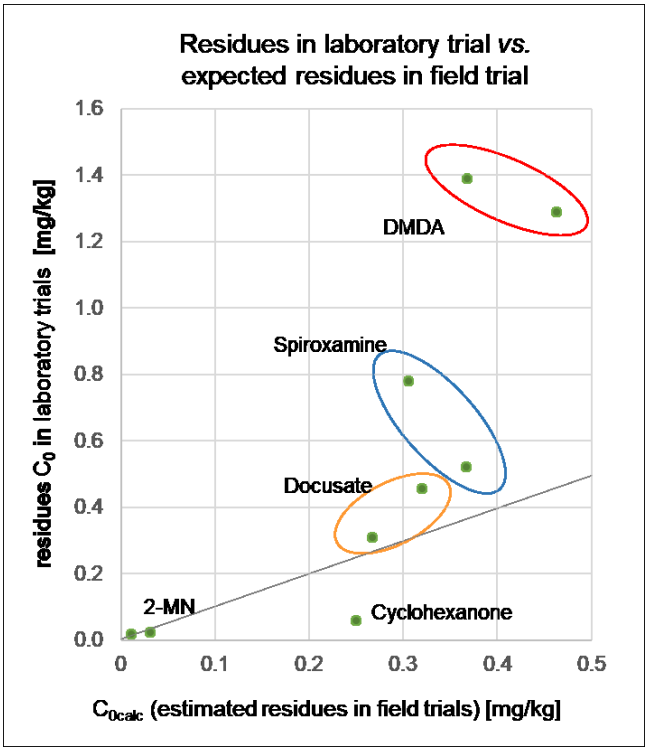


Figure 4: Residues measured in the laboratory tests 1 hour after treatment or 10 minutes after treatment (cyclohexanone) vs. the day 0 values estimated for field conditions according to [11] ($C_{0,calc}$). For data points above the line, the residues measured in the laboratory tests are greater than expected under field conditions.

For the less volatile substances docusate, DMDA and spiroxamine, the values measured in the laboratory tests shortly after treatment were higher than the estimated value $C_{0,calc}$, but were on the same order of magnitude (higher by a factor of max. 3.8 for DMDA). Conversely, for the more volatile compounds cyclohexanone and 2-methylnaphthalene, the measured residues were lower than the calculated values. This indicates that only a low amount of these compounds ends up on the surfaces of the produce, or that they evaporate from them very quickly. The estimates do not provide any indication of the expected residues at the time of harvesting, several days or weeks after application.

2.8 Summary and outlook

The studies conducted in this sub-project have shown that with the established methods for analysing the residues of plant protection product active substances (QuEChERS in combination with GC or LC-MS/MS), various plant protection product co-formulants that are frequently used can be verified specifically and sensitively. At the same time, it has been confirmed that the analysis of co-formulants presents some challenges, in particular due to the background levels and blank values.

The laboratory tests indicated that the assumption that co-formulants can lead to verifiable residues is correct. This was confirmed with the detection of the co-formulant docusate in market samples. It has been found that the amount of residue is heavily influenced not only by the concentration in the spray mixture but also by the volatility of the compound examined. It was not possible to determine the extent to which other substance properties (in particular the octanol/water partition coefficient) could have an impact on the amount and behaviour of the residues.

Only field tests will be able to provide information on the actual residues of plant protection product co-formulants in foods of plant origin. Such tests are planned for the next sub-project. Selecting suitable co-formulants (or plant protection products) and crops will be of major importance here.

The experience gained in the laboratory tests shows that it is less worthwhile to investigate very volatile compounds outdoors since barely any verifiable residues are to be expected. At the very least, the prospect of investigating solvent naphtha or other mixtures, as well as substances very widely used in other applications, must be reviewed carefully beforehand and the amount and source of background contamination and blank values must be known. DMDA and docusate, a solvent and a surfactant, are two compounds that appear readily suited to investigation in the field.

In addition to the fruits or the vegetables cultivated for their fruit that were tested in the laboratory, further vegetable crops, in particular those belonging to the group of leafy vegetables, should be investigated, since somewhat higher levels of residue are expected here.

2.9 Acknowledgements

We would like to thank our project partner, the Federal Food Safety and Veterinary Office (FSVO), for making this project possible, as well as Christoph Geiser, Ursina Zürcher and Jürg Zarn for the interesting discussions and excellent cooperation. We are very grateful to Thomas Poiger from Agroscope for his expert assistance with analytical issues, critical input on the project, and valuable comments on the report. Ignaz Bürge and Daniel Baumgartner from Agroscope assisted us with gas chromatography mass spectrometry and with developing a basis for estimating residues, respectively. We would like to extend our sincere thanks for this.

3 Introduction

3.1 Background

The Plant Protection Products Ordinance (PlantPPO) defines residues as one or more substances present in, or on, plants or plant products, edible animal products, drinking water or elsewhere in the environment and resulting from the use of a plant protection product (PPP), including their metabolites, breakdown or reaction products. The PlantPPO further specifies that residues of plant protection products must not produce any harmful effects on human or animal health or on groundwater.

A plant protection product (PPP formulation) generally consists of one or more active substances, and co-formulants. In practice, the risk assessment and the regulation of residues are limited exclusively to the active substances in the plant protection products and their metabolites and breakdown or reaction products. Annex 2 of the PestRO[8], for example, only lists maximum residue levels for active substances in plant protection products. Co-formulants of plant protection products are not considered. Since, when using plant protection products, the co-formulants are applied to the crop in the same way as the active substances, it must be assumed that they will also be found on the food products. At present, however, it is not possible to assess the resulting risk to consumers. This is because, on the one hand, the exposure level is unknown, and on the other hand, the co-formulants have in most cases not been sufficiently characterised from a toxicological standpoint. In the “Action plan for plant protection products” [12], therefore, a corresponding research need is identified: “In the context of the further development of the risk assessment of plant protection products for consumers, risks arising from co-formulants in food products are to be identified and, where necessary, new measures defined.”

In Switzerland, several hundred different plant protection (PP) products with varying compositions and formulation types are authorised. While the contents of the active substance are declared on every pack of a PP product, co-formulants do not generally need to be publicly disclosed. An exception to this is co-formulants that contribute to the classification of a PP product in terms of specific effects, such as acute toxicity, skin corrosion, or sensitisation[9]. These co-formulants must be declared, but there is no requirement to specify the amount. Some of the publicly available safety data sheets for the PP products contain information on co-formulants, but this is usually incomplete. Although the complete composition of the plant protection product formulations is part of the dossier that must be submitted to the responsible authority for authorisation, this is confidential.

An overview of the various formulation types, their prevalence and the average composition of PP products was drawn up in the preliminary study for this project [7]. This evaluation showed that the main components by far in PP products are the active substance or substances, and water (in liquid formulations) or inert carrier materials such as powdered stone (in solid formulations), followed by the substance group of surfactants. An exception to this is products that are formulated as emulsion concentrates (EC), which contain 70%

[8] FDHA Ordinance on the Maximum Residue Levels for Pesticides in or on Products of Plant and Animal Origin (PestRO) of 16 December 2016 (version: 1 May 2018)

[9] See the European Regulation on the classification, labelling and packaging of substances and mixtures (Regulation (EC) No 1272/2008, Art. 18).

solvent as co-formulant on average. In 2015, the formulation type EC was the fourth most commonly sold by quantity.

3.2 Aims of the sub-project on substance selection and method development

Together with future work of a more in-depth nature, this sub-project aims to establish a basis for risk assessment and to enable risks arising from co-formulant residues in food products to be identified and, where necessary, measures to be defined for the protection of consumers. The first step is to gather information on the potential exposure, in other words the type and amount of co-formulant residues on food products.

Therefore, co-formulants or components thereof are to be selected and examined in order to determine their suitability for further residue investigations. This includes the development of analytical methods for these substances, as well as simple laboratory tests that can provide some indication of the behaviour of the co-formulants on harvested produce. The findings obtained in this way will form the basis for planning further sub-projects, including field tests. Research shall also be carried out into the availability of past scientific studies or reports on the determination and prevalence of PP co-formulants (literature study).

The following points had to be taken into consideration when selecting co-formulants:

- the substances should be present in PP products at a sufficiently high concentration,
- they should have significant sales volumes,
- if possible, they should be representative of a category of co-formulants,
- they should be strong candidates for the development of a reliable, and if possible simple, analysis at trace levels, and
- reference material must be available so that the residues can be reliably quantified.

When selecting the harvested produce on which to conduct initial tests, the following aspects in particular had to be considered:

- the harvested produce selected should be representative of a group of food products that are consumed in significant quantities,
- they should be easy to handle in a laboratory test setting,
- they should be available all year round,
- they should not make analysis unnecessarily difficult,
- if possible, PP products with the selected co-formulants should be approved for use on the corresponding crops.

Developing the analytical methods for co-formulants in plant matrices posed the following challenges in particular. These challenges in turn had an impact on which substances were selected for further investigation:

- Background levels: Co-formulants are generally not used exclusively in PP products, but can also occur from other sources. This can lead to background contamination on the plants, and can also cause difficulties during analysis (blank values).
- Co-formulants often do not consist of a single substance but are themselves mixtures of substances. Since in each case the individual components, which only make up part of the overall co-formulant, are analysed, this results in a higher limit of quantification and limitations for quantification.
- The analysis must be adapted for different matrices (e.g. fatty or acidic foodstuffs, foodstuffs containing chlorophyll)

When developing a method, existing standard procedures from the field of pesticide residue analysis had to be adapted for the study of plant material. Depending on the substance properties, LC-MS/MS or GC-MS(/MS) presented themselves as suitable detection methods.

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4 Details and results for the selected co-formulants

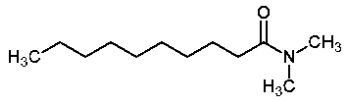
In this project, the four co-formulants N,N-dimethyldecanamide, solvent naphtha, cyclohexanone and dioctyl sulfosuccinate were investigated. These are found in the plant protection products Input (W6392), Slick (W5056), Milbeknock (W6526) and Armicarb (W6432). Since solvent naphtha is a mixture of various aromatic hydrocarbons, the components 2-methylnaphthalene, 1-methylnaphthalene and biphenyl, which are found in this co-formulant, were selected for the method development and the tests. Apples, peppers and tomatoes were the harvested produce used because they are available in shops at all times, and experience has shown that they rarely cause matrix problems due to interfering signals during analysis. The analytical methods, the storage tests and the determination of residue on the harvested produce are described below for each of these individual co-formulants or co-formulant components.

4.1 Co-formulant N,N-dimethyldecanamide (solvent)

4.1.1 Introduction and literature

The preliminary study on co-formulants in plant protection products [7] showed that N,N-dimethyldecanamide (DMDA) is a solvent in emulsion concentrates that is used in large quantities. According to estimates, approximately 9 tonnes of this solvent were sold in Switzerland in 2015 as a constituent of plant protection products. By quantity, it is the seventh most commonly used solvent in PP products. According to our assessment, this solvent was well-suited to helping us answer the questions posed in this sub-project. The physicochemical properties of dimethyldecanamide are summarised in the table below.

Table 9: Designations, structural formula and physicochemical properties of DMDA.

Designations	N,N-Dimethyldecanamide N,N-Dimethylcapramide N,N-Dimethylcaprinamide Decanoic acid dimethylamide
Structural formula	
CAS no.	14433-76-2
Molecular weight (g/mol)	199.335
Molecular formula	$C_{12}H_{25}NO$
Appearance, properties	Liquid, yellowish
Melting point	-7 °C
Initial boiling point	291 °C at 1,013 hPa
Vapour pressure	0.11 Pa at 25 °C
Relative density	0.88
Water solubility	0.34 g/L at 20 °C
Partition coefficient: n-octanol/water log P_{OW}:	3.44

Source: ECHA Submission Dossier[10]

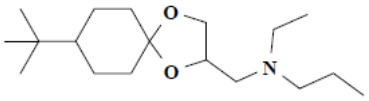
In addition to N,N-dimethyldecanamide, the similar substance N,N-dimethyloctanamide is also found in PP products. This is also used as a solvent. In the US, these two solvents are approved as constituents in agrochemical products [13]. The use of DMDA has already been discussed in sections 2.2 and 2.3. No further literature on DMDA as a co-formulant in PP products or as a residue on harvested produce was found.

Dimethyldecanamide is a pure substance that has no isomers. In preliminary tests, when analysed via both gas and liquid chromatography, it yielded with both techniques good signals that were easy to identify.

In addition to the co-formulant N,N-dimethyldecanamide, the active substance spiroxamine, which is also a constituent of the product Input, was also measured.

[10] ECHA dossier submission on N,N-dimethyldecan-1-amide: https://echa.europa.eu/de/registration-dossier/-/registered-dossier/15021/1

Table 10: Designations, structural formula and physicochemical properties of spiroxamine.

Designations	Spiroxamine 8-tert-butyl-1,4-dioxaspiro[4.5]decan-2-ylmethyl(ethyl)(propyl)amine (ISO)
Structural formula	
CAS no.	118134-30-8
Molecular weight (g/mol)	297.5
Molecular formula	$C_{18}H_{35}NO_2$
Melting point	< - 170 °C
Vapour pressure	4 / 6 x 10^{-3} at 20 °C
Water solubility	470 / 340 (pH 5) at 20 °C 14 / 10 (pH 9) at 20 °C
Partition coefficient: n-octanol/water log POW:	2.8 / 3.0 at pH 7

Source: EFSA conclusion 2010 on spiroxamine http://www.efsa.europa.eu/en/efsajournal/pub/1719. The active substance consists of two stereoisomers, so two values are stated in each case.

4.1.2 Analytical method for the co-formulant N,N-dimethyldecanamide and the active substance spiroxamine

The samples were prepared according to the QuEChERS method, which is widely used in the field of pesticide residue analysis [9]. The procedure is described in detail in Appendix 6.2 and 6.3.

10 g of homogenised sample were extracted with 10 mL of acetonitrile and then purified with QuEChERS tube I (partitioning of acetonitrile/saline aqueous phase). 1 mL of the acetonitrile phase was then poured into QuEChERS tube II (dispersive solid phase extraction with primary secondary amine solid phase). Part of the residue after centrifuging (extract II) was analysed using LC-MS/MS.

Table 11: Analytical method for DMDA: parameters and procedure.

	N,N-Dimethyldecanamide	Spiroxamine
Extraction agent	Acetonitrile	Acetonitrile
Purification I (liquid/liquid partitioning, QuEChERS I)	Yes	Yes
Purification II (dispersive solid phase, QuEChERS II)	Yes	Yes
MS MRM quantification	m/z 200.2 → m/z 102.2	m/z 298.3 → m/z 100.1
MS MRM confirmation	m/z 200.2 → m/z 116.1	m/z 298.3 → m/z 116.1
Calibration (5 points)	1 mg/L to 0.01 mg/L	1 mg/L to 0.01 mg/L
Correlation coefficient r	0.9965	0.9986
Limit of Quantification[1]	0.002 mg/kg	0.002 mg/kg
Blank value	≤ 0.0005 mg/L	≤ 0.0005 mg/L
Recovery rate for pepper [2] (5 separate processes)	99%	105%
Recovery rate for apple [2] (5 separate processes)	92%	99%
Expanded uncertainty (estimated, see 6.3)	± 20%	± 20%

[1] *Limit of Quantification: signal/noise is ≥ 10*

[2] *At concentrations of 0.5 mg/kg (n=2), 0.1 mg/kg (n=2) and 0.03 mg/kg (n=1)*

Typical chromatograms are presented in Figures 5 and 6 below.

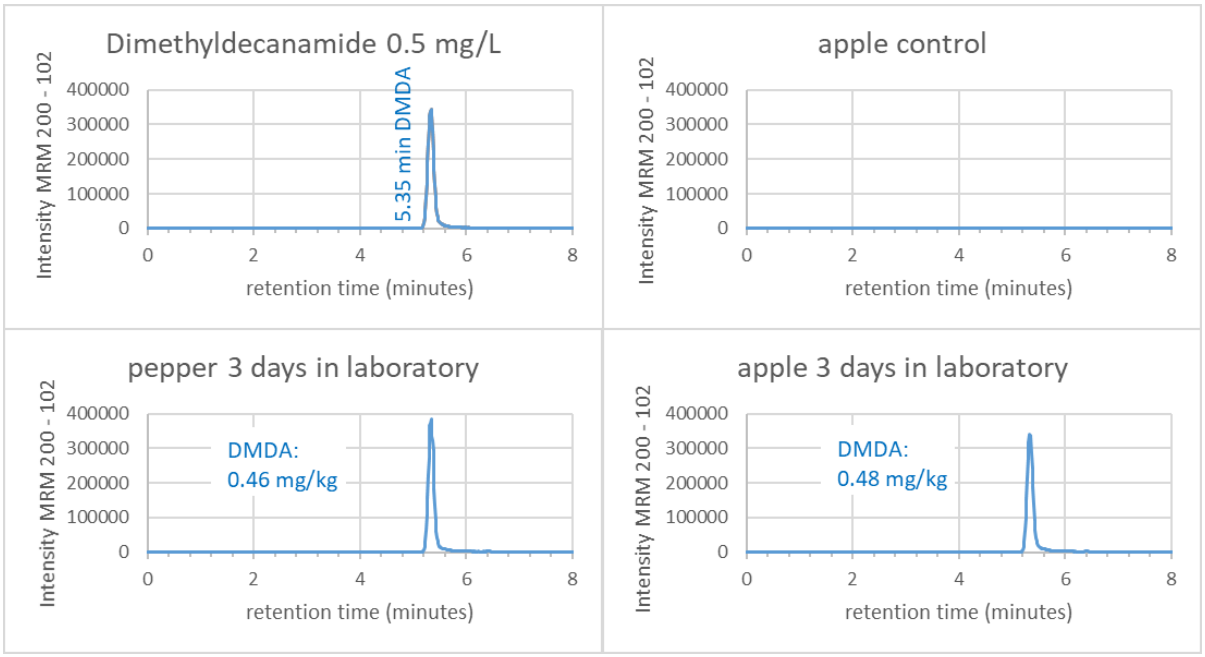


Figure 5: Chromatograms for dimethyldecanamide determination using LC-MS/MS. One standard (0.5 mg/L), one pepper sample after 3 days, one apple sample after 3 days and one apple control are shown. Retention time of dimethyldecanamide (DMDA): 5.35 minutes.

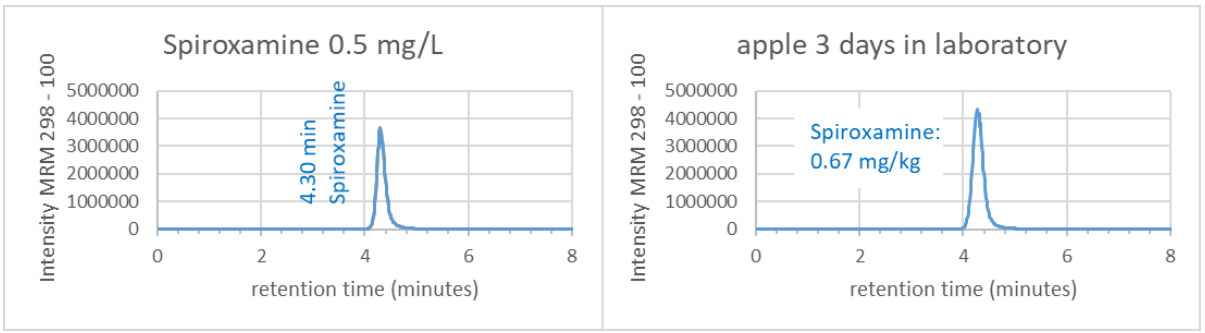


Figure 6: Chromatograms for spiroxamine determination using LC-MS/MS. One standard (0.5 mg/L) and one apple sample after 3 days are shown. Retention time of spiroxamine: 4.30 minutes.

4.1.3 Laboratory tests with apple and pepper

The PP product Input, federal registration no. W6392, was selected for the storage tests. Input has a declared spiroxamine content of 30.5 % (300 g/L). The safety data sheet for Input specifies a N,N-dimethyldecanamide content of > 20 % (safety data sheet, Bayer Switzerland, Input, CH version dated 24/01/2018[11]). Our own measurements (GC-FID) gave

[11] MSDS for Input, Bayer https://www.google.ch/url?sa=t&rct=j&q=&esrc=s&source=web&cd=6&cad=rja&uact=8&ved=2ahUKEwjcpKaS44HgAhWpsaQKHeK1D4UQFjAFegQICRAC&url=https%3A%2F%2Fpim.bayercropscience.ch%2Fsdb.pdfstream%3Fprod-uct%3D38%26lang%3Dde&usg=AOvVaw3w5jpcnAsFOL72egkbEO6-

a DMDA content of 38.6 % in the product batch available to us. The PP product Input is permitted for use in agriculture (cereals), but not on the crops apples or peppers. However, it is suitable for use as a model of a PP product that contains a solvent with low volatility in a high concentration.

Peppers and apples were purchased from a retail outlet (30 of each, purchased on 24/07/2018). This harvested produce, with the exception of the control, was immersed in a spray mixture of the PP product Input with a concentration of 0.1%. The harvested products (apples, peppers) were held by their stalk with tweezers and immersed in the “spray mixture” up to approx. 80% of their height. Complete immersion of the harvested produce was avoided. This prevented the spray mixture from running into the depression at the base of the stalk, which could lead to locally excessive and non-reproducible residues. The concentrations of dimethyldecanamide and spiroxamine in this spray mixture were 386 and 306 mg/L.

The treated peppers and apples were left to dry for an hour. Approximately half the produce was then stored in open trays in the laboratory in a fume cupboard for 1 or 3 days. The other half was placed on the roof of the laboratory (Müller-Thurgau-Strasse 29, 8820 Wädenswil, Switzerland, N47.221792° E8.677035°), where it was exposed to solar radiation, a higher temperature, and air circulation (wind) (24/07/2018 to 27/07/2018). This three-day period was characterised by hot, sunny summer weather, with temperatures above 30 °C and no rain. The mean temperature was 25.0 °C and the maximum was 31.6 °C. The weather data from Agrometeo for the fruit growing station in Wädenswil can be found in Table 35 of the appendix. Accordingly, a significant change in the appearance and consistency of the samples was observed, in connection with water loss (the samples appeared almost “cooked”, Figure 7).

Figure 7: Peppers and apples from the storage test: left: stored in the laboratory for 15 hours, right: stored on the roof for 3 days.

Treatment with the spray liquid resulted in residues of approx. 1.3 mg/kg dimethyldecanamide and 0.5 to 0.8 mg/kg spiroxamine (concentrations 1 h after application, Fig. 8 above and Table 12). Residues of neither DMDA nor spiroxamine were measured in the untreated control samples (Table 12).

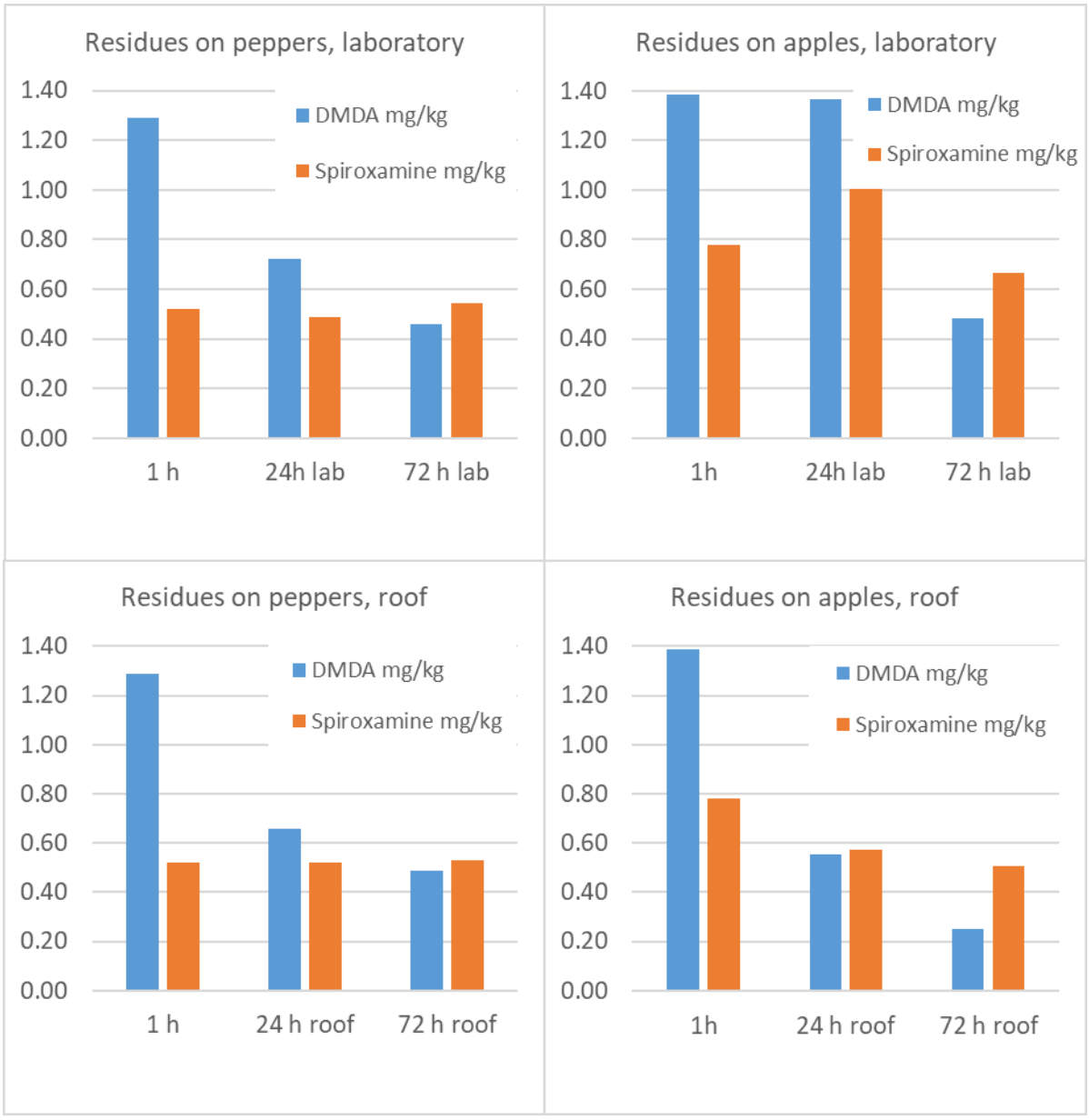


Figure 8: Residues of dimethyldecanamide and spiroxamine on peppers (left) and apples (right) after 1 hour and after storage in the laboratory for 1 day and 3 days (top) or after storage on the roof for 1 day and 3 days (bottom, sample after 1 h is identical to that of the laboratory test in each case). At the time t=0, the peppers and apples were immersed once in a spray mixture (0.1% plant protection product Input).

Storage in the laboratory: slow decrease in concentration of DMDA, virtually no decrease in spiroxamine

In the laboratory test with **peppers**, the concentration of the co-formulant dimethyldecanamide decreased continuously over the 3 days to approximately 1/3 of the

original value. During this time, the concentration of the active substance spiroxamine remained at approx. 0.5 mg/kg. In **apples**, the residues were slightly higher than in peppers. Furthermore, the concentration of the co-formulant after 1 day was still almost at the initial level, but this also decreased to approximately one-third after 3 days. The concentration of spiroxamine on apples was also unexpectedly high in the sample after 24 h. The high values after 24 h can be attributed to the variance in the test procedure and the measurement uncertainty.

Table 12: Residues of dimethyldecanamide and spiroxamine on peppers and apples in mg/kg. The results are shown in graph format in Figure 8.

	Peppers DMDA (mg/kg)	Peppers Spiroxamine (mg/kg)	Apples DMDA (mg/kg)	Apples Spiroxamine (mg/kg)
Blank	< LOQ	< LOQ.	< LOQ	< LOQ
Control	< LOQ	< LOQ	< LOQ	< LOQ
1 h	1.29	0.52	1.39	0.78
24 h lab	0.72	0.49	1.37	1.01
24 h roof	0.66	0.52	0.55	0.57
72 h lab	0.46	0.55	0.48	0.67
72 h roof	0.49	0.53	0.25	0.50

< LOQ : not quantifiable (limit of quantification 0.002 mg/kg)
Concentration in the spray mixture: DMDA 386 mg/L; spiroxamine 306 mg/L.

Storage outdoors: slightly faster decrease in DMDA, virtually no decrease in spiroxamine

For the produce stored on the roof, the amount of residues and the changes over time were similar to those stored in the laboratory at room temperature (Fig. 8, Table 12). In apples alone, the amount of co-formulant residue after storage for 3 days on the roof was only half as high as *that of the samples stored in the laboratory*.
The water loss for the produce on the roof was estimated to be < 20% (visual assessment). Although not especially high, the water loss in the peppers and apples could have influenced the progression over time to a certain degree (slightly higher concentration of residues).

Interpretation, expectations

Since the co-formulant dimethyldecanamide performs the role of a solvent, especially in formulations of the emulsion concentrate type, it was expected that its concentration on harvested produce stored on the roof at elevated temperatures, in direct sunlight and in the ambient air (convection) would decrease more quickly than for the produce stored in the laboratory. This was not confirmed; the differences between the two storage methods were low. Dimethyldecanamide may have been retained in hydrophobic structures of the peppers and apples (e.g. wax layer). Furthermore, DMDA has comparatively low volatility for a solvent.

4.2 Co-formulant solvent naphtha (solvent)

4.2.1 Introduction and literature

Solvent naphtha is used as a solvent in plant protection product formulations. It is a mixture of various, primarily aromatic hydrocarbons, and is obtained from crude oil. One trade name of this widely used co-formulant is Solvesso 200 ND. ND stands for naphthalene-depleted and indicates that the naphthalene content has been reduced to < 1%.
Solvent naphtha is used in large quantities in emulsion concentrates as a solvent [7]. According to our estimates, approximately 53 tonnes of this solvent were sold in Switzerland in 2015 as a constituent of plant protection products. By quantity, it is the third most commonly used solvent in PP products.

Although solvent naphtha was deemed to be difficult to analyse owing to its many components, it was selected for this sub-project because it is so significant in quantity terms and because it is regarded as one of the classic solvents used in emulsion concentrates. In order to simplify the tests, the measurements were restricted to the model substances (co-formulant components) 2-methylnaphthalene, 1-methylnaphthalene and biphenyl.

The physicochemical properties of solvent naphtha, 2-methylnaphthalene, 1-methylnaphthalene and biphenyl are summarised in the tables below.

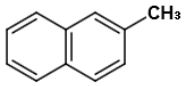
Table 13: Designations, structural formula and physicochemical properties of solvent naphtha.

Designations	Solvent Naphtha, heavy aromatic Solvesso 200 ND Hydrocarbons, C10-C13, aromatics, <1% naphthalene
Structural formula	Mixture of aromatic hydrocarbons with C10 to C13
CAS no.	64742-94-5
Molecular weight (g/mol)	variable
Molecular formula	
Melting point	173 - 179 °C
Boiling point/range	200 °C – 310 °C
Vapour pressure	< 0.1 kPa (0.75 mm Hg) at 25 °C
Relative density	0.951 – 1.051 g/cm³, at 15 °C
Water solubility	negligible

Source: ECHA substance information for solvent naphtha [12]

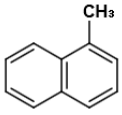
[12] ECHA substance information for solvent naphtha (petroleum), heavy arom. https://echa.europa.eu/de/substance-information/-/substanceinfo/100.059.253

Table 14: Designations, structural formula and physicochemical properties of 2methylnaphthalene.

Designations	2-Methylnaphthalene beta-Methylnaphthalene
Structural formula	
CAS no.	91-57-6; methylnaphthalene, not specified 1321-94-4
Molecular weight (g/mol)	142.20
Appearance, properties	Crystalline, white, aromatic odour
Melting point	34.6 °C
Initial boiling point	241.1 °C
Vapour pressure	7.3 Pa at 25 °C
Vapour density	3.39 (air = 1.0)
Density:	1.006 g/cm^3 at 25 °C
Water solubility	24.6 mg/L at 25 °C
Partition coeff.: n-octanol/water log POW:	3.86

Source: NIH ToxNet[13], Gestis[14] and PubChem[15] databases

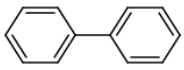
Table 15: Designations, structural formula and physicochemical properties of 1-methylnaphthalene.

Designations	1-Methylnaphthalene alpha-Methylnaphthalene
Structural formula	
CAS no.	90-12-0; methylnaphthalene, not specified 1321-94-4
Molecular weight (g/mol)	142.20
Appearance, properties	liquid, light yellow, aromatic odour
Melting point/range:	-31 °C
Initial boiling point	245 °C
Vapour pressure	8.9 Pa at 25 °C
Density:	1.02 g/cm^3 at 20 °C
Water solubility	25.8 mg/L at 25 °C
Partition coeff.: n-octanol/water log POW:	3.87

Source: NIH ToxNet, Gestis and PubChem databases

[13] https://chem.nlm.nih.gov/chemidplus/rn/91-57-6
[14] http://gestis.itrust.de/nxt/gateway.dll/gestis_de/000000.xml?f=templates$fn=default.htm$vid=gestisdeu:sdb-deu$3.0
[15] https://pubchem.ncbi.nlm.nih.gov/compound/7055#section=Top

Table 16: Designations, structural formula and physicochemical properties of biphenyl.

Designations	Biphenyl Diphenyl Phenylbenzene 1,1'-Biphenyl E 230
Structural formula	
CAS no.	92-52-4
Molecular weight (g/mol)	154.21
Appearance, properties	Crystalline, light yellow
Melting point/range:	68 - 70 °C
Initial boiling point	255 °C
Vapour pressure	0.01 hPa at 25 °C
Density:	1.04 g/cm^3 (20 °C)
Water solubility	6.9 mg/L at 25 °C
Partition coeff.: n-octanol/water log POW:	3.98

Source: NIH ToxNet, Gestis and PubChem databases

In Sweden in 2011, medium aromatic solvent naphtha was one of the top 10 most frequently used substances in chemical products [14]. According to the statistics, there were 3280 products containing medium aromatic solvent naphtha at that time, corresponding to a total quantity of 18,856 tonnes.

Biphenyl used to be authorised as a pesticide active substance and is therefore often also included in investigations on PP product residues. Although it is no longer authorised as an active substance, low concentrations of biphenyl have been found in parsley (0.01 to 0.12 mg/kg) and other kitchen herbs [15, 16]. It has been suggested that biphenyl reaches the crops via the air. No studies on biphenyl residues arising from PP product formulations containing solvent naphtha have been conducted to date.

The use of solvent naphtha has already been discussed in sections 2.2 and 2.3. No further literature on solvent naphtha or the three selected components as a co-formulant in PP products or as a residue on harvested produce was found.

Solvent naphtha, and in particular the individual model substances, can be readily analysed using gas chromatography. However, it is not possible to separate out all the individual components of solvent naphtha.

4.2.2 Analytical method for the co-formulant components methylnaphthalene and biphenyl

The samples were prepared analogously (modification: extraction with ethyl acetate) to the widely used QuEChERS method [9]. The procedure is described in detail in Appendix 6.2 and 6.4. Measurement was conducted on GC-MS in single ion monitoring (SIM) mode. The parameters of the analytical method for methylnaphthalene and biphenyl are summarised in the table below.

Table 17: Analytical method for components of solvent naphtha: parameters and procedure.

	2-Methylnaphthalene	1-Methylnaphthalene	Biphenyl
Extraction agent	Ethyl acetate	Ethyl acetate	Ethyl acetate
Purification I (liquid/liquid partitioning, QuEChERS I)	Yes	Yes	Yes
Purification II (dispersive solid phase, QuEChERS II)	Yes	Yes	Yes
MS quantification (SIM)	m/z 142.1	m/z 142.1	m/z 154.1
Calibration (6 points)	1 to 0.005 mg/L	1 to 0.005 mg/L	1 to 0.005 mg/L
Correlation coefficient r	0.9914	0.9909	0.9932
Limit of Quantification[1] (calibration solutions)	0.002 mg/kg	0.002 mg/kg	0.002 mg/kg
Blank value	approx. 0.006 mg/kg	approx. 0.003 mg/kg	approx. 0.001 mg/kg
Recovery rate for tomatoes [2] (6 separate processes)	97%	93%	92%
Recovery rate for apples [2] (6 separate processes)	108%	103%	104%
Expanded uncertainty (same as DMDA, see 6.3)	± 20%	± 20%	± 20%

[1] *Limit of Quantification: signal/noise is ≥ 10*

[2] *At concentrations of 0.5 mg/kg (n=4) and 0.1 mg/kg (n=2)*

The active substance difenoconazole, which is found in the formulation used, could not be determined using this GC-MS method because the signal was much too insensitive (SIM m/z 323, retention time 25.4 min, double peak for the two diastereomers, see 6.4).

Typical chromatograms for methylnaphthalene and for biphenyl are shown in the two figures below.

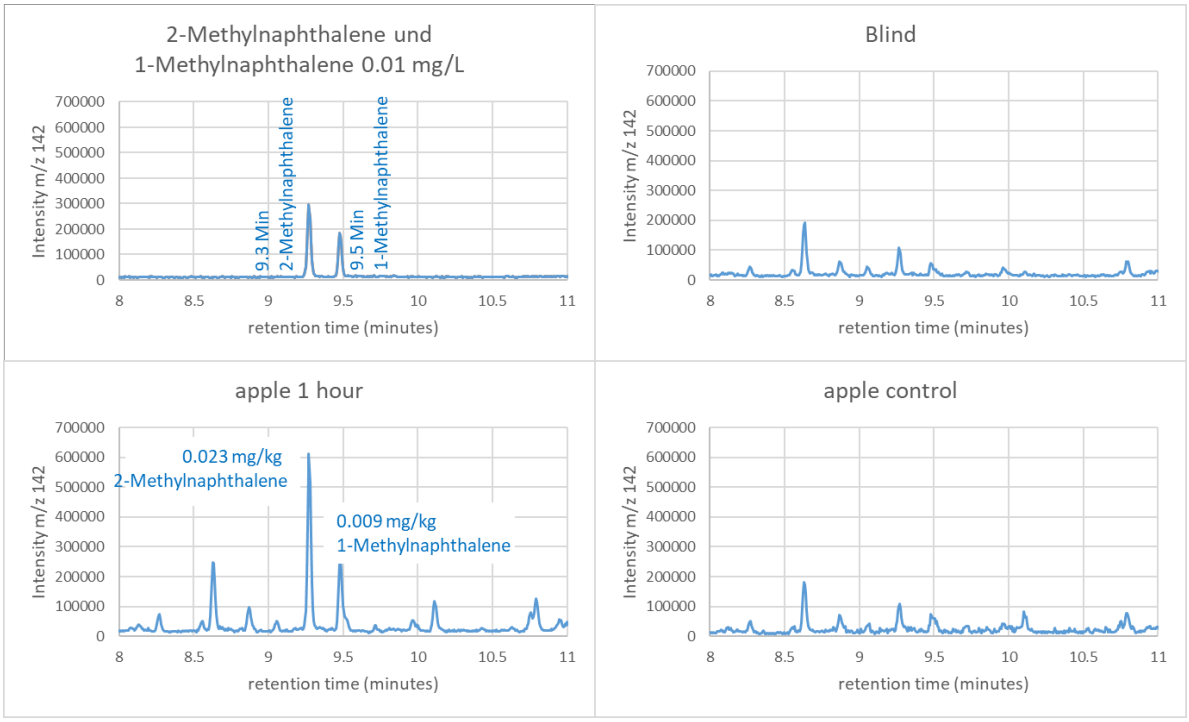


Figure 9: Chromatograms for methylnaphthalene determination using GC-MS. One standard (0.01 mg/L), one blank sample, one apple sample after 1 hour and one apple control are shown. Retention time of 2-methylnaphthalene: 9.3 minutes; retention time of 1-methylnaphthalene: 9.5 minutes.

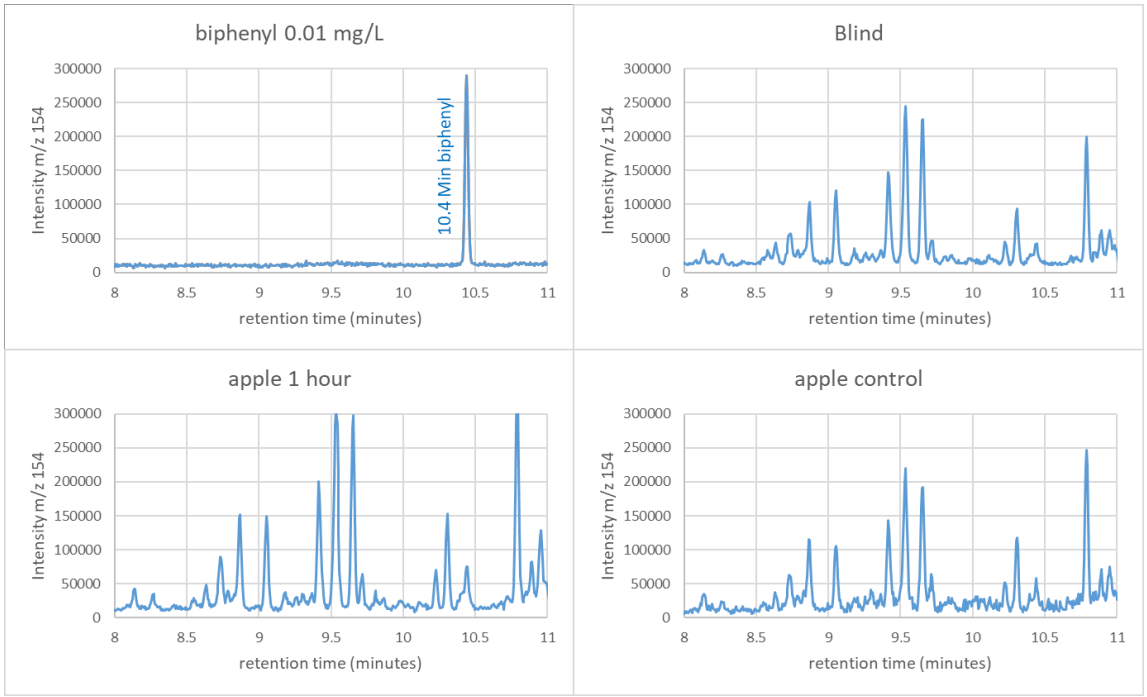


Figure 10: Chromatograms for biphenyl determination using GC-MS. One standard (0.01 mg/L), one blank sample, one apple sample after 1 hour and one apple control are shown. No biphenyl residues were found on the apple sample after 1 hour. Retention time of biphenyl: 10.4 minutes.

4.2.3 Laboratory tests with apple and tomato

The plant protection product Slick (fed. registration number W5056) was selected for the storage experiments. The product contains the active substance difenoconazole in a concentration of 23.5% (250 g/L). According to the safety data sheet[16] from the company, the content of solvent naphtha (petroleum, heavy aromatic) is between 50 and 70%. Our own measurements (GC-FID) showed that the batch of the plant protection product Slick used in the experiment contained 6.2% 2-methylnaphthalene, 3.1% 1-methylnaphthalene and only 0.16% biphenyl in the formulation.

Tomatoes and apples were purchased from a retail outlet (36 and 30 pieces, respectively; purchased on 25/09/2018). This harvested produce, with the exception of the control, was immersed in a spray mixture of the PP product Slick with a concentration of 0.05% (concentration of 2-methylnaphthalene, 1-methylnaphthalene and biphenyl in the spray mixture: 31, 16 and 0.8 mg/L respectively). The fruits were held by their stalk with tweezers and immersed in the “spray mixture” up to approx. 80% of their height, then left to dry for 1 hour. Approximately half of the fruits were stored in open trays in the laboratory in a fume cupboard for 1 or 3 days. The other half were exposed to atmospheric conditions on the roof of the laboratory building. This three-day period was characterised by sunny autumn weather, with temperatures above 20 °C and no rain. The mean temperature was 12.7 °C and the maximum was 21.9 °C. The weather data from Agrometeo for the fruit growing station in Wädenswil can be found in Table 36 of the appendix.

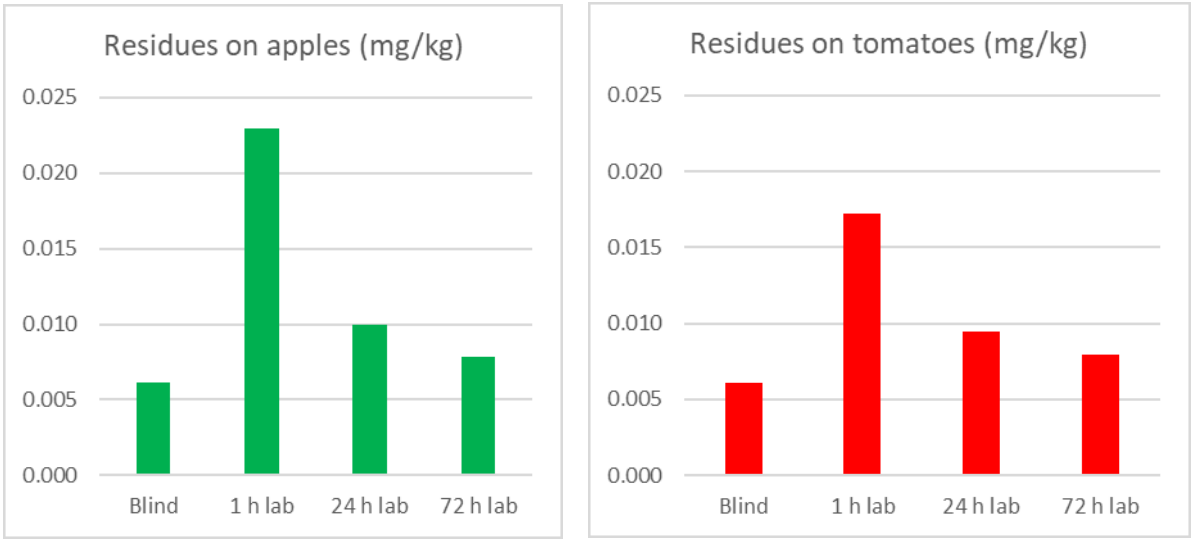


Figure 11: Blank value and residues of 2-methylnaphthalene on apples (left) and tomatoes (right) after storage in the laboratory for 1 hour, 1 day and 3 days. At the time t=0, the apples and tomatoes were immersed once in a spray mixture (0.05% plant protection product Slick).

[16] Safety data sheet for Slick (W5056) from Syngenta Agro AG, dated 03/01/2017 https://www.google.ch/url?sa=t&rct=j&q=&esrc=s&source=web&cd=6&cad=rja&uact=8&ved=2ahUKEwit7r7b5IHgAhWKEVAKHZ2EA0kQFjAFegQIBBAC&url=https%3A%2F%2Fwww.syngenta.ch%2Fsites%2Fg%2Ffiles%2Fzhg441%2Ff%2Fsd_slick_d.pdf%3Ftoken%3D1511172337&usg=AOvVaw38ORUUDNALFOLyN5PhUctM

It should be noted that the residue levels were already in the range of the blank value after one day of storage.

Table 18: Residues of methylnaphthalene and biphenyl on apples and tomatoes in mg/kg.

	2-Methylnaphthalene (mg/kg)		1-Methylnaphthalene (mg/kg)		Biphenyl (mg/kg)	
	Apples	Tomatoes	Apples	Tomatoes	Apples	Tomatoes
Blank	0.006	0.006	0.003	0.003	< LOQ (0.001)	< LOQ (0.001)
Control	0.006	0.003	0.003	0.002	< LOQ (0.001)	< LOQ (0.001)
1 h	0.023	0.017	0.009	0.008	0.002	< LOQ (0.0017)
24 h lab	0.010	0.010	0.005	0.005	0.002	< LOQ (0.0018)
24 h roof	0.012	0.008	0.006	0.004	0.002	< LOQ (0.0018)
72 h lab	0.008	0.008	0.004	0.004	0.002	< LOQ (0.0017)
72 h roof	0.009	0.006	0.004	0.003	0.002	< LOQ (0.0017)

< LOQ: not quantifiable (limit of quantification 0.002 mg/kg)
An estimate is provided in parentheses; however, this is below the limit of quantification.
Concentration in the spray mixture: 2-methylnaphthalene 31 mg/L; 1-methylnaphthalene 16 mg/L; biphenyl 0.8 mg/L.

In these storage experiments, the residues of **1-methylnaphthalene** and **biphenyl** were always below 0.01 mg/kg and only higher than the blank value by an insignificant amount (Table 18). Owing to the low residues, the results for these two co-formulant components of solvent naphtha are not discussed any further.

In contrast, the residues of **2-methylnaphthalene** were approx. 0.02 mg/kg, but decreased very rapidly and were already in the range of 0.01 mg/kg after 24 hours. Since concentrations in the range of from 0.003 to 0.006 mg/kg were also found in untreated control samples and in the blank sample, a further interpretation of the change in the residues over time is not possible.

The cause of the presence in control and blank samples could not be precisely determined. It was assumed that the QuEChERS tube I and II are responsible for the blank values, since the solvent used – ethyl acetate – did not exhibit an elevated content of the analytes. To date, it has not been possible to make further attempts to reduce or completely eliminate the blank values.

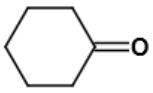
4.3 Co-formulant cyclohexanone (solvent)

4.3.1 Introduction and literature

Cyclohexanone is another solvent that is used in large quantities in emulsion concentrates [7]. According to estimates, approximately 20 tonnes of this solvent were sold in Switzerland in 2015 as a constituent of plant protection products. By quantity, it is the fourth most commonly used solvent in PP products in Switzerland [7]. The use of cyclohexanone has already been discussed in sections 2.2 and 2.3. No further literature on cyclohexanone as a co-formulant in PP products or as a residue on harvested produce was found.

The physicochemical properties of cyclohexanone are summarised in the table below.

Table 19: Designations, structural formula and physicochemical properties of cyclohexanone.

Designations	Cyclohexanone Anonsextone Ketohexamethylene Anone Sextone Pimelic ketone
Structural formula	
CAS no.	108-94-1
EC number	203-631-1
Molecular weight (g/mol)	98.15
Appearance, properties	clear, liquid, colourless, peppermint-like aroma
Melting point/freezing point:	-31 °C
Initial boiling point	154.3 °C
Vapour pressure	700 Pa at 30 °C
Relative density	0.9465 g/cm^3 at 20 °C
Water solubility	86 g/L at 20 °C
Partition coeff.: n-octanol/water log POW:	0.86 at 25 °C

Source: ECHA Registration Dossier[17]

Cyclohexanone is a pure substance with no isomers that can be readily analysed using gas chromatography.

4.3.2 Analytical method

The samples were prepared analogously (modification: extraction with ethyl acetate) to the widely used QuEChERS method [9]. The procedure is described in detail in Appendix 6.2 and 6.5. Measurement was conducted on GC-MS in single ion monitoring (SIM) mode. The parameters of the analytical method for cyclohexanone are summarised in the table below, and typical chromatograms are shown in Figure 12.

[17] ECHA substance information for cyclohexanone: https://echa.europa.eu/de/substance-information/-/substanceinfo/100.003.302

In the chromatogram for the *apple control*, a signal can be identified at 5.1 minutes (retention time of cyclohexanone). The signal is roughly as high as in the blank sample. It is therefore **not** a cyclohexanone residue in the apple control but rather the blank value.

Table 20: Analytical method for cyclohexanone: parameters and procedure.

Extraction agent	Ethyl acetate
Purification I (liquid/liquid partitioning, QuEChERS I)	Yes
Purification II (dispersive solid phase, QuEChERS II)	No
MS quantification (SIM)	m/z 98
Calibration (6 points)	1 to 0.005 mg/L
Correlation coefficient r	0.9968
Limit of Quantification[1] (calibration solutions)	0.002 mg/L
Blank value	approx. 0.022 mg/kg
Recovery rate for apple [2] (6 separate processes)	97%
Expanded uncertainty (same as DMDA, see 6.3)	± 20%

[1] *Limit of Quantification: signal/noise is ≥ 10*

[2] *At concentrations of 0.9 mg/kg (n=2), 0.4 mg/kg (n=2) and 0.1 mg/kg (n=2)*

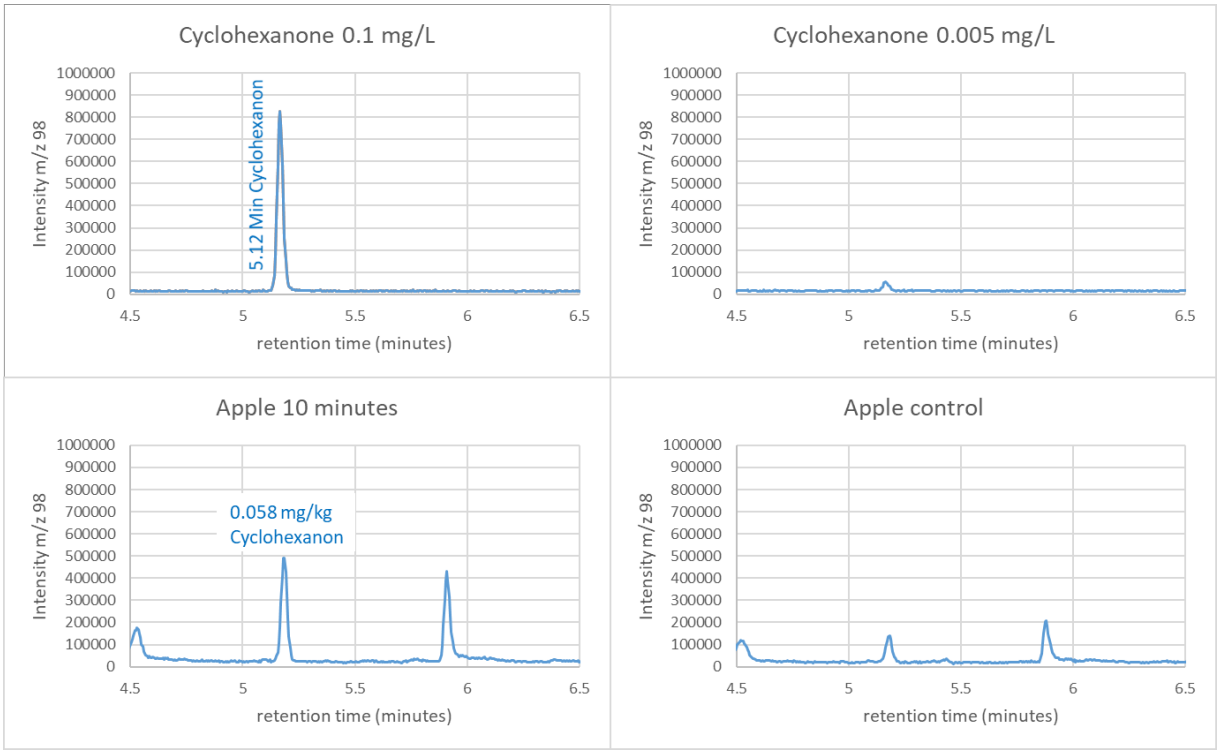


Figure 12: Chromatograms for cyclohexanone determination using GC-MS. One standard (0.1 mg/L), one low standard (0.005 mg/L), one apple sample after 10 minutes and one apple control are shown. Retention time of cyclohexanone: 5.12 minutes.

4.3.3 Laboratory tests with apple

The plant protection product Milbenknock (fed. registration number W6526) was selected for the storage experiments. The product contains the active substance milbemectin in a concentration of 1% (9.3 g/L). The safety data sheet[18] from the company declares the cyclohexanone content to be 10–25%. Measurement via GC-MS indicated that the content of the co-formulant cyclohexanone in the formulation was 20%.

Apples were purchased from a retail outlet (20 apples, purchased on 03/12/2018). At the time t=0, these apples – with the exception of the control – were immersed in a spray mixture of the PP product Milbenknock with a concentration of 0.125%. The concentration of cyclohexanone in the spray mixture was therefore 250 mg/L. The apples were held by their stalk with tweezers and immersed in the “spray mixture” up to approx. 80% of their height. The treated apples were left to dry for 10 minutes and were then stored in an open tray in the laboratory in a fume cupboard for 1 hour, 1 day and 3 days.

Owing to the high volatility of cyclohexanone, the additional data point after 10 minutes was measured for this experiment. 10 minutes after treatment with the Milbeknock spray mixture, the apples were quickly cut up and frozen immediately at -45 °C.

[18] MSDS for Milbeknock dated 8/02/2017, Omya Schweiz
https://www.google.ch/url?sa=t&rct=j&q=&esrc=s&source=web&cd=3&cad=rja&uact=8&ved=2ahUKEwjxwK7U5oHgAhUBKFAKHRIRCx0QFjACegQIAhAC&url=https%3A%2F%2Fwww.omya.com%2FAgroDocs%2FMilbeknock-D-70208.pdf&usg=AOvVaw1FteGFTCzX9E3DidoLxjED

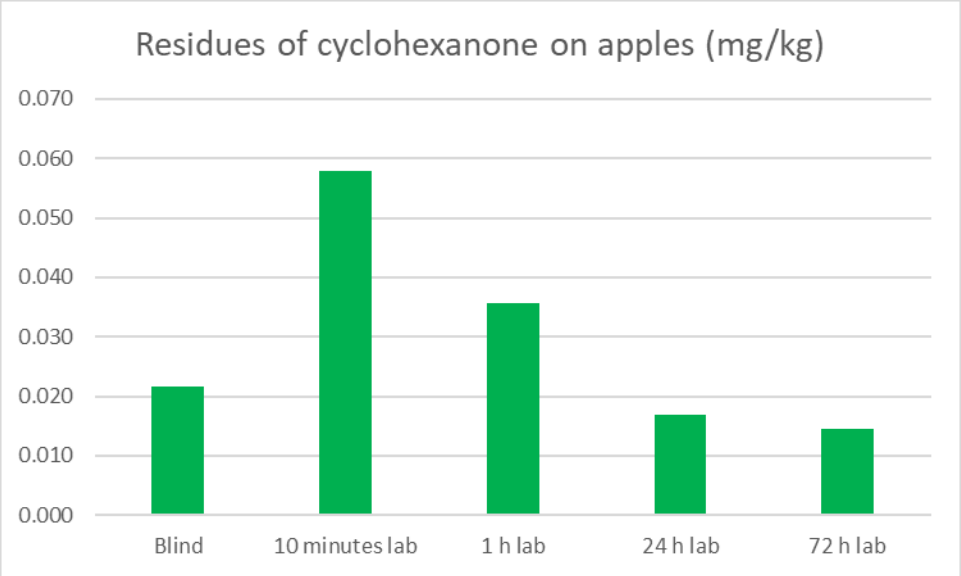


Figure 13: Blank value and residues of cyclohexanone on apples after storage in the laboratory for 10 minutes, 1 hour, 1 day and 3 days. At the time t=0, the apples were immersed once in a spray mixture (0.125% plant protection product Milbeknock).

Table 21: Residues of cyclohexanone on apples in mg/kg.

	Cyclohexanone (mg/kg)	Comments
Blank	0.022	*Single values 0.018; 0.026*
Control	0.013	*Single values 0.018; 0.009*
10 min	0.058	*Single values 0.065; 0.051*
1 h	0.036	*Single values 0.040; 0.032*
24 h	0.017	*Single values 0.018; 0.016*
72 h	0.014	*Single values 0.018; 0.011*

Concentration in the spray mixture: Cyclohexanone 250 mg/L

Immersing the apples in the spray mixture resulted in residues of approx. 0.06 mg/kg; however, these decreased very rapidly (Fig. 13, Table 21). After 24 hours, the measured concentrations were the same as for the control and the blank value, which was relatively high at 0.02 mg/kg and displayed a large fluctuation between the two individual values. The cause of the elevated blank value could not be precisely determined. It was assumed that QuEChERS tube I is responsible for the blank values. In test experiments where QuEChERS tube I and tube II were used for purification, the blank value was slightly higher than the one shown here.

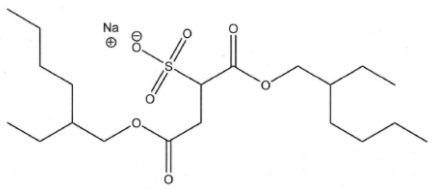
4.4 Co-formulant dioctyl sulfosuccinate sodium salt (surfactant)

4.4.1 Introduction and literature

Dioctyl sulfosuccinate sodium salt is commonly called docusate sodium. For this reason, the short name “docusate” is used for this surfactant in this report. Docusate is used in moderate quantities in plant protection products as a wetting agent. According to our estimates, approximately 1.1 tonnes of this surfactant were sold in Switzerland in 2015 as a constituent of plant protection products [7].

The physicochemical properties of docusate are summarised in Table 22.

Table 22: Designations, structural formula and physicochemical properties of docusate.

Designations	**Docusate** sodium Dioctyl sulfosuccinate sodium salt Di(2-ethylhexyl) sodium sulfosuccinate Sodium bis(2-ethylhexyl)sulfosuccinate Dioctyl sulfosuccinic acid, sodium salt Sodium Bis(2-ethylhexyl)sulfosuccinate Alternative: Potassium and calcium salts
Structural formula	
CAS no.	577-11-7
EC number	209-406-4
Molecular weight (g/mol)	444.56
Molecular formula	$C_{20}H_{37}NaO_7S$
Appearance, properties	White, solid
Melting point	167.5 (165–170) °C
Initial boiling point and boiling range	- (disintegrates at temperatures above 200 °C)
Vapour pressure	1.63 * 10^{-12} Pa at 25 °C (calculated)
Relative density	1.146 at 20 °C
Water solubility	8.17 g/L at 20 °C
Partition coeff.: n-octanol/water log POW:	1.998 at 20 °C

Source: ECHA Registration Dossier[19]

In the PubChem database[20], docusate is described as a universal surfactant, wetting agent and solubiliser for the pharmaceutical, cosmetics and food industries. Docusate is used as a laxative in the pharmaceutical industry and as an additive that acts as an emulsifier and wetting agent in the food industry. Soft drinks can contain up to 10 ppm docusate.

[19] ECHA substance information for docusate sodium: https://echa.europa.eu/de/substance-information/-/substanceinfo/100.008.553

[20] PubChem,Open Chemistry Data base, Compound Summary for CID 23673837, https://pubchem.ncbi.nlm.nih.gov/compound/Docusate_sodium#section=Top

Docusate is used industrially in bonding and sealing chemicals, in adsorption materials, finishing agents, pigments, processing aids, surface-active preparations and in chemicals for adjusting viscosity. The use of docusate has already been discussed in sections 2.2 and 2.3. No further literature on docusate as a co-formulant in PP products or as a residue on harvested produce was found.

According to our assessment, docusate was well-suited to helping us answer the questions posed in this sub-project, since it is a pure substance and has no isomers.

4.4.2 Analytical method

The samples were prepared according to the widely used QuEChERS method [9]. The procedure is described in detail in Appendix 6.2 and 6.6. Measurement was conducted on LC-MS/MS: MRM in positive mode. The parameters of the analytical method for docusate are summarised in the table below.

Table 23: Analytical method for docusate: parameters and procedure.

Extraction agent	Acetonitrile
Purification I (liquid/liquid partitioning, QuEChERS I)	Yes
Purification II (dispersive solid phase, QuEChERS II)	No (Part of the analyte is lost during this purification)
MS MRM quantification	m/z 423.5 → m/z 199.1
MS MRM confirmations	m/z 423.5 → m/z 311.2 m/z 423.5 → m/z 181.0
Calibration (7 points)	2 mg/L to 0.002 mg/L
Correlation coefficient r	0.9988
Limit of Quantification[1]	0.002 mg/L
Blank value	≤ 0.0005 mg/kg
Recovery rate for pepper [2] (8 separate processes)	94.6%
Recovery rate for apple [3] (4 separate processes)	99.7%
Recovery rate for aubergine [4] (1 separate process)	91%
Recovery rate for cucumber [5] (2 separate processes)	97%
Recovery rate for tomato [5] (2 separate processes)	93%
Recovery rate for courgette [5] (2 separate processes)	95%
Expanded uncertainty (Based on laboratory precision, 5-fold determination at a concentration of 0.02 mg/kg, see 6.6)	± 23%

[1] *Limit of Quantification: signal/noise is ≥ 10*

[2] *At concentrations of 0.8 mg/kg (n=2), 0.4 mg/kg (n=3) and 0.2 mg/kg (n=3)*

[3] At concentrations of 0.8 mg/kg (n=2) and 0.2 mg/kg (n=2)

[4] At a concentration of 0.8 mg/kg (n=1)

[5] At concentrations of 0.8 mg/kg (n=1) and 0.2 mg/kg (n=1)

The measurement uncertainty for the determination of docusate was determined by measuring the laboratory precision. The docusate content of the pepper sample of 03/12/2018 was measured five times. The results were used to calculate the relative standard deviation and the expanded uncertainty (see also 6.6 Measurement uncertainty).

Extract II from QuEChERS purification could **not** be used because some of the analyte was lost during purification, resulting in a lower recovery rate (the recovery rate for pepper was 49% (n=8), apple 59% (n=4)). It is assumed that other anionic surfactants are lost during purification with QuEChERS tube II too, since they bind to the solid phase with primary secondary amine that is contained in tube II. For this reason, the extract after purification I (liquid/liquid partitioning, QuEChERS I) was always used for the docusate measurements.

Typical chromatograms are presented in Figure 14 below.

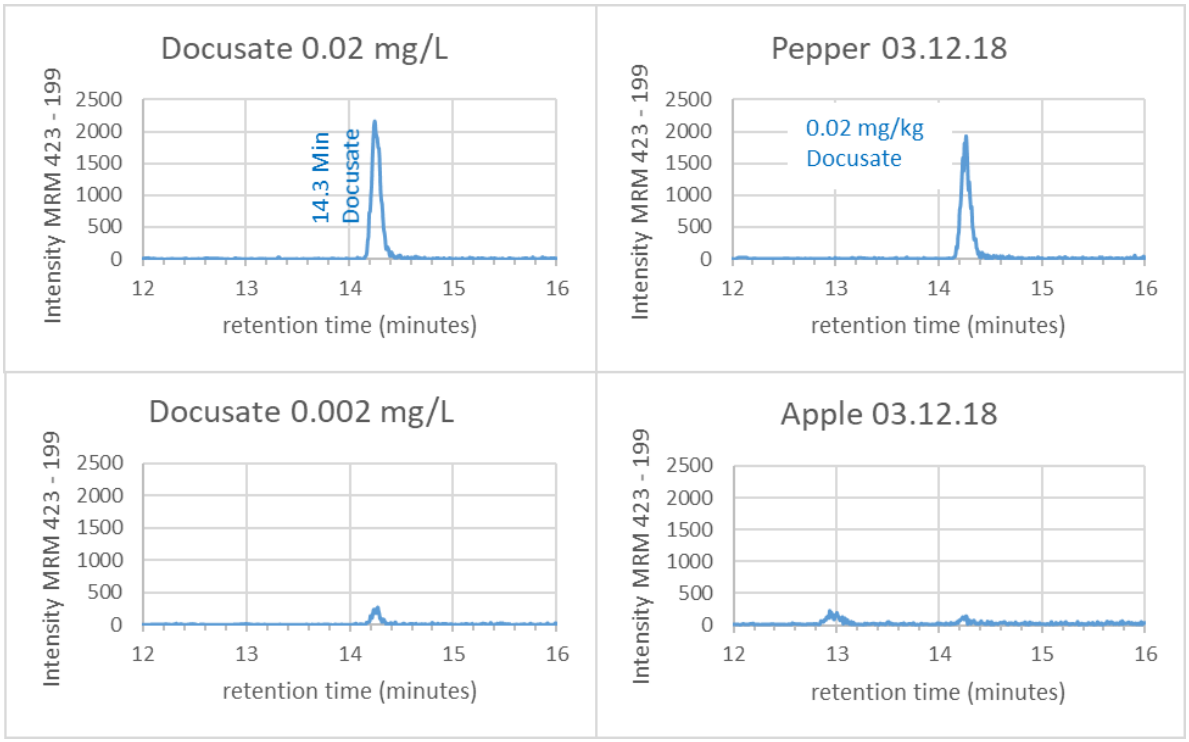


Figure 14: Chromatograms for docusate determination using LC-MS/MS. One standard (0.02 mg/L), one standard at the limit of quantification (0.002 mg/L), one pepper sample with residues (0.02 mg/kg) and one apple sample without residues are shown. Retention time of docusate: 14.3 minutes.

4.4.3 Laboratory tests with pepper and apple

The plant protection product Armicarb (fed. registration number W6432) was selected for the storage experiments. The product contains the active substance potassium bicarbonate in a concentration of 85%. The safety data sheet[21] from the company declares the docusate (dioctyl sodium sulfosuccinate) content to be <15%. Our measurement via LC-MS/MS indicated that the content of the co-formulant docusate in the formulation was 8.9%.

Peppers and apples were purchased from a retail outlet (18 and 8 pieces, respectively, purchased on 03/12/2018). With the exception of the control, the produce was immersed in a spray mixture of the PP product Armicarb with a concentration of 0.3% (only to a height of approx. 80% in the same way as for the other experiments). The concentration of docusate in the spray mixture was therefore 267 mg/L. The treated peppers and apples were left to dry for one hour. The peppers were then stored in an open tray in the laboratory in a fume cupboard for 1 and 3 days.

The results of the residue determinations are presented in Figure 15 and Table 24.

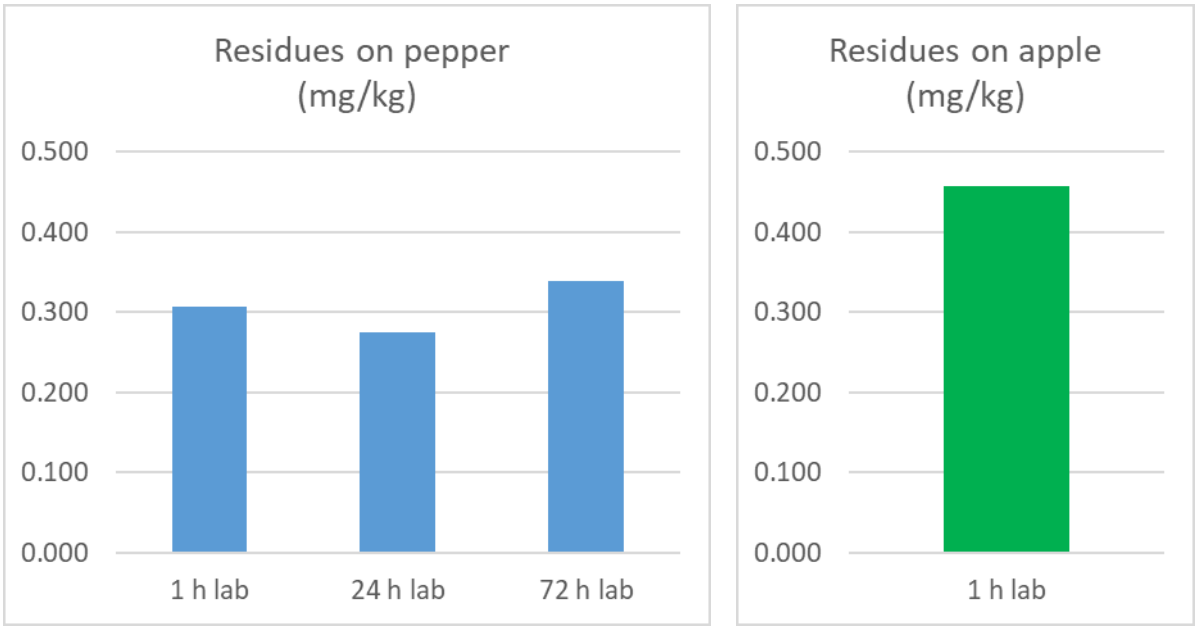


Figure 15: Residues of docusate in mg/kg on pepper (left) and apples (right) after storage in the laboratory for 1 hour, 1 day and 3 days. At the time t=0, the peppers and apples were immersed once in a spray mixture (0.3% plant protection product Armicarb).

[21] MSDS for Armicarb dated 11/07/2016, Stähler, http://www.staehler.ch/de/produkte/info/armicarb.html

Table 24: Residues of docusate on peppers and apples in mg/kg.

	Peppers Docusate (mg/kg)	Apples Docusate (mg/kg)
Blank	< LOQ (0.0006)	< LOQ (0.0006)
Control	0.022	< LOQ (0.001)
1 h	0.308	0.457
24 h lab	0.275	
72 h lab	0.339	

An estimate is provided in parentheses; however, this is below the limit of quantification.
Concentration in the spray mixture: Docusate 267 mg/L

Immersing the **peppers** in the spray mixture resulted in residues of approx. 0.3 mg/kg, which remained roughly constant over the three days. The pepper control sample already exhibited docusate residues of 0.02 mg/kg. This concentration was significantly higher than the blank value for the method (0.0006 mg/kg).

Residues of approximately 0.5 mg/kg were found on the apples. In contrast to the experiment with the peppers, there were no measurable docusate residues on the control sample here. Samples were not examined at later points in time.

4.4.4 Residues on market samples

The control sample (pepper, 03/12/2018) from the laboratory test had docusate residues. For this reason, further vegetables were purchased from a retail outlet on 12/12/2018 in order to investigate whether this was an isolated case or whether other vegetables might also have docusate residues. The pepper control sample of 03/12/2018 was repeated as a double determination. The retention samples for the controls, which had been produced and frozen over the course of the year for this project, were also analysed for docusate residues. The results are shown in Table 25.

Table 25: Docusate residues in mg/kg on varieties of fruit and vegetables from Swiss retail trade.

Produce	Date of purchase	Docusate (mg/kg)	Number of determinations
Aubergine	12/12/2018[1]	0.110	2
Pepper	03/12/2018[1]	0.020	5
Cucumber	12/12/2018[1]	0.010	2
Organic pepper	12/12/2018[1]	0.005	2
Courgette	12/12/2018[1]	0.005	2
Pepper	12/12/2018[1]	0.003	2
Apple	24/07/2018[2]	0.002	2
Apple	25/09/2018[2]	< LOQ	2
Apple	03/12/2018[2]	< LOQ	2
Pepper	24/07/2018[2]	< LOQ	2
Tomatoes	25/09/2018[2]	< LOQ	2
Tomatoes	12/12/2018[1]	< LOQ	2

< LOQ: limit of quantification 0.002 mg/kg
[1] Vegetables from 3 and 12 December 2018 including organic pepper: origin – Spain
[2] Fruit and vegetables from 24 July and 25 September 2018: origin not recorded

Apart from one exception, all vegetable types investigated that were purchased in December 2018 had docusate residues. The highest measured value was 0.11 mg/kg.

However, since the surfactant docusate is not used exclusively in plant protection products, it cannot be concluded with certainty that the residues of this surfactant come from the application of plant protection products. If the results of the storage tests described in section 4.4.3 are considered, it nonetheless appears plausible that the application of PP products contributed to these residues.

4.5 Discussion of the results

In the preceding sections, the residues in the storage tests were presented as a function of storage time. The concentration in the spray liquid was stated for each of the individual co-formulant components. It is evident that there are significant differences in concentration (see overview in Table 4). The concentrations differ by a factor of up to 500. It is therefore unsurprising that the residues on the produce also vary significantly.

Owing to the different concentrations in the spray liquid, it is not possible to compare the residue concentrations directly. The residue values at the earliest sampling time after application (1 hour or 10 minutes) were therefore normalised to a reference concentration of 1 kg/hL (=10 g/L) in the spray liquid (Table 26). The normalised residues vary over a much smaller range of just 2.3–36 mg/kg (a factor of 15), which should be more representative of the differing behaviour of the substances.

Table 26: For the three co-formulants investigated that are used as solvents, the concentration of the residues, the normalised residues and the vapour pressure are stated. Data is also provided for the active substance spiroxamine and the surfactant docusate.

Co-formulant component or active substance	Concentration in the spray mixture (mg/L)	Produce	Residue (mg/kg) after 1 hour or 10 min	Residue (mg/kg) normalised to 1 kg/hL	Vapour pressure (Pa)
Cyclohexanone	250	Apple	0.058	2.3	700
2-Methylnaphthalene	31	Apple	0.023	7.4	3.7
2-Methylnaphthalene	31	Tomato	0.017	5.5	3.7
Dimethyldecanamide	386	Apple	1.385	35.9	0.11
Dimethyldecanamide	386	Pepper	1.29	33.4	0.11
Docusate	267	Apple	0.457	17.1	1.6×10^{-12}
Docusate	267	Pepper	0.308	11.5	1.6×10^{-12}
Spiroxamine	306	Apple	0.780	25.5	$4 / 6 \times 10^{-3}$
Spiroxamine	306	Pepper	0.52	17.0	$4 / 6 \times 10^{-3}$

For the solvents, there was a correlation between the standardised residues and the vapour pressure (see Fig. 3):

- Cyclohexanone, the most volatile of the solvents investigated, was found on the produce despite its high volatility and did not evaporate when the spray mixture was produced. However, the storage test showed that the concentration decreased rapidly on the apple. This decrease undoubtedly begins within the first 10 minutes after treatment. However, this phase cannot be recorded experimentally because the spray liquid must first dry before the apples can be prepared and frozen.

- 2-Methylnaphthalene, the second most volatile of the solvents investigated (which were present in the spray mixture in a sufficient concentration as to be subsequently detectable in the produce), resulted in normalised residues that were approximately 3 times as high as those of cyclohexanone, even though the absolute residues were lower. The storage test indicated a continuous decrease in the concentration of 2-methylnaphthalene on the apple. This decrease is much slower than in cyclohexanone. The co-formulant component 2-methylnaphthalene makes up

approx. 10% of solvent naphtha 200 ND. In order to estimate the overall residues of all components, the residue of 2-methylnaphthalene must be multiplied by at least a factor of 10. The gas chromatogram for solvent naphtha shows that the other co-formulant components are less volatile than 2-methylnaphthalene and must therefore remain on the produce for longer in practice.

- N,N-Dimethyldecanamide has the lowest volatility of the three solvents tested. The normalised residues were on the same order of magnitude as those of the active substance spiroxamine and the surfactant docusate. In the case of dimethyldecanamide, evaporation can only have had a low impact on the amount of residues after one hour. This is also confirmed by the slow decrease over the 3-day storage period.

Biphenyl, which was sporadically confirmed in food monitoring programmes, could not be verified in the laboratory tests, possibly due to the very low concentrations in the product. In this project, the products were only applied once. Although the residues could accumulate in the case of multiple applications, this would primarily only happen if the substances settle in hydrophobic compartments of the produce. Nonetheless, on the basis of the present results, it would appear that solvent naphtha can be ruled out as the cause of the presence of biphenyl in food products.

High blank values were found for methylnaphthalene and cyclohexanone. This demonstrates that it can be very difficult to reliably determine widely used substances in trace quantities. To date, only these two solvents have been affected, but expanding the range of substances to include other co-formulants may well bring other cases to light.

In the laboratory experiment, the anionic surfactant docusate left residues that could still be measured over 3 days and that did not change significantly in terms of quantity over time. In the case of docusate, verifiable residues in the control sample could not be attributed to blank values. Exploratory market monitoring conducted as a result of this indicated that almost all vegetable samples purchased in December 2018 had residues of docusate. However, the majority of these residues were below or in the range of 0.01 mg/kg. When determining pesticide residues with multi-methods, this concentration is often the limit of quantification and is defined as the default maximum residue limit (MRL) in the EU, unless higher limits are necessary for the permitted application of plant protection products. However, it is not possible to conclude with certainty that the residues measured here are the result of treatment with plant protection products, since other sources are also conceivable.

Overall, it is clear that co-formulants can leave residues on foods of plant origin. Initial field tests will indicate whether measurable residues can also occur on harvested produce under practical conditions when plant protection products are used.

5 Bibliography

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6 Appendix

6.1 Materials, equipment

1-Methylnaphthalene	1-Methylnaphthalene, 96%, Acros Organics, Catalogue Number 12716, Lot A0388530
2-Methylnaphthalene	2-Methylnaphthalene, 96%, Acros Organics, Catalogue Number 12717, Lot A0371695
Acetonitrile	HPLC gradient grade, VWR, HiPerSolv Chromanorm
Ammonium acetate	puriss p.a., ACS reagent, Fluka, No. 101174715
Ammonium formate	99%, Acros Organics, No. 401152500
Biphenyl	Biphenyl, 99%, Acros Organics, Catalogue Number 10625, Lot A0382502
Cyclohexanone	Cyclohexanone, ≥ 99.5%, Sigma-Aldrich, Product Number: 29140, Lot STBD0350V
Difenoconazole	Difenoconazole, Pestanal, Sigma-Aldrich, Product No. 36531, Lot SZBF205XV
Docusate	Dioctyl sulfosuccinate, sodium salt, 96%, Acros Organics, Catalog Number 11710, Lot A0394647
Ethyl acetate	For residue analysis, Fluka
GC-MS/MS	Agilent 6890N, Combi PAL, Quattro Micro GC-MS/MS
LC-MS/MS	Agilent 1100 series (high pressure gradient), LC PAL, API 4000
Knife mill	Grindomix GM 300, Retsch, Haan, Germany
Methanol	HPLC gradient grade, VWR, HiPerSolv Chromanorm
Microcentrifuge	UniCFUGE 3 LLG Labware
N,N-Dimethyldecanamide	N,N-Dimethyldecanamide, 98%, Alfa Aesar, Product No. L08731, Lot 10174436
Plant protection product A	Input with 38.6% DMDA and 30.5% spiroxamine
Plant protection product B	Slick with 50–70% solvent naphtha (of which 6.2% 2methylnaphthalene; 3.1% 1-methylnaphthalene; 0.16% biphenyl) and 23.5% difenoconazole
Plant protection product C	Milbenknock with 20% cyclohexanone and 1% milbemectin
Plant protection product D	Armicarb with 8.9% docusate and 85% potassium bicarbonate
Spiroxamine	Bayer, Lot: M00298, 98.3%, internal lab number W2024
Freezer	UNI 21 (to -45 °C)
GC-MS separation column	DB 5 MS, 30 m x 0.32 mm x 0.25 µm,
LC-MS/MS separation column	Gemini-NX 5µ C18 110A, 150 mm x 2.0 mm, Phenomenex No. 007-4454-B0) with precolumn Gemini-NX 5u C18 110A, 4 mm x 2.0 mm, Security Guard Cartridges, Phenomenex No. AJ0-8367)
Tube I QuEChERS	DisQuE 50mL tube/AOAC – acetate, Waters, No. 186004571 (QuEChERS, 1.5 g sodium acetate and 6 g MgSO4)
Tube II QuEChERS	DisQuE 2ml tube – AOAC, Waters, No. 186004572 (QuEChERS, 150mg MgSO4 and 50 mg PSA)
Centrifuge	Eppendorf tabletop centrifuge 25804

6.2 Sample preparation

Harvested produce was purchased from a retail outlet and either prepared directly as described below or dipped in a spray mixture at the time t=0 for the storage tests.
To do this, a beaker was filled with 600 mL tap water and positioned on a magnetic stirrer in a fume cupboard, and a magnetic stirring rod was added. The corresponding quantity of plant protection product was then weighed and added to the beaker. A homogeneous spray mixture was produced by stirring.
For the individual storage tests, the following quantities were weighed and added:

4.1.3	Input W6392	Concentration 0.1%	600 mg Input in 600 ml water
4.2.3	Slick W5056	Concentration 0.05%	300 mg Slick in 600 ml water
4.3.3	Milbeknock W6526	Concentration 0.125%	750 mg Milbeknock in 600 ml water
4.4.3	Armicarb W6432	Concentration 0.3%	1800 mg Armicarb in 600 ml water

The spray mixture was stirred further in order to guarantee a uniform distribution. The harvested products (apples, peppers, tomatoes) were held by their stalk with tweezers and immersed in the “spray mixture” up to approx. 80% of their height. Complete immersion was avoided so that no spray mixture could run into the depression at the base of the stalk, which would lead to locally excessive and non-reproducible residues. The produce treated in this way was then dried in open trays, sampled, and stored for up to 3 days in the laboratory or outdoors.

Samples were taken at various times (4–6 whole fruits in each case) and cut into pieces of approx. 4 cm using a knife in order to facilitate homogenisation later on (inedible parts, such as the stalk, were removed beforehand). They were then placed in a zip lock bag that was firmly sealed. The sample was then frozen at -45 °C at least overnight. The frozen sample was placed in the knife mill and milled while frozen
(10 seconds 500 rpm; 20 seconds 1000 rpm; 70 seconds 1500 rpm; direction of rotation (knife cutting)). The homogenate was still frozen after milling. For each analysis, 10 g (± 0.1 g) of homogenate were weighed into a 50 mL centrifuge tube made of polypropylene (caution: work quickly so that the homogenate does not thaw). The rest of the homogenate was kept as retention samples (in a polyethylene box, PEHD) and stored at -18 °C. The centrifuge tubes containing the weighed samples were also stored at -18 °C until they were used for processing and analysis.

Processing was based on the QuEChERS multi-method for pesticides [9], which was modified depending on the analyte and analytical equipment (ethyl acetate or acetonitrile as extraction agent, skipping purification and direct use of extract I). No internal standards were used.

Processing sequence:

- Leave centrifuge tubes containing samples to thaw at room temperature for approx. 1 hour
- Add 10 mL solvent
  Acetonitrile for dimethyldecanamide, spiroxamine and docusate → HPLC;
  Ethyl acetate for methylnaphthalene, biphenyl and cyclohexane → GC;
- Shake for 1 min
- Add salt from pouch QuEChERS I (DisQuE 50mL tube/AOAC – acetate QuEChERS)
- Shake for 1 min

- Centrifuge for 5 min 3500 rpm
  ➔ **Extract I** vials for GC (cyclohexanone determination) or for HPLC (docusate determination)
- Add 1 mL extract I to QuEChERS tube II (DisQuE 2ml tube – AOAC)
  Shake for 1 min
- Centrifuge with microcentrifuge: 1 min 6000 rpm
- Use pipette to dispense 600 ul of supernatant into vial
  ➔ **Extract II** vials for GC (methylnaphthalene and biphenyl determination) or for HPLC (dimethyl-decanamide and spiroxamine determination)

6.3 Description of method for dimethyldecanamide and spiroxamine

The samples were prepared as described in section 6.2, using **acetonitrile** as the extraction agent. Purification was conducted up to **extract II**, which was then poured into the vials for LC-MS/MS.
The chromatographic determination of the analytes was conducted as follows: HPLC system Agilent 1100, autosampler HTS PAL (CTC, Zwingen), no column thermostat (air-conditioned laboratory, 22 °C). The separation column (Gemini-NX 5µm C18 110A, 150 mm x 2.0 mm) was connected to an API 4000 triple quadrupole mass spectrometer (Sciex, Framingham, MA) with a turbo ion spray (TIS) source in positive mode.
Eluent flow 0.2 mL/min, high pressure gradient:

0 min	25% 5mM ammonium acetate / 75% methanol
4 min	5% 5mM ammonium acetate / 95% methanol
9 min	5% 5mM ammonium acetate / 95% methanol
9.1 min	25% 5mM ammonium acetate / 75% methanol
10.6 min	25% 5mM ammonium acetate / 75% methanol

Retention time of dimethyldecanamide: 5.32 min; spiroxamine 4.32 min

Mass spectrometer, MRM positive, (EP 10; CAD 4; CUR 10; GS1 20; GS2 30; IS 4200; Temp 400)

Analyte	MRM m/z	Time ms	DP	CE	CXP
Dimethyldecanamide	200.2 -> 102.2	200	80	30	10
Dimethyldecanamide confirmation	200.2 -> 116.1	200	80	28	10
Spiroxamine	298.3 -> 100.1	200	95	42	16
Spiroxamine confirmation	298.3 -> 144.1	200	95	27	7

The chromatograms of the first (more intensive) mass transfer were evaluated using the software Analyst based on peak areas and external calibration.

Measurement uncertainty
The expanded uncertainty for the determination of dimethyldecanamide and spiroxamine was not determined by measuring the laboratory precision but was instead estimated. The individual values of the double determinations from the storage test were taken into consideration:

Äpfel		Serie 180831	Serie 180830				
Analyt		mg/kg	mg/kg	MW	SD	RSD	2 * RSD
DMDA	72hDach	0.232	0.272	0.252	0.028	11%	**22%**
Spiroxamin	72hDach	0.460	0.549	0.505	0.063	12%	**25%**
DMDA	72hLab	0.450	0.517	0.484	0.047	10%	**20%**
Spiroxamin	72hLab	0.619	0.712	0.666	0.066	10%	**20%**
DMDA	24hDach	0.551	0.552	0.552	0.001	0%	0%
Spiroxamin	24hDach	0.573	0.576	0.575	0.002	0%	1%
DMDA	24hLab	1.430	1.300	1.365	0.092	7%	13%
Spiroxamin	24hLab	0.941	1.070	1.006	0.091	9%	18%
DMDA	1h	1.450	1.320	1.385	0.092	7%	13%
Spiroxamin	1h	0.795	0.765	0.780	0.021	3%	5%
					MW	7%	14%

and

Peperoni		Serie 180831	Serie 180830				
Analyt		mg/kg	mg/kg	MW	SD	RSD	2 * RSD
DMDA	72hDach	0.496	0.474	0.485	0.016	3%	6%
Spiroxamin	72hDach	0.534	0.528	0.531	0.004	1%	2%
DMDA	72hLab	0.484	0.435	0.460	0.035	8%	15%
Spiroxamin	72hLab	0.550	0.541	0.546	0.006	1%	2%
DMDA	24hDach	0.652	0.665	0.659	0.009	1%	3%
Spiroxamin	24hDach	0.509	0.530	0.520	0.015	3%	6%
DMDA	24hLab	0.744	0.696	0.720	0.034	5%	9%
Spiroxamin	24hLab	0.484	0.489	0.487	0.004	1%	1%
DMDA	1h	1.310	1.270	1.290	0.028	2%	4%
Spiroxamin	1h	0.496	0.540	0.518	0.031	6%	12%
					MW	3%	6%

The relative standard deviation (RSD) of the double determinations was multiplied by expansion factor 2. The expanded uncertainty was derived from the highest values (bold) and was estimated to be **20%** after rounding.

6.4 Description of method for methylnaphthalene and biphenyl

The samples were prepared as described in section 6.2, using **ethyl acetate** as the extraction agent. Purification was conducted up to **extract II**, which was then poured into the vials for GC-MS.

The chromatographic determination of the analytes was conducted as follows: GC-MS instrument: Agilent 6890N (Santa Clara, CA), COMBI PAL autosampler (CTC Analytics, Zwingen, Switzerland).

The GC was coupled to a Quattro Micro triple quadrupole mass spectrometer (Micromass, Manchester, UK) with electron impact ionisation (70 eV, 200 °C) in single ion monitoring mode.

Separation column: DB 5 MS column (5% phenyl-, 95% methylpolysiloxane, 30 m, 0.32 mm i.d., 0.25 µm film), with retention gap (1m, 0.53 mm i.d.)

GC interface at 250 °C; 1 µL split/splitless injection (280 °C, initial 60 s splitless)

Temperature program, 60 °C, 3 min isotherm, with 10 °C/min up to 300 °C, 6 min isotherm at 300 °C; run time 33 minutes; constant flow, helium: 4 mL/min.
MS conditions:
SIM, solvent delay 3 min; acquisition 3.1 to 33 min;

m/z 142.1, dwell 0.08 sec	2-methylnaphthalene	t_R 9.27 min and
	1-methylnaphthalene	t_R 9.48 min
m/z 154.1, dwell 0.08 sec	biphenyl	t_R 10.44 min
m/z 323.3, dwell 0.08 sec	difenoconazole	t_R 25.28 and 25.35 min

The chromatograms were evaluated using the software QuanLynx based on peak areas and external calibration.

6.5 Description of method for cyclohexanone

The samples were prepared as described in section 6.2, using **ethyl acetate** as the extraction agent. Purification was conducted up to **extract I**, which was then transferred into the vials for GC-MS.
The chromatographic determination of the analytes was conducted as follows: GC-MS instrument: Agilent 6890N (Santa Clara, CA), COMBI PAL autosampler (CTC Analytics, Zwingen, Switzerland).
The GC was coupled to a Quattro Micro triple quadrupole mass spectrometer (Micromass, Manchester, UK) with electron impact ionisation (70 eV, 200 °C) in single ion monitoring mode.
Separation column: DB 5 MS column (5% phenyl-, 95% methylpolysiloxane, 30 m, 0.32 mm i.d., 0.25 µm film), with retention gap (1m, 0.53 mm i.d.)
GC interface at 250 °C; 1 µL split/splitless injection (280 °C, initial 60 s splitless)

Temperatur program, 40 °C, 4 min isotherm, with 10 °C/min up to 80 °C, with 50 °C/min up to 300 °C, 1.6 min isotherm at 300 °C; run time 14 minutes
Constant flow, helium: 4 mL/min.
MS conditions:
SIM, solvent delay 3 min; acquisition 3.1 to 33 min;

m/z 98, Dwell 0.1 sec	cyclohexanone	t_R 5.18 min
m/z 55, Dwell 0.1 sec	cyclohexanone confirmation	t_R 5.18 min

The chromatograms were evaluated using the software QuanLynx based on peak areas and external calibration.

6.6 Description of method for docusate

The samples were prepared as described in section 6.2, using **acetonitrile** as the extraction agent. Purification was conducted up to **extract I**, which was poured into the vials for LC-MS/MS.
The chromatographic determination of the analytes was conducted as follows: HPLC system Agilent 1100 series, autosampler HTS PAL (CTC, Zwingen), no column thermostat (air-conditioned laboratory, 22 °C). The separation column (Gemini-NX 5µm C18 110A, 150 mm x 2.0 mm) was connected to an API 4000 triple quadrupole mass spectrometer (Sciex, Framingham, MA) with a turbo ion spray (TIS) source in positive mode.
Eluent flow 0.2 mL/min, high pressure gradient:

HPLC

0.0 min	50%	5mM ammonium formate /	50% methanol
15.0 min	0%	5mM ammonium formate /	100% methanol
19.0 min	0%	5mM ammonium formate /	100% methanol
19.1 min	50%	5mM ammonium formate /	50% methanol
22.0 min	50%	5mM ammonium formate /	50% methanol

Retention time of docusate: 14.3 min

MS/MS, MRM positive, (EP 10; CAD 4; CUR 10; GS1 20; GS2 20; IS 5000; Temp 400)

Analyte	MRM m/z	Time ms	DP	CE	CXP
Docusate	423.5 -> 199.1	150	81	13	12
Docusate confirmation	423.5 -> 311.2	150	81	9	20
Docusate confirmation 2	423.5 -> 181.0	150	81	27	30

The chromatograms of the first MRM were evaluated using the software Analyst based on peak areas and external calibration.

Measurement uncertainty

The expanded uncertainty for the determination of docusate was determined by measuring the laboratory precision. The docusate content of the pepper sample of 03/12/2018 was measured five times (2 series on 11/12/2018 and 13/12/2018 respectively):

Pepper from 03/12/2018	No.	Docusate (mg/kg)
Series 181211a	P1a	0.0227
Series 181211b	P1b	0.0211
Series 181211a	P1c	0.0212
Series 181213a	P1d	0.0178
Series 181213b	P1e	0.0176
Mean		0.0201
Relative standard deviation (RSD)		11.3%
Expanded uncertainty (2 * RSD)		**23%**

The relative standard deviation of the 5-fold determination is multiplied by expansion factor 2. This gives an expanded measurement uncertainty of 23%.

6.7 Table overview of results (single values)

The single values and the coding in the sequences for the measurements conducted are shown here. For all data points specified, double determination was performed (in two different sequences). Calibration and quantification were carried out directly with the device programs Analyst (LC-MS/MS) or QuanLynx (GC-MS).

*Table 27: Residues of **DMDA** in mg/kg; series LC-MS/MS for 180731a and 180731b; stored from 24/07/18 to 27/07/18*

	Code	Series 180731a	Code	Series 180731b	Mean
Blank	Blank1	< LOQ	Blankb	< LOQ	
Pepper					
Control	P1a contr	< LOQ	P1b contr	< LOQ	
1 h	P6a10 1h	1.31	P6b10 1h	1.27	1.29
24 h lab	P5a 24h lab	0.744	P5b 24h lab	0.696	0.72
24 h roof	P4a 24h roof	0.652	P4b 24h roof	0.665	0.66
72 h lab	P3a 72h lab	0.484	P3b 72h lab	0.435	0.46
72 h roof	P2a 72h roof	0.496	P2b 72h roof	0.474	0.49
Apple					
Control	Ap1a contr	< LOQ	Ap1b contr	< LOQ	
1 h	A6a10 1h	1.45	A6b10 1h	1.32	1.39
24 h lab	A5a10 24h lab	1.43	A5b10 24h lab	1.3	1.37
24 h roof	A4a 24h roof	0.551	A4b 24h roof	0.552	0.55
72 h lab	A3a 72h lab	0.45	A3b 72h lab	0.517	0.48
72 h roof	A2a 72h roof	0.232	A2b 72h roof	0.272	0.25

*Table 28: Residues of **spiroxamine** in mg/kg; series LC-MS/MS for 180731a and 180731b; stored from 24/07/18 to 27/07/18*

	Code	Series 180731a	Code	Series 180731b	Mean
Blank	Blank1	< LOQ	Blankb	< LOQ	
Pepper					
Control	P1a contr	< LOQ	P1b contr	< LOQ	
1 h	P6a10 1h	0.496	P6b10 1h	0.54	0.52
24 h lab	P5a 24h lab	0.484	P5b 24h lab	0.489	0.49
24 h roof	P4a 24h roof	0.509	P4b 24h roof	0.53	0.52
72 h lab	P3a 72h lab	0.55	P3b 72h lab	0.541	0.55
72 h roof	P2a 72h roof	0.534	P2b 72h roof	0.528	0.53
Apple					
Control	Ap1a contr	< LOQ	Ap1b contr	< LOQ	
1 h	A6a10 1h	0.795	A6b10 1h	0.765	0.78
24 h lab	A5a10 24h lab	0.941	A5b10 24h lab	1.07	1.01
24 h roof	A4a 24h roof	0.573	A4b 24h roof	0.576	0.57
72 h lab	A3a 72h lab	0.619	A3b 72h lab	0.712	0.67
72 h roof	A2a 72h roof	0.460	A2b 72h roof	0.549	0.50

*Table 29: Residues of **2-methylnaphthalene** in mg/kg; series GC-MS for 181106 and 181108; stored from 25/09/18 to 28/09/18*

	Code	Series 181106	Code	Series 181108	Mean
Blank	Blank1	0.0032	Blank2	0.0090	0.0061
Apple					
Control	A1a	0.0035	A1b	0.0083	0.0059
1 h	A6a	0.0224	A6b	0.0235	0.0230
24 h lab	A5a	0.0060	A5b	0.0139	0.0100
24 h roof	A4a	0.0077	A4b	0.0158	0.0118
72 h lab	A3a	0.0041	A3b	0.0115	0.0078
72 h roof	A2a	0.0048	A2b	0.0132	0.0090
Tomatoes					
Control	T1a	0.0034	T1b	0.0035	0.0035
1 h	T6a	0.0171	T6b	0.0174	0.0173
24 h lab	T5a	0.0056	T5b	0.0134	0.0095
24 h roof	T4a	0.0058	T4b	0.0095	0.0077
72 h lab	T3a	0.0051	T3b	0.0108	0.0080
72 h roof	T2a	0.0043	T2b	0.0083	0.0063

*Table 30: Residues of **1-methylnaphthalene** in mg/kg; series GC-MS for 181106 and 181108; stored from 25/09/18 to 28/09/18*

	Code	Series 181106	Code	Series 181108	Mean
Blank	Blank1	0.0016	Blank2	0.0039	0.0028
Apple					
Control	A1a	0.0024	A1b	0.0041	0.0033
1 h	A6a	0.0090	A6b	0.0096	0.0093
24 h lab	A5a	0.0033	A5b	0.0069	0.0051
24 h roof	A4a	0.0041	A4b	0.0073	0.0057
72 h lab	A3a	0.0022	A3b	0.0055	0.0039
72 h roof	A2a	0.0024	A2b	0.0063	0.0044
Tomatoes					
Control	T1a	0.0020	T1b	0.0025	0.0023
1 h	T6a	0.0081	T6b	0.0081	0.0081
24 h lab	T5a	0.0039	T5b	0.0062	0.0051
24 h roof	T4a	0.0034	T4b	0.0053	0.0044
72 h lab	T3a	0.0028	T3b	0.0053	0.0041
72 h roof	T2a	0.0024	T2b	0.0040	0.0032

*Table 31: Residues of **biphenyl** in mg/kg; series GC-MS for 181106 and 181108; stored from 25/09/18 to 28/09/18*

	Code	Series 181106	Code	Series 181108	Mean
Blank	Blank1	0.0010	Blank2	0.0014	0.0012
Apple					
Control	A1a	0.0012	A1b	0.0015	0.0014
1 h	A6a	0.0021	A6b	0.0025	0.0023
24 h lab	A5a	0.0016	A5b	0.0028	0.0022
24 h roof	A4a	0.0015	A4b	0.0026	0.0021
72 h lab	A3a	0.0015	A3b	0.0024	0.0020
72 h roof	A2a	0.0016	A2b	0.0024	0.0020
Tomatoes					
Control	T1a	0.0014	T1b	0.0015	0.0015
1 h	T6a	0.0017	T6b	0.0017	0.0017
24 h lab	T5a	0.0014	T5b	0.0022	0.0018
24 h roof	T4a	0.0015	T4b	0.0021	0.0018
72 h lab	T3a	0.0016	T3b	0.0017	0.0017
72 h roof	T2a	0.0016	T2b	0.0017	0.0017

*Table 32: Residues of **cyclohexanone** on apple in mg/kg; series GC-MS for 181220 and 181221; stored from 3/12/18 to 6/12/18*

	Code	Series 181220	Code	Series 181221	Mean
Blank	Blanka	0.0176	Blankb	0.0257	0.022
Control	A1a	0.0179	A1b	0.0088	0.013
10 min	A5a	0.0654	A5b	0.0505	0.058
1 h	A4a	0.04	A4b	0.0315	0.036
24 h lab	A3a	0.018	A3b	0.0157	0.017
72 h lab	A2a	0.0178	A2b	0.0111	0.014

*Table 33: Residues of **docusate** in mg/kg; series LC-MSMS for 181211a and b; stored from 3/12/18 to 6/12/18*

	Code	Series 181211a	Code	Series 181211b	Mean
Blank	Blank1	0.00085	Blank2	0.000214	0.0006
Pepper					
Control	P1a	0.0227	P1b	0.0211	0.022
1 h	P4a	0.29	P4b	0.325	0.308
24 h lab	P3a	0.279	P3b	0.271	0.275
72 h lab	P2a	0.347	P2b	0.33	0.339
Control	P1c	0.0212			
Apple					
Control	AD1a	0.00106	AD1b	0.000923	0.001
1 h	A6a	0.454	A6b	0.46	0.457

*Table 34: Residues of **docusate** in mg/kg; series LC-MSMS for 181213a and b; purchased on 12/12/2018 and retention samples*

	Code	Series 181213a	Code	Series 1812113b	Mean
Blank	Blanka	0.000383	Blankb	0.000277	0.0003
Apples July 18	A07a	0.00185	A07b	0.00234	0.0021
Peppers July 18	P07a	0.000824	P07b	0.00069	0.0008
Apples Sept 18	A09a	0.000868	A09b	0.000616	0.0007
Tomatoes Sept 18	T09a	0.000377	T09b	0.000559	0.0005
Peppers 03/12/18	P1d	0.0178	P1e	0.0176	0.0177
Organic peppers 12/12/18	PB12a	0.00495	PB12b	0.00562	0.0053
Peppers 12/12/18	P12a	0.00343	P12b	0.00347	0.0035
Tomatoes 12/12/18	T12a	0.00173	T12b	0.00167	0.0017
Courgettes 12/12/18	Z12a	0.00459	Z12b	0.00455	0.0046
Aubergines 12/12/18	Au12a	0.112	Au12b	0.107	0.1095
Cucumbers 12/12/18	G12a	0.0098	G12b	0.0101	0.0100

6.8 Table overview of weather data

Table 35: Weather data during storage from 24 to 27/7/2018 from Agrometeo for the fruit growing station in Wädenswil. The weather station is directly adjacent to the laboratory building.

Date, time	Mean temperature (ºC)	Precipitation (mm)	Global radiation (WH/m2)
24/07/2018 15:00	30	0	344
24/07/2018 16:00	30.2	0	308
24/07/2018 17:00	29.3	0	99
24/07/2018 18:00	27.8	0	30
24/07/2018 19:00	25.6	0	2
24/07/2018 20:00	24.2	0	0
24/07/2018 21:00	23	0	0
24/07/2018 22:00	21.9	0	0
24/07/2018 23:00	20.5	0	0
25/07/2018 00:00	20	0	0
25/07/2018 01:00	20.2	0	0
25/07/2018 02:00	20.3	0	0
25/07/2018 03:00	18.9	0	0
25/07/2018 04:00	18.6	0	18
25/07/2018 05:00	20.9	0	116
25/07/2018 06:00	22.7	0	274
25/07/2018 07:00	23.8	0	437
25/07/2018 08:00	25.1	0	583
25/07/2018 09:00	26.5	0	699
25/07/2018 10:00	27.9	0	784
25/07/2018 11:00	29.1	0	802
25/07/2018 12:00	29.8	0	785
25/07/2018 13:00	30.5	0	727
25/07/2018 14:00	31	0	625
25/07/2018 15:00	31.2	0	489
25/07/2018 16:00	31.2	0	328
25/07/2018 17:00	29.6	0	92
25/07/2018 18:00	28	0	21
25/07/2018 19:00	25	0	2
25/07/2018 20:00	23.3	0	0
25/07/2018 21:00	23.2	0	0
25/07/2018 22:00	22.6	0	0
25/07/2018 23:00	22.5	0	0
26/07/2018 00:00	21.2	0	0
26/07/2018 01:00	20.3	0	0
26/07/2018 02:00	19.2	0	0
26/07/2018 03:00	18.7	0	0
26/07/2018 04:00	18.6	0	16
26/07/2018 05:00	21.7	0	130
26/07/2018 06:00	23.2	0	299
26/07/2018 07:00	23.8	0	459
26/07/2018 08:00	24.8	0	601
26/07/2018 09:00	26.2	0	708
26/07/2018 10:00	28.7	0	782
26/07/2018 11:00	30.4	0	792
26/07/2018 12:00	30.8	0	708
26/07/2018 13:00	31.1	0	720
26/07/2018 14:00	31.4	0	616
26/07/2018 15:00	31.3	0	477
26/07/2018 16:00	30.7	0	323
26/07/2018 17:00	29.2	0	86
26/07/2018 18:00	27.7	0	17
26/07/2018 19:00	25.6	0	2
26/07/2018 20:00	23.4	0	0
26/07/2018 21:00	22.4	0	0
26/07/2018 22:00	22.5	0	0
26/07/2018 23:00	22.1	0	0
27/07/2018 00:00	19.7	0	0
27/07/2018 01:00	19	0	0
27/07/2018 02:00	18.4	0	0

Date, time	Mean temperature (ºC)	Precipitation (mm)	Global radiation (WH/m2)
27/07/2018 03:00	17.6	0	0
27/07/2018 04:00	17.9	0	15
27/07/2018 05:00	20.3	0	132
27/07/2018 06:00	22.7	0	305
27/07/2018 07:00	23.5	0	466
27/07/2018 08:00	24.2	0	608
27/07/2018 09:00	25.4	0	716
27/07/2018 10:00	27.2	0	787
27/07/2018 11:00	28.5	0	812
27/07/2018 12:00	29.3	0	783
27/07/2018 13:00	30.4	0	714
27/07/2018 14:00	31	0	614
27/07/2018 15:00	31.6	0	485
Mean	**25.0**	**0**	**284**
Maximum	31.6	0	812
Minimum	17.6	0	0

Table 36: Weather data during storage from 25 to 28/9/2018 from Agrometeo for the fruit growing station in Wädenswil. The weather station is directly adjacent to the laboratory building.

Date, time	Mean temperature (ºC)	Precipitation (mm)	Global radiation (WH/m2)
25/09/2018 11:00	14.8	0	634
25/09/2018 12:00	15.5	0	595
25/09/2018 13:00	15.5	0	506
25/09/2018 14:00	15.5	0	380
25/09/2018 15:00	15.1	0	231
25/09/2018 16:00	14.1	0	77
25/09/2018 17:00	12.7	0	4
25/09/2018 18:00	11.6	0	0
25/09/2018 19:00	10.9	0	0
25/09/2018 20:00	10.2	0	0
25/09/2018 21:00	8.9	0	0
25/09/2018 22:00	7.4	0	0
25/09/2018 23:00	7.4	0	0
26/09/2018 00:00	6.1	0	0
26/09/2018 01:00	5.8	0	0
26/09/2018 02:00	5.6	0	0
26/09/2018 03:00	5.6	0	0
26/09/2018 04:00	5	0	0
26/09/2018 05:00	4.5	0	5
26/09/2018 06:00	6.5	0	59
26/09/2018 07:00	7.7	0	63
26/09/2018 08:00	8.9	0	150
26/09/2018 09:00	11.1	0	539
26/09/2018 10:00	12.6	0	605
26/09/2018 11:00	13.8	0	621
26/09/2018 12:00	15.6	0	558
26/09/2018 13:00	17.1	0	471
26/09/2018 14:00	18.1	0	370
26/09/2018 15:00	18.2	0	214
26/09/2018 16:00	17	0	73
26/09/2018 17:00	14.3	0	7
26/09/2018 18:00	13.4	0	0
26/09/2018 19:00	12.3	0	0
26/09/2018 20:00	12.2	0	0
26/09/2018 21:00	11.4	0	0
26/09/2018 22:00	10.8	0	0
26/09/2018 23:00	10.3	0	0
27/09/2018 00:00	9.8	0	0
27/09/2018 01:00	9.9	0	0
27/09/2018 02:00	9.2	0	0
27/09/2018 03:00	9.2	0	0

Date, time	Mean temperature (ºC)	Precipitation (mm)	Global radiation (WH/m2)
27/09/2018 04:00	9.3	0	0
27/09/2018 05:00	9.2	0	5
27/09/2018 06:00	10.8	0	55
27/09/2018 07:00	12.3	0	38
27/09/2018 08:00	13.3	0	157
27/09/2018 09:00	15.2	0	527
27/09/2018 10:00	16.8	0	593
27/09/2018 11:00	18.4	0	607
27/09/2018 12:00	20	0	571
27/09/2018 13:00	21	0	488
27/09/2018 14:00	21.9	0	368
27/09/2018 15:00	21.6	0	215
27/09/2018 16:00	19.9	0	65
27/09/2018 17:00	17.4	0	3
27/09/2018 18:00	15.8	0	0
27/09/2018 19:00	13.6	0	0
27/09/2018 20:00	13.2	0	0
27/09/2018 21:00	12.4	0	0
27/09/2018 22:00	12.4	0	0
27/09/2018 23:00	11.5	0	0
28/09/2018 00:00	11.2	0	0
28/09/2018 01:00	11.1	0	0
28/09/2018 02:00	10.4	0	0
28/09/2018 03:00	10.4	0	0
28/09/2018 04:00	10.6	0	0
28/09/2018 05:00	10.3	0	5
28/09/2018 06:00	12.2	0	53
28/09/2018 07:00	13.8	0	43
28/09/2018 08:00	14.8	0	161
28/09/2018 09:00	17.1	0	505
28/09/2018 10:00	18.6	0	576
28/09/2018 11:00	20	0	591
Mean	**12.7**	**0**	**161**
Maximum	21.9	0	634
Minimum	4.5	0	0

6.9 Abbreviations and explanations

2-MN	2-Methylnaphthalene
Fig.	Figure
Limit of Quantification	Signal/noise is at least 10
Blank	For an analysis, pure water is weighed instead of the sample and prepared according to the analytical method specifications, and the extract is then measured on the analytical apparatus.
FSVO	Federal Food Safety and Veterinary Office
FOAG	Federal Office for Agriculture
CAS no.	Chemical Abstract Service number
DMDA	Dimethyldecanamide; N,N-dimethyldecanamide
DOCUSATE	Dioctyl sulfosuccinate, docusate
Double determination	2 weighing operations and 2 separate procedures are carried out using material from one sample
EC	Formulation type: emulsion concentrate
GC-MS/MS	Gas chromatograph coupled to a mass spectrometer (triple quadrupole)
GC-FID	Gas chromatography with flame-ionisation detection
Control	For an analysis, the produce as purchased from a retail outlet is homogenised, weighed, and prepared according to the analytical method specifications, and the extract is then measured on the analytical apparatus.
LC-MS/MS	Liquid chromatograph coupled to a mass spectrometer (triple quadrupole)
SOL	Solvent
MRM	Multiple reaction monitoring
n.a.	not available
LOQ	limit of quantification (The signal is below the limit of quantification. In exceptional cases, an estimate for the concentration can be made)
< LOQ	below limit of quantification
n.v.	not verifiable, i.e. no signal; concentration is below the verification limit.
PP	Plant protection
PP product	A plant protection product consists of one or more active substances, possibly a synergist or a safener, and co-formulants.
PlantPPO	Plant Protection Products Ordinance
SIM	Single ion monitoring
SP	Formulation type: water-soluble powder
Tab.	Table
% (w/v)	% weight/volume