European Commission

ERM
European Reference Materials

CERTIFICATION REPORT

The certification of the mass fraction of pesticides in soya: ERM®-BC700

EUR 29625 EN

European Commission
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JRC Science Hub
https://ec.europa.eu/jrc

JRC115354

EUR 29625 EN

ISBN 978-92-79-98884-4 (PDF)
ISSN 1831-9424 (online)
doi: 10.2760/064105

Luxembourg: Publications Office of the European Union, 2019

Abstract

This report describes the production of ERM-BC700, which is a soya material certified for the mass fraction of selected pesticides. This material was produced following ISO Guide 34:2009 and is certified in accordance with ISO Guide 35:2006.
Soya beans (Glycine max) originating from Uruguay were sprayed with 11 pesticides in solution. The beans were air dried, cryo-milled and homogenised. Between unit-homogeneity was quantified and stability during dispatch and storage were assessed in accordance with ISO Guide 35:2006.
The material was characterised by an interlaboratory comparison of laboratories of demonstrated competence and adhering to ISO/IEC 17025. Technically invalid results were removed but no outlier was eliminated on statistical grounds only.
Uncertainties of the certified values were calculated in accordance with the Guide to the Expression of Uncertainty in Measurement (GUM) and include uncertainties related to possible inhomogeneity, instability and characterisation.
The material is intended for the quality control / assessment of method performance. As with any reference material, it can be used for establishing control charts or validation studies. The CRM is available in glass vials containing 32 g of cryo-milled soya powder which were sealed under an atmosphere of nitrogen. The minimum amount of sample to be used is 2 g.

CERTIFICATION REPORT

The certification of the mass fraction of pesticides in soya: ERM®-BC700

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Disclaimer

Certain commercial equipment, instruments, and materials are identified in this paper to specify adequately the experimental procedure. In no case does such identification imply recommendation or endorsement by the European Commission, nor does it imply that the material or equipment is necessarily the best available for the purpose.

Summary

This report describes the production of ERM-BC700, which is a soya material certified for the mass fraction of selected pesticides. This material was produced following ISO Guide 34:2009 [1] and is certified in accordance with ISO Guide 35:2006 [2].

Soya beans (*Glycine max*) originating from Uruguay were sprayed with 11 pesticides in solution. The beans were air dried, cryo-milled and homogenised.

Between unit-homogeneity was quantified and stability during dispatch and storage were assessed in accordance with ISO Guide 35:2006 [2].

The material was characterised by an interlaboratory comparison of laboratories of demonstrated competence and adhering to ISO/IEC 17025 [3]. Technically invalid results were removed but no outlier was eliminated on statistical grounds only.

Uncertainties of the certified values were calculated in accordance with the Guide to the Expression of Uncertainty in Measurement (GUM) [4] and include uncertainties related to possible inhomogeneity, instability and characterisation.

The material is intended for the quality control / assessment of method performance. As with any reference material, it can be used for establishing control charts or validation studies. The CRM is available in glass vials containing 32 g of cryo-milled soya powder which were sealed under an atmosphere of nitrogen. The minimum amount of sample to be used is 2 g.

The following values were assigned:

	Mass Fraction[1)]	
	Certified value [4)] [mg/kg]	Uncertainty [5)] [mg/kg]
Azoxystrobin	**0.46**	**0.05**
Carbendazim [2)]	**0.197**	**0.019**
Chlorpyrifos	**0.067**	**0.006**
Cypermethrin	**0.052**	**0.010**
Diazinon	**0.068**	**0.006**
Dieldrin [3)]	**0.075**	**0.007**
(α+β)-Endosulfan[3)]	**0.49**	**0.05**
Imidacloprid [2)]	**0.075**	**0.009**
Iprodione	**0.104**	**0.015**
Methomyl [2)]	**0.046**	**0.006**
Tebuconazole	**0.048**	**0.005**

[1)] reported on dry mass basis and corrected for recovery

[2)] as obtained by liquid chromatography/tandem mass spectrometry

[3)] as obtained by gas chromatography

[4)] Certified values are values that fulfil the highest standards of accuracy and represent the unweighted mean value of the means of accepted sets of data, each set being obtained in a different laboratory and/or with a different method of determination. The certified value and its uncertainty are traceable to the International System of Units (SI).

[5)] The uncertainty is the expanded uncertainty of the certified value with a coverage factor k = 2 corresponding to a level of confidence of about 95 % estimated in accordance with ISO/IEC Guide 98-3, Guide to the Expression of Uncertainty in Measurement (GUM:1995), ISO, 2008.

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Glossary

ANOVA	Analysis of variance
b	Slope in the equation of linear regression $y = a + bx$
CEN	European Committee for Standardization
CRM	Certified reference material
d-SPE	Dispersive solid phase extraction
EC	European Commission
ECD	Electron capture detection
EI	Electron ionisation
EN	European norm (standard)
ERM®	Trademark protected code used by the EC for CRMs
ESI	Electro spray ionisation
EU	European Union
EURL-FV	European Union Reference Laboratory for Pesticides in Fruits and Vegetables
GC	Gas chromatography
GC-ECD	Gas chromatography-electron capture detection
GC-MS	Gas chromatography-mass spectrometry
GUM	Guide to the Expression of Uncertainty in Measurement
HPLC	High performance liquid chromatography
ID	Isotope dilution
IDMS	isotope dilution mass spectrometry
ILC	Interlaboratory comparison
ISO	International Organization for Standardization
IUPAC	International Union of Pure and Applied Chemistry
JRC	Joint Research Centre of the European Commission
k	Coverage factor
KFT	Karl Fischer titration
LC-MS	Liquid chromatography-mass spectrometry
LOD	Limit of detection
LOQ	Limit of quantification
MQC	Method quality control
MRL	Maximum residue level
MS	Mass spectrometry
$MS_{between}$	Mean of squares between-unit from an ANOVA
MS_{within}	Mean of squares within-unit from an ANOVA
n	Number of replicates per unit

N	Number of samples (units) analysed
PCB	Polychlorinated biphenyl
PLE	Pressurised liquid extraction (= accelerated solvent extraction)
PSA	Particle size analysis
PT	Proficiency testing
PTV	Programmable temperature vaporiser
QA	Quality assurance
QC	Quality control
rel	Index denoting relative figures (uncertainties etc.)
RM	Reference material
RM Unit	Reference Materials Unit of Directorate F at JRC
RRF	Relative response factor
RSD	Relative standard deviation
RSE	Relative standard error (=RSD/$\sqrt{n}$)
RT	Room temperature
s	Standard deviation
s_{bb}	Between-unit standard deviation; an additional index "rel" is added when appropriate
$s_{between}$	Standard deviation between groups as obtained from ANOVA; an additional index "rel" is added as appropriate
SANTE	Directorate General for Health and Food Safety
se	Standard error
SI	International System of Units
s_{meas}	Standard deviation of measurement data; an additional index "rel" is added as appropriate
s_{ns}	Standard deviation of results of normal stock samples
SPE	Solid phase extraction
s_{within}	Standard deviation within groups as obtained from ANOVA; an additional index "rel" is added as appropriate
s_{wb}	Within-unit standard deviation
T	Temperature
t	Time
t_i	Time point for each replicate
$t_{\alpha,\,df}$	Critical t-value for a t-test, with a level of confidence of 1-α and df degrees of freedom
t_{sl}	Proposed shelf life
TPhP	Triphenyl phosphate
u	standard uncertainty
U	expanded uncertainty

u^*_{bb}	Standard uncertainty related to a maximum between-unit inhomogeneity that could be hidden by method repeatability; an additional index "rel" is added as appropriate
u_{bb}	Standard uncertainty related to a possible between-unit inhomogeneity; an additional index "rel" is added as appropriate
u_c	combined standard uncertainty; an additional index "rel" is added as appropriate
u_{cal}	Standard uncertainty of calibration
u_{char}	Standard uncertainty of the material characterisation; an additional index "rel" is added as appropriate
u_{CRM}	Combined standard uncertainty of the certified value; an additional index "rel" is added as appropriate
U_{CRM}	Expanded uncertainty of the certified value; an additional index "rel" is added as appropriate
u_Δ	Combined standard uncertainty of measurement result and certified value
u_{lts}	Standard uncertainty of the long-term stability; an additional index "rel" is added as appropriate
u_{meas}	Standard measurement uncertainty
U_{meas}	Expanded measurement uncertainty
u_{rec}	Standard uncertainty related to possible between-unit inhomogeneity modelled as rectangular distribution; an additional index "rel" is added as appropriate
u_{sts}	Standard uncertainty of the short-term stability; an additional index "rel" is added as appropriate
u_t	Standard uncertainty of trueness
$\bar{y}$	Arithmetic mean
α	significance level
Δ_{meas}	Absolute difference between mean measured value and the certified value
$\nu_{s,meas}$	Degrees of freedom for the determination of the standard deviation s_{meas}
$\nu_{MSwithin}$	Degrees of freedom of MS_{within}

1 Introduction

1.1 Background

To ensure safe food for the citizens, European legislation establishes maximum residue levels (MRL) of pesticide residues in food stuffs. Compliance with these levels prior to commercialization of products inside the EU is required [5]. To perform the control and monitoring, laboratories need reliable analytical methodologies which are developed and validated in accordance with the standard requirements listed in ISO/IEC 17025 [3]. Usually multi-residual analytical methods are developed as a way to optimise the process. Yet the list of active compounds and its metabolites which are authorised and banned for use in the EU is very extensive. Therefore the validation of the analytical methods has become a complex and highly time consuming task. The effort required is significantly increased when considering the large amount of different food commodities, giving rise to thousands of matrix/pesticide combinations.

Being aware of this challenge, the European Commission's Directorate for Health and Food Safety (DG SANTE) developed guidance on the validation of analytical procedures for pesticides which is regularly updated [6]. The guidance document groups certain commodities in categories. For vegetables and fruits, cereals and food of animal origin, five commodity clusters are distinguished based on composition:

- high water content
- high acid content and high water content
- high sugar and low water content
- high oil content
- high starch and/or protein content with low water and fat content

Each category can be represented by one typical commodity that would be employed during method validation resulting in a significant simplification for the laboratories. Still, all active compounds and metabolites within the scope of the analytical method need full validation including the assessment of accuracy.

According to ISO/IEC 17025 the use of reference materials and the participation in proficiency testing schemes are essential tools for assuring and controlling the quality of analytical data [3]. Likewise the validation guidance document specifies Certified Reference Materials (CRMs) as the preferable option to provide evidence of analytical method performance [6]. CRMs are used for verification of the accuracy, trueness, for the estimation of uncertainty and to establish the traceability of analytical results. Yet, for pesticide analysis, access to matrix CRMs is currently limited.

To cover this need and to contribute to the harmonisation of reliable analytical results, and thus to the proper implementation of EU legislation, the Joint Research Centre of the European Commission collaborated with the European Union Reference Laboratory for Pesticides in Fruits and Vegetables (EURL-FV) to select representative combinations of plant species and pesticides to produce CRMs.

Soya bean (*Glycine max*) was chosen as a characteristic matrix for the high oil content commodity. Eleven pesticides were then carefully selected based on their use in the commodity as well as different chemical properties for the production of this soya based CRM.

1.2 Choice of the material

Soya beans were selected as matrix to represent groups of food commodities with high oil content [6]. Soya beans (*Glycine max*) originating from Uruguay were provided by Pharmacognosy & Natural products, Faculty of Chemistry, University of the Republic (UdelaR), Montevideo, UY.

Target pesticides were carefully chosen based on a number of criteria with the intention to achieve a broad coverage of different aspects such as the chemical compound families, their intended use, the simplicity/difficulty of analysis as well as to include pesticides which are authorised or banned for use in the EU.

The soya was spiked with the following pesticides: azoxystrobin, carbendazim, chlorpyrifos, cypermethrin, diazinon, diedrin, (α+β)-endosulfan, imidacloprid, iprodione, methomyl, and tebuconazole. Annex A includes information about the different compounds.

The nominal mass fraction levels for the pesticide residues in the soya material were chosen within the range of the MRLs established by the EU legislation. The target concentrations in the matrix were set at three different nominal levels, 0.5 mg/kg for azoxystrobin and (α+β)-endosulfan, 0.2 mg/kg for carbendazim, and 0.05 mg/kg for the remaining compounds.

1.3 Design of the CRM project

The project was designed, managed and developed at the European Commission, Joint Research Centre (EC-JRC), Directorate F – Health, Consumers and Reference Materials, with the participation of the EURL-FV, particularly contributing to the selection of the matrix and pesticides for the CRM.

Analytical methodologies were developed and validated in-house by the JRC to support the different steps of the CRM production. Tandem mass spectrometry coupled to liquid chromatography (LC-MS/MS) and gas chromatography (GC-MS/MS) based on multi-residue methods were applied during the feasibility studies to optimise the material processing conditions as well as for the assessment of homogeneity and stability of the pesticides in the CRM.

Characterisation was based on an interlaboratory comparison involving a number of expert laboratories in the field of pesticide residue analysis. Selected laboratories to take part in the material certification campaign were ISO/IEC 17025 accredited for the particular applications. The participants in the characterisation phase were instructed to apply their own validated analytical methodology for the determination of pesticides in soya. Together with the candidate CRM ERM-BC700, the laboratories received an additional method quality control sample (MQC) of blank soya material. The laboratories were instructed to report the blank values found. Furthermore, the MQC sample could be used for the laboratory's quality control and/or to be used for matrix matched calibration purposes.

2 Participants

2.1 Project management and evaluation

European Commission, Joint Research Centre, Directorate F – Health, Consumers and Reference Materials, Geel, BE
(accredited to ISO Guide 34 for production of certified reference materials, accreditation by BELAC, accreditation number 268-RM)

2.2 Processing

European Commission, Joint Research Centre, Directorate F – Health, Consumers and Reference Materials, Geel, BE
(accredited to ISO Guide 34 for production of certified reference materials, accreditation by BELAC, accreditation number 268-RM)

2.3 Homogeneity and Stabilty study

European Commission, Joint Research Centre, Directorate F – Health, Consumers and Reference Materials, Geel, BE
(accredited to ISO Guide 34 for production of certified reference materials, accreditation by BELAC, accreditation number 268-RM; measurements under the scope of ISO/IEC 17025 accreditation by BELAC, accreditation number 268-TEST)

2.4 Characterisation

AGQ Labs & Technological Services, Sevilla, ES
(measurements under the scope of ISO/IEC 17025 accreditation by ENAC; accreditation number 305/LE1323)

European Commission, Joint Research Centre, Directorate F – Health, Consumers and Reference Materials, Geel, BE
(measurements under the scope of ISO/IEC 17025 accreditation by BELAC, accreditation number 268-TEST)

GALAB Laboratories GmbH, Geesthacht, DE
(measurements under the scope of ISO/IEC 17025 accreditation by Deutsche Akkreditierungsstelle; accreditation number D-PL-14234-01-00)

Institut Dr. Wagner Lebensmittel Analytik GmbH, Lebring, AT
(measurements under the scope of ISO/IEC 17025 accreditation by Akkreditierung Österreich, accreditation number 0239)

Istituto Zooprofilattico Sperimentale dell'Umbria e delle Marche, Perugia, IT
(measurements under the scope of ISO/IEC 17025 accreditation by Accredia, accreditation number 0217)

Laboratório Regional de Veterinária e Segurança Alimentar, Funchal, PT
(measurements under the scope of ISO/IEC 17025 accreditation by Instituto Português de Acreditação; accreditation number L0509-1)

Labor Friedle GmbH, Tegernheim, DE
(measurements under the scope of ISO/IEC 17025 accreditation by Deutsche Akkreditierungsstelle; accreditation number D-P-14646-03-00)

Livsmedelsverket, Uppsala, SE
(measurements under the scope of ISO/IEC 17025 accreditation by SWEDAC; accreditation number 1457)

Norwegian Institute of Bioeconomy Research, Biotechnology and Plant Health, Ås, NO
(measurements under the scope of ISO/IEC 17025 accreditation by Norsk Akkreditering; accreditation number TEST 035)

Pesticides Control Laboratory, Department of Agriculture, Food and the Marine, Backweston, Celbridge, Co Kildare, IE
(measurements under the scope of ISO/IEC 17025 accreditation by Irish National Accreditation Board; accreditation number 121T)

Reactiva Laboratorio S.L., Almería, ES
(measurements under the scope of ISO/IEC 17025 accreditation by ENAC; accreditation number 543/LE1458)

The Food and Environment Research Agency, York, UK
(measurements under the scope of ISO/IEC 17025 accreditation by UKAS; accreditation number 1652)

3 Material processing and process control

3.1 Origin/Purity of the starting material

Soya beans (*Glycine max*) provided by Pharmacognosy & Natural products, Faculty of Chemistry, University of the Republic (UdelaR), Montevideo, UY, was used as base material. Analytical tests were done prior to the processing to verify the content of the selected pesticides. Soya beans from this source were available in two grades, one larger bulk amount (57 kg) with a low level of pesticides and a smaller amount (5.7 kg) with no detectable pesticide contamination.

3.2 Processing

The soya beans were visually inspected and pods and straws were removed by hand. A smaller amount of the low level contamination beans were placed in batches of 1 kg on trays on which 50 mL of a methanol water mixture with the eleven different pesticides was sprayed, in a custom-made spraying chamber (see Figure 1). In total 15 kg of soya beans were spiked in this way. The spray-chamber was attached to a Nederman point-extraction (Helsingborg, SE) with strong evacuation to the outside. During feasibility studies conducted prior to processing (results not shown) it was found that the spraying process results in losses of up to 80 %. Therefore the concentration levels of pesticides were adapted in the spraying solution to compensate for these losses so that the final material reached the intended target levels. Directly after spraying, the trays were transferred to a glove-box where the soya beans were air-dried for 24 hours.

The spiked soya beans were then milled using a Palla VM-KT vibrating cryogenic mill (KHD Humboldt Wedag, Cologne, DE). The mill was cooled down with liquid nitrogen to -196 °C prior to use. Once the temperature increased above -100 °C during milling a new cooling sequence was commenced until all soya beans had been milled. During this step the temperature of the milled product remained below -96 °C.

After being cold-sieved (using liquid nitrogen) over a 500 µm stainless steel mesh (Russel Finex, London, UK), the fraction <500 µm of the soya bean powder was cold-mixed in a three-dimensional mixer for one hour (Dyna-Mix CM200, Basel, CH). For this mixing a gentle mixing program suitable for a fat-rich matrix like soya bean powder was used. Samples were taken to measure the pesticide levels of soya bean powder to confirm spiking levels.

The remaining low level contaminated (but un-spiked) soya beans were milled in the same way as the spiked soya beans and sieved over a 500 µm sieve. The 5.7 kg of blank soya beans underwent the same treatment. Stepwise dilutions were thereafter performed by mixing the spiked soya bean powder with the low level soya and blank soya beans using the Dyna-mix CM200 to approximately reach the nominal concentration levels targeted for the pesticides. The stepwise dilutions were performed in the following way. First 3.9 kg of blank soybean powder was mixed with 3.8 kg of the spiked soybean powder and homogenised in the Dyna-mix CM200 for 30 minutes. In the next step, 1.8 kg of the blank and 6.0 Kg of the spiked soybean powder was added and homogenised again using the Dyna-mix CM200 during 30 minutes. Finally 42.0 kg of the low level contaminated soybean powder was added and mixed for 180 minutes in the Dyna-mix CM200.

Samples were taken for the determination of the pesticide levels as well as for measuring water content and particle size distribution. After this, the resulting 57.5 kg of powder was stored at -20 °C. Prior to aliquoting the total bulk was mixed again and then 32 g powder was filled into 125 mL amber glass bottles with a break ring seal in an atmosphere of dry nitrogen. The bottles were immediately closed by hand inside the glovebox after filling, thereafter labelled and placed into a pre-labelled sachet made of aluminium-coated poly-ethylene foil, and thermo-sealed.

Samples were taken for the determination of the pesticide levels as well as for measuring water content and particle size distribution.

Figure 1: Spraying of pesticide solution on the soya beans

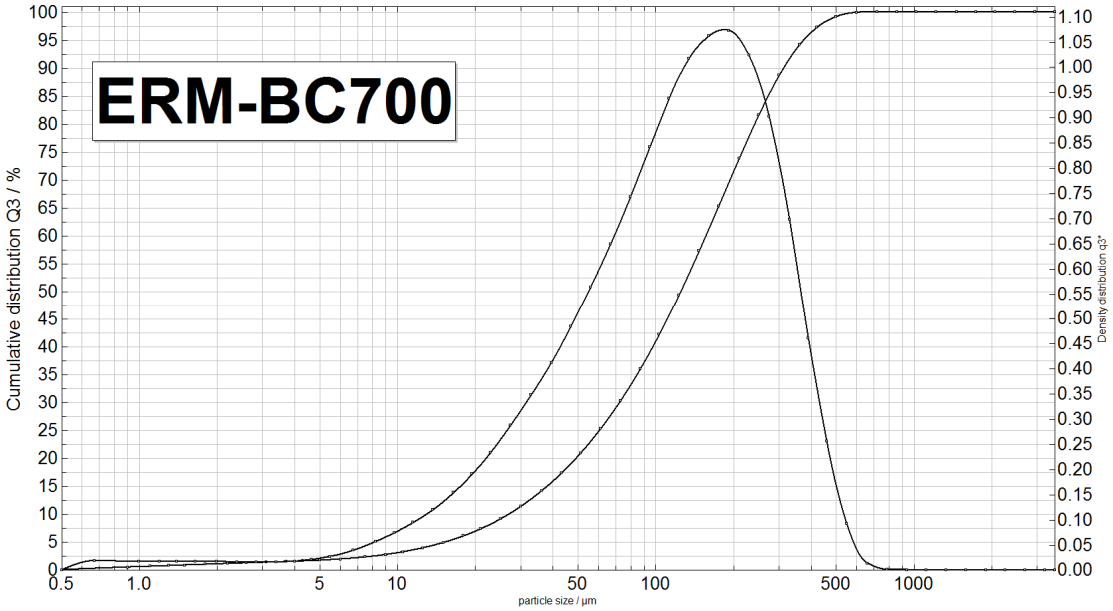
3.3 Process control

The water content in the final product of soya bean powder was measured in duplicate in five samples covering the filling sequence using Volumetric-Karl Fischer titration (Metrohm, Herisau, CH). The average result was 8.06 ± 0.19 % (m/m) (expanded uncertainty).

Micrographs reveal different fractions due to shape and colour-differences and can provide an accurate estimate of the particle size base on direct comparison with a certified length scale.

Results for the particle size distribution showed an average deviation for the X_{10}, X_{50} and X_{90} size classes (on the cumulative particle size distribution curve) well below 20 % which is the acceptance criterion.

Figure 2: Results of the particle size analysis of ERM-BC700 based on laser diffraction



Upper band limit (size classes)	Average particle size / µm, (n = 10)	Standard deviation / µm	Relative standard deviation / %
X_{10}	27.03	3.93	14.54
X_{50}	125.10	4.08	3.26
X_{90}	315.96	22.49	7.12

4 Homogeneity

A key requirement for any reference material aliquoted into units is equivalence between those units. In this respect, it is relevant whether the variation between units is significant compared to the uncertainty of the certified value, but it is not relevant if this variation between units is significant compared to the analytical variation. Consequently, ISO Guide 34 [1] requires RM producers to quantify the between unit variation. This aspect is covered in between-unit homogeneity studies.

The within-unit inhomogeneity does not influence the uncertainty of the certified value when the minimum sample intake is respected, but determines the minimum size of an aliquot that is representative for the whole unit.

4.1 Between-unit homogeneity

The between-unit homogeneity was evaluated to ensure that the certified values of the pesticides in the CRM are valid for all units of the material, within the stated uncertainties.

The number of units selected corresponds to approximately the cube root of the total number of units produced. Thirteen units were selected using a random stratified sampling scheme covering the whole batch for the between-unit homogeneity test. For this, the batch was divided into groups (with a similar number of units) and one unit was selected randomly from each group. Three independent samples were taken from each selected unit, and analysed by GC-MS/MS and LC-MS/MS. The measurements were performed under repeatability conditions, and in a randomised manner to be able to separate a potential analytical drift from a trend in the filling sequence. The results are shown as graphs in Annex B.

Regression analyses were performed to evaluate potential trends in the analytical sequence as well as trends in the filling sequence. One trend in the filling sequence was observed for cypermethrin at a 95 % confidence level. Some significant (95 % confidence level) trends in the analytical measurement sequence were visible, pointing at a changing parameter, e.g. a signal drift in the analytical system. The correction of biases, even if they are statistically not significant, was found to combine the smallest uncertainty with the highest probability to cover the true value [7]. Correction of trends is therefore expected to improve the sensitivity of the subsequent statistical analysis through a reduction in analytical variation without masking potential between-unit heterogeneities. As the analytical sequence and the unit numbers were not correlated, trends significant on at least a 95 % confidence level were corrected as shown below:

$$x_{i\_corr} = x_i - b \cdot i \qquad \textbf{Equation 1}$$

b **=** slope of the linear regression

i **=** position of the result in the analytical sequence

The trend-corrected dataset was assessed for consistency using Grubbs outlier tests at a confidence level of 99 % on the individual results and on the unit means. One outlying unit mean was detected for cypermethrin, however the trend in filling sequence disappeared. Since no technical reason for the outlier could be found, all the data were retained for statistical analysis.

Quantification of between-unit inhomogeneity was undertaken by analysis of variance (ANOVA), which separates the between-unit variation (s_{bb}) from the within-unit variation (s_{wb}). The latter is equivalent to the method repeatability if the individual samples were representative for the whole unit.

Evaluation by ANOVA requires mean values per unit, which follow at least a unimodal distribution and results for each unit that follow unimodal distributions with approximately the same standard deviations. Too few data are available for the unit means to make a clear statement of the distribution. Therefore, it was checked visually whether all individual data

follow a unimodal distribution using histograms and normal probability plots. Minor deviations from unimodality of the individual values do not significantly affect the estimate of between-unit standard deviations. The results of all statistical evaluations are given in Table 1.

Table 1: Results of the statistical evaluation of the homogeneity studies

Pesticide	Trends (before correction)*		Outliers** (after correction)		Distribution (after correction)	
	Analytical sequence	Filling sequence	Individual results	Unit means	Individual results	Unit means
Azoxystrobin[2]	yes	no	none	none	normal	unimodal
Carbendazim[2]	no	no	none	none	normal	normal
Chlorpyrifos[2]	yes	no	none	none	normal	normal
Cypermethrin[1]	yes	yes	none	one	normal	normal
Diazinon[1]	yes	no	none	none	unimodal	unimodal
Dieldrin[1]	no	no	none	none	normal	normal
(α+β)-Endosulfan[1]	no	no	none	none	normal	unimodal
Imidacloprid[2]	no	no	none	none	normal	unimodal
Iprodione[1]	no	no	none	none	normal	bimodal
Methomyl[2]	yes	no	none	none	normal	unimodal
Tebuconazole[2]	no	no	none	none	normal	normal

* 95 % confidence level
** 99 % confidence level
[1] Determined by GC-MS/MS
[2] Determined by LC-MS/MS

It should be noted that $s_{bb,rel}$ and $s_{wb,rel}$ are estimates of the true standard deviations and are therefore subject to random fluctuations. Therefore, the mean square between groups ($MS_{between}$) can be smaller than the mean squares within groups (MS_{within}), resulting in negative arguments under the square root used for the estimation of the between-unit variation, whereas the true variation cannot be lower than zero. In this case, u^*_{bb}, the maximum inhomogeneity that could be hidden by method repeatability, was calculated as described by Linsinger *et al.* [8]. u^*_{bb} is comparable to the LOD of an analytical method, yielding the maximum inhomogeneity that might be undetected by the given study setup.

Method repeatability ($s_{wb,rel}$), between–unit standard deviation ($s_{bb,rel}$) and $u^*_{bb,rel}$ were calculated as:

$$s_{wb,rel} = \frac{\sqrt{MS_{within}}}{\bar{y}} \qquad \textbf{Equation 2}$$

$$s_{bb,rel} = \frac{\sqrt{\frac{MS_{between} - MS_{within}}{n}}}{\bar{y}} \qquad \textbf{Equation 3}$$

$$u^*_{bb,rel} = \frac{\sqrt{\frac{MS_{within}}{n}\sqrt[4]{\frac{2}{v_{MSwithin}}}}}{\bar{y}} \qquad \textbf{Equation 4}$$

MS_{within} mean of squares within-unit from an ANOVA

$MS_{between}$ mean of squares between-unit from an ANOVA

$\bar{y}$ mean of all results of the homogeneity study

n mean number of replicates per unit

$v_{MSwithin}$ degrees of freedom of MS_{within}

However, a different approach was adopted for cypermethrin for which one outlying mean was detected. In this case between-unit inhomogeneity was modelled as a rectangular distribution limited by the largest outlying mean, and the rectangular standard uncertainty of homogeneity was estimated by:

$$u_{rec} = \frac{\left|outlier - \bar{y}\right|}{\sqrt{3} \cdot \bar{y}} \qquad \textbf{Equation 5}$$

$\bar{y}$ mean of all results of the homogeneity study

It should be mentioned that the outlying means are a result of presence of outlying individual values and do not necessarily reflect the real distribution of these elements in the material.

The data does not follow a uni-modal distribution for iprodione. Therefore, u_{rec} was estimated using a rectangular distribution between the highest and lowest unit mean [9]. The uncertainty in those cases is given as:

$$u_{rec} = \frac{\left|highest\ result - lowest\ result\right|}{2 \cdot \sqrt{3} \cdot \bar{y}} \qquad \textbf{Equation 6}$$

The results of the evaluation of the between-unit variation are summarised in Table 2. The resulting values from the above equations were converted into relative uncertainties. In most cases, the uncertainty contribution for homogeneity was determined by the method repeatability.

Table 2: Results of the homogeneity studies

Pesticide	$s_{wb,rel}$ [%]	$s_{bb,rel}$ [%]	$u^*_{bb,rel}$ [%]	$u_{rec,rel}$ [%]	$u_{bb,rel}$ [%]
Azoxystrobin[2]	2.4	n.c.	0.7	n.a.	0.7
Carbendazim[2]	2.6	0.4	0.8	n.a.	0.8
Chlorpyrifos[2]	2.5	n.c.	0.8	n.a.	0.8
Cypermethrin[1]	n.a	n.a	n.a	4.2	4.2
Diazinon[1]	3.0	1.1	0.5	n.a.	1.1
Dieldrin[1]	2.4	1.6	0.4	n.a.	1.6
(α+β)-Endosulfan[1]	4.5	1.4	0.8	n.a.	1.4
Imidacloprid[2]	3.6	0.7	1.1	n.a.	1.1
Iprodione[1]	n.a	n.a	n.a	4.0	4.0
Methomyl[2]	7.7	n.c.	2.3	n.a.	2.3
Tebuconazole[2]	4.7	1.9	1.4	n.a.	1.9

[1] Determined by GC-MS/MS
[2] Determined by LC-MS/MS
n.c.: cannot be calculated as $MS_{between} < MS_{within}$
n.a.: not applicable

The homogeneity study showed no outlying means or trends in the filling sequence with the exception of cypermethrin. Therefore, for all the analytes except cypermethrin and iprodione, the between-unit standard deviation can be used as estimate of u_{bb}. As u^*_{bb} sets the limits of the study to detect inhomogeneity, the larger value of s_{bb} and u^*_{bb} is adopted as uncertainty contribution to account for potential inhomogeneity.

A bimodal distribution was found for iprodione. Taking the two mean values into account, the inhomogeneity as quantified as u_{rec} is still sufficiently small to make the material useful. Therefore, u_{rec} was used as estimate of u_{bb} for this pesticide

One outlying mean was found for cypermethrin. However, taking these extreme values into account, the inhomogeneity as quantified as u_{rec} is still sufficiently small to make the material useful. Therefore, u_{rec} was used as estimate of u_{bb} for this pesticide.

4.2 Within-unit homogeneity and minimum sample intake

The within-unit homogeneity is closely correlated to the minimum sample intake. Due to this correlation, individual aliquots of a material will not contain the same amount of analyte if the sample intake is too small. The minimum sample intake is the minimum amount of sample that is representative for the whole unit and thus should be used in an analysis. Using sample sizes equal or above the minimum sample intake guarantees the certified value within its stated uncertainty.

Homogeneity and stability experiments were performed using a sample intake of 2 g soya powder when applying a GC-MS/MS method whereas 1 g was taken for LC-MS/MS. The sample intake for both measurement techniques gives acceptable repeatability/intermediate precision, demonstrating that the within-unit inhomogeneity no longer contributes to analytical variation.

The minimum sample intake was established from the results of the characterisation study, using the method information supplied by the participants. The smallest sample intake that still yielded results with acceptable accuracy to be included in the respective studies was taken as minimum sample intake. Using the data from Annex E, a minimum sample intake of 2 g soya was established, which is independent from the measurement technique applied.

5 Stability

Time, temperature, light (including ultraviolet radiation) and water content were regarded as the most relevant influences on the stability of the materials. The influence of ultraviolet or visible light was minimised by storing the material in vials in non-transparent sachets. In addition, the certified reference materials are stored in the dark and dispatched in boxes, thus removing any possibility of degradation by light. The water content was adjusted to an optimum during processing and is close to the natural water content in soya. Therefore, only the influences of time and temperature needed to be investigated.

Stability testing is necessary to establish the conditions for storage (long-term stability) as well as the conditions for dispatch of the materials to the customers (short-term stability). During transport, especially in summer time, temperatures up to 60 °C can be reached and stability under these conditions must be demonstrated, if the samples are to be transported without any additional cooling.

The stability studies were carried out using an isochronous design [10]. In this approach, samples were stored for a particular length of time at different temperature conditions. Afterwards, the samples were moved to conditions where further degradation can be assumed to be negligible (reference conditions). At the end of the isochronous storage, the samples were analysed simultaneously under repeatability conditions. Analysis of the material (after various exposure times and temperatures) under repeatability conditions greatly improves the sensitivity of the stability tests.

5.1 Short-term stability study

For the short-term stability study, samples were stored at -20 °C and 4 °C for 0, 1, 2 and 4 weeks (at each temperature). The reference temperature was set to -70 °C. Two units per storage time were selected using a random stratified sampling scheme. From each unit, three subsamples were measured by LC-MS/MS and GC-MS/MS. The measurements were performed under repeatability conditions, and a randomised sequence was used to differentiate any potential analytical drift from a trend over storage time.

The data were evaluated individually for each temperature. The results were screened for outliers using the single and double Grubbs test on a confidence level of 99 %. Some outlying individual results were found (**Table 3: Results of the short-term stability tests**). All the outliers detected at -20 °C corresponded to the same subsample. One additional outlier was detected for diazinon at 4 °C. As no technical reason for the outliers could be found all data were retained for statistical analysis.

In addition, the data were evaluated against storage time, and regression lines of mass fraction versus time were calculated, to test for potential increases/decrease of the mass fraction of the pesticides due to shipping conditions. The slopes of the regression lines were tested for statistical significance. Only one trend was statistically significant at a 95 % confidence level at one of the storage temperatures.

The results of the measurements are shown in Annex C. The results of the statistical evaluation of the short-term stability are summarised in **Table 3**.

Table 3: Results of the short-term stability tests

Pesticide	Number of individual outlying results*		Significance of the trend **	
	-20 ºC	4 ºC	-20 ºC	4 ºC
Azoxystrobin[2]	none	none	no	no
Carbendazim[2]	none	none	yes	no
Chlorpyrifos[2]	none	none	no	no
Cypermethrin[1]	one (retained)	none	no	no
Diazinon[1]	one (retained)	one (retained)	no	no
Dieldrin[1]	one (retained)	none	no	no
(α+β)-Endosulfan[1]	none	none	no	no
Imidacloprid[2]	none	none	no	no
Iprodione[1]	none	none	no	no
Methomyl[2]	none	none	no	no
Tebuconazole[2]	none	none	no	no

[1] Determined by GC-MS/MS
[2] Determined by LC-MS/MS
* 99 % confidence level
** 95 % confidence level

A positive trend was observed for carbendazim at -20 °C. As the analyte cannot be created in the sample, a positive trend could only be due to degradation of the matrix. This, however, should be seen for all measurands, which is not the case. The observed trend was therefore regarded as statistical artefact. The absence of degradation was confirmed by a subsequent stability study conducted at the same temperature for a period of 12 (results not shown) and 24 months which did not reveal any significant trend for carbendazim.

Standard shipment conditions: The material shall be shipped under cooled conditions to ensure temperatures are kept below +20 °C upon arrival.

5.2 Long-term stability study

For the long-term stability study, samples were stored at -20 °C and -40 °C for 0, 8, 16 and 24 months (at each temperature). The reference temperature was set to -70 °C. Two samples per storage time were selected using a random stratified sampling scheme. From each, three subsamples were measured by GC-MS/MS and LC-MS/MS. The measurements were performed under repeatability conditions, in a random sequence to be able to separate any potential analytical drift from a trend over storage time. The results were reported as mass fractions of pesticides in soya.

The long-term stability data were evaluated individually for each temperature. The results were screened for outliers using the single and double Grubbs test at a confidence level of 99 %. Some outlying individual results were found at -20 °C (Table 4). As no technical reason for the outliers could be found all data were retained for statistical analysis.

In addition, the data were plotted against storage time and linear regression lines of mass fraction versus time were calculated. The slopes of the regression lines were tested for statistical significance (loss/increase due to storage). No significant trend was detected at a 95 % confidence level with the exception of azoxystrobin (-20 °C).

The results of the long-term stability measurements are shown in Annex D. The results of the statistical evaluation of the long-term stability study are summarised in Table 4.

Table 4: Results of the long-term stability tests

Pesticide	Number of individual outlying results*		Significance of the trend**	
	-40 °C	-20 ºC	-40 °C	-20 ºC
Azoxystrobin[2]	none	none	no	yes
Carbendazim[2]	none	one (retained)	no	no
Chlorpyrifos[2]	none	none	no	no
Cypermethrin[1]	none	none	no	no
Diazinon[1]	none	none	no	no
Dieldrin[1]	none	none	no	no
(α+β)-Endosulfan[1]	none	none	no	no
Imidacloprid[2]	none	none	no	no
Iprodione[1]	none	one (retained)	no	no
Methomyl[2]	none	none	no	no
Tebuconazole[2]	none	none	no	no

[1] Determined by GC-MS/MS
[2] Determined by LC-MS/MS
* 99 % confidence level
** 95 % confidence level

Two outliers were statistically significant on a 99 % confidence level. As in both cases no technical explanation could be found, the outliers were retained for further statistical evaluation.

A positive trend was observed for azoxystrobin at -20 °C. As the analyte cannot be created in the sample, a positive trend could only be due to degradation of the matrix. This, however, should be seen for all measurands, which is not the case. This trend was not observed for azoxystrobin measured by GC-MS/MS (results not shown). The observed trend was therefore regarded as statistical artefact.

5.3 Estimation of uncertainties

Due to the intrinsic variation of measurement results, no study can entirely rule out degradation of materials, even in the absence of statistically significant trends. It is therefore necessary to quantify the potential degradation that could be hidden by the method repeatability, i.e. to estimate the uncertainty of stability. This means that, even under ideal conditions, the outcome of a stability study can only be that there is no detectable degradation within an uncertainty to be estimated.

The uncertainties of stability during dispatch and storage were estimated, as described in [11] for each pesticide. In this approach, the uncertainty of the linear regression line with a slope of zero was calculated. The uncertainty contributions u_{sts} and u_{lts} were calculated as the product of the chosen transport time/shelf life and the uncertainty of the regression lines as:

$$u_{sts,rel} = \frac{s_{rel}}{\sqrt{\sum \left(t_i - \bar{t}\right)^2}} \cdot t_{tt} \qquad \textbf{Equation 7}$$

$$u_{lts,rel} = \frac{s_{rel}}{\sqrt{\sum \left(t_i - \bar{t}\right)^2}} \cdot t_{sl} \qquad \textbf{Equation 8}$$

s_{rel} relative standard deviation of all results of the stability study

t_i time elapsed at time point *i*

$\bar{t}$ mean of all t_i

t_{tt} chosen transport time (1 week at 4 ºC)

t_{sl} chosen shelf life (24 months at -20 ºC)

The following uncertainties were estimated:

- $u_{sts,rel}$, the uncertainty of degradation during dispatch. This was estimated from the 4 °C studies. The uncertainty describes the possible change during a dispatch at 4 °C lasting for one week.
- $u_{lts,rel}$, the stability during storage. This uncertainty contribution was estimated from the -20 °C studies. The uncertainty contribution describes the possible degradation during 24 months storage at -20 °C.

The results of these evaluations are summarised in Table 5.

Table 5: Uncertainties of stability during dispatch and storage. $u_{sts,rel}$ was calculated for a temperature of 4 °C and 1 week; $u_{lts,rel}$ was calculated for a storage temperature of -20 °C and 24 months

Pesticide	$u_{sts,rel}$ [%]	$u_{lts,rel}$ [%]
Azoxystrobin[2]	0.3	1.4
Carbendazim[2]	0.3	1.9
Chlorpyrifos[2]	0.5	1.8
Cypermethrin[1]	0.3	1.2
Diazinon[1]	0.3	1.1
Dieldrin[1]	0.2	0.9
(α+β)-Endosulfan[1]	0.3	1.9
Imidacloprid[2]	0.4	2.1
Iprodione[1]	0.7	1.8
Methomyl[2]	0.7	3.4
Tebuconazole[2]	0.5	1.9

[1] Determined by GC-MS/MS
[2] Determined by LC-MS/MS

The material showed no significant degradation for transport below 4 °C. The material should be shipped with cooling.

After the certification study, the material will be included in the JRC's regular stability monitoring programme, to control its further stability.

6 Characterisation

The material characterisation is the process of determining the property values of a reference material.

This was based on an interlaboratory comparison of expert laboratories, i.e. the pesticide mass fractions of the material were determined in different laboratories that applied different measurement procedures to demonstrate the absence of a measurement bias. Due to the nature of the analytes however, all participants used liquid and/or gas chromatographic methods, in most cases followed by mass spectrometric detection, for the measurements. One exception is the use of gas chromatography - electron capture detection. This approach aims at randomisation of laboratory bias, which reduces the combined uncertainty.

6.1 Selection of participants

Twelve laboratories were selected based on criteria that comprised both technical competence and quality management aspects. Each participant was required to operate a quality system and to deliver documented evidence of its laboratory proficiency in the field of pesticide measurements in relevant matrices. To this end the laboratories had to submit results for intercomparison exercises or method validation reports. Having a formal accreditation was not mandatory, but meeting the requirements of ISO/IEC 17025 was obligatory. Where measurements are covered by the scope of accreditation, the accreditation number is stated in the list of participants (Section 2.4).

6.2 Study setup

Each laboratory received two units of the CRM and was requested to provide six independent results, three per unit. This means that each laboratory had to carry out six extractions and clean-ups. The units for material characterisation were selected using a random stratified sampling scheme and covered the whole batch. The sample preparations and measurements had to be spread over at least two days to ensure intermediate precision conditions. The water content had to be determined in each unit and results are reported on dry mass basis and corrected for recovery.

Each participant received additionally a sample labelled as method quality control (MQC). The material was a milled soya free of pesticides to be measured as blank. It could optionally be employed as well for any recovery tests by the participant laboratories. The results obtained for the blank sample were used to support the evaluation of the characterisation results.

Laboratories were also requested to give estimations of the expanded uncertainties of the mean value of the six results. No approach for the estimation of the measurement uncertainty was prescribed, i.e. top-down and bottom-up were regarded as equally valid procedures.

6.3 Methods used

A variety of extraction and clean-up methods with different quantification approaches were used to characterise the material. Some pesticides were analysed by a combination of GC and LC methods while others were exclusively analysed using one of the methods. The combination of results from methods based on different principles mitigates undetected method bias.

All methods used during the characterisation study are summarised in Annex E. The laboratory code (e.g. L01) is a random number and does not correspond to the order of laboratories in Section 2.4. The lab-method code is amended with an abbreviation of the measurement method used, (e.g. LC-MS/MS). Different codes were assigned to laboratories providing data for particular pesticides using more than one method of determination.

6.3.1 Dry mass determination

For all measurements carried out during certification (characterisation studies) the following protocol for dry mass determination was applied:

1 g sample (in duplicate), oven dried at 103 °C for 1 hour

The water content determined by the laboratories was in the range of 5 - 8 %. However, results within each laboratory were consistent and in agreement with the results from the processing control (Section 3.3).

6.4 Evaluation of results

The characterisation study resulted in 10-15 datasets per pesticide. All individual results of the participants, grouped per pesticide are displayed in tabular and graphical form in Annex F.

6.4.1 Technical evaluation

The obtained data were first checked for compliance with the requested analysis protocol and for their validity based on technical reasons. The following criteria were considered during the evaluation:

- appropriate validation of the measurement procedure
- compliance with the analysis protocol: sample preparations and measurements performed on two days, and the analytical sequence and water content determination.
- absence of values given as below limit of detection or below limit of quantification
- method performance, MQC sample measured to confirm absence (<LOQ) of target pesticides
- according to SANTE/111813/2017 [6] the method repeatability should be not higher than 20%.

Based on the above criteria all datasets were technically valid with the exception of the following cases. It was reported from L09 that the recoveries of the MQC sample were outside the laboratory's normal acceptable range for cypermethrin and methomyl. The results for these two pesticides were therefore not used for the evaluation. Furthermore it was noted that the method repeatability exceeded 20% for diazinon and dieldrin for L09, endosulfan for L11 and chlorpyrifos and cypermethin for L12. The results from these laboratories for these pesticides are therefore not included in the evaluation.

6.4.2 Statistical evaluation

The datasets accepted based on technical reasons were tested for normality of dataset means using kurtosis/skewness tests and normal probability plots and were tested for outlying means using the Grubbs test and using the Cochran test for outlying standard deviations, (both at a 99 % confidence level). Standard deviations within (s_{within}) and between ($s_{between}$) laboratories were calculated using one-way ANOVA. The results of these evaluations are shown in Table 6.

Table 6: Statistical evaluation of the technically accepted datasets for ERM-BC700. *p*: number of technically valid datasets

Pesticide	*p*	Outliers		Normally distributed	Statistical parameters			
		Means	Variances		Mean [mg/kg]	*s* [mg/kg]	$s_{between}$ [mg/kg]	s_{within} [mg/kg]
Azoxystrobin	14	0	6	yes	0.4559	0.0897	0.0885	0.0358
Carbendazim	12	0	3	yes	0.1970	0.0281	0.0275	0.0140
Chlorpyrifos	13	0	1	yes	0.0666	0.0093	0.0092	0.0034
Cypermethrin	10	0	3	yes	0.0523	0.0141	0.0140	0.0028
Diazinon	14	0	2	yes	0.0676	0.0092	0.0089	0.0056
Dieldrin	11	0	1	yes	0.0749	0.0103	0.0100	0.0061
(α+β)-Endosulfan	11	0	0	yes	0.4871	0.0731	0.0723	0.0261
Imidacloprid	11	0	0	yes	0.0747	0.0123	0.0122	0.0045
Iprodione	13	0	3	yes	0.1042	0.0210	0.0206	0.0101
Methomyl	10	0	2	yes	0.0463	0.0071	0.0070	0.0032
Tebuconazole	13	0	2	yes	0.0475	0.0078	0.0077	0.0038

The laboratory means follow normal distributions and none of the datasets contain outlying means. The datasets are therefore consistent and the mean of laboratory means is a good estimate of the true value. Standard deviations between laboratories are considerably larger than the standard deviation within laboratories, showing that confidence intervals of replicate measurements are unsuitable as estimate of measurement uncertainty.

The statistical evaluation flags the datasets laboratories L04, L05, L07, L08, L09 and L12 as outlying variances for azoxystrobin. Similarly, other outlying variances are identified for a number of pesticides as follows; Laboratories L05, L09 and L11 show outlying variances for carbendazim and iprodione. The same situation is valid for the variance reported by L07 for chlorpyrifos, L05, L06 and L07 for cypermethrin, L05 and L12 for diazinon, and L11 for dieldrin. Laboratories L05 and L07 are outlying variances for methomyl and L05 and L12 are outliers for tebuconazole. This merely reflects the fact that different methods have different intrinsic variability. As all measurement methods were found technically sound, all results were retained.

The uncertainty related to the characterisation is estimated as the standard error of the mean of laboratory means (Table 7).

Table 7: Uncertainty of characterisation for ERM-BC700

Pesticide	*p*	Mean [mg/kg]	*s* [mg/kg]	u_{char} [mg/kg]
Azoxystrobin	14	0.4559	0.0897	0.0240
Carbendazim	12	0.1970	0.0281	0.0081
Chlorpyrifos	13	0.0666	0.0093	0.0026
Cypermethrin	10	0.0523	0.0141	0.0045
Diazinon	14	0.0676	0.0092	0.0025
Dieldrin	11	0.0749	0.0103	0.0031
(α+β)-Endosulfan	11	0.4871	0.0731	0.0221
Imidacloprid	11	0.0747	0.0123	0.0037
Iprodione	13	0.1042	0.0210	0.0058
Methomyl	10	0.0463	0.0071	0.0023
Tebuconazole	13	0.0475	0.0078	0.0022

7 Value Assignment

Certified values were assigned for 11 pesticides.

Certified values are values that fulfil the highest standards of accuracy. Procedures at the JRC, Directorate F require generally pooling of not less than 6 datasets to assign certified values. Full uncertainty budgets in accordance with the 'Guide to the Expression of Uncertainty in Measurement' [4] were established.

Additional material information refers to values that were obtained in the course of the study. For example, results reported from only one or two laboratories or in cases where individual measurement uncertainty is high, would fall under this category.

7.1 Certified values and their uncertainties

The unweighted mean of the means of the accepted datasets as shown in Table 6 were assigned as certified values for each of the 11 pesticides.

The assigned uncertainty consists of uncertainties relating to characterisation, u_{char} (Section 6), potential between-unit inhomogeneity, u_{bb} (Section 4.1), and potential degradation during transport, u_{sts}, and long-term storage, u_{lts} (Section 5). In some cases the uncertainty related to inhomogeneity/degradation during transport/long-term storage was found to be negligible. These different contributions were combined to estimate the relative expanded uncertainty of the certified value ($U_{CRM,\,rel}$) with a coverage factor k given as:

$$U_{CRM,rel} = k \cdot \sqrt{u_{bb,rel}^2 + u_{sts,rel}^2 + u_{lts,rel}^2 + u_{char,rel}^2} \qquad \textbf{Equation 9}$$

- u_{char} was estimated as described in Section 6.4.2
- u_{bb} was estimated as described in Section 4.1.
- u_{sts} and u_{lts} were estimated as described in section 5.3

A coverage factor k of 2 was applied, to obtain the expanded uncertainties. The certified values and their uncertainties are summarised in Table 8.

Table 8: Certified values and their uncertainties for ERM-BC700

Pesticide	Certified value[1)] [mg/kg]	u_{char} [mg/kg]	u_{bb} [mg/kg]	u_{sts} [mg/kg]	$u_{lts,}$ [mg/kg]	U_{CRM} [3)] [mg/kg]
Azoxystrobin	**0.46**	0.0240	0.0033	0.0011	0.0064	**0.05**
Carbendazim	**0.197**	0.0081	0.0015	0.0006	0.0038	**0.019**
Chlorpyrifos	**0.067**	0.0026	0.0005	0.0003	0.0012	**0.006**
Cypermethrin	**0.052**	0.0045	0.0022	0.0002	0.0006	**0.010**
Diazinon	**0.068**	0.0025	0.0007	0.0002	0.0008	**0.006**
Dieldrin	**0.075**	0.0031	0.0012	0.0002	0.0007	**0.007**
(α+β)-Endosulfan	**0.49**	0.0221	0.0066	0.0016	0.0095	**0.05**
Imidacloprid	**0.075**	0.0037	0.0008	0.0003	0.0016	**0.009**
Iprodione	**0.104**	0.0058	0.0041	0.0007	0.0018	**0.015**
Methomyl	**0.046**	0.0023	0.0011	0.0003	0.0016	**0.006**
Tebuconazole	**0.048**	0.0022	0.0009	0.0002	0.0009	**0.005**

[1)]: reported on dry mass basis (Section 6.3.1) and corrected for recovery
[2)] The certified value is above/below this level (with a 95 % confidence level).
[3)] Expanded (k = 2) and rounded uncertainty.

7.2 Additional material information

α-Endosulfan and β-Endosulfan were reported individually and as a sum of (α+β)-Endosulfan by the participants. As there are no specific information regarding the distribution of α- and β-endosulfan from the homogeneity study and the stability studies, the results should be regarded as informative only on the general composition of the material and cannot be, in any case, used as certified or indicative value.

Pesticide	Mass Fraction Range[1)] [mg/kg]
α-Endosulfan	0.08-0.15
β-Endosulfan	0.28-0.47

[1)] reported on dry mass basis and corrected for recovery

Endosulfan sulphate was not detected in the material.

8 Metrological traceability and commutability

8.1 Metrological traceability

Identity

Pesticides are chemically clearly defined substances. Identity was confirmed by mass spectrometry following either gas or liquid chromatography. The participants used different methods for the sample preparation as well as for the final determination, demonstrating absence of measurement bias. The identity of the measurand is therefore structurally defined when the determination is based on the application of the different techniques. The identity of the measurand is method defined in those cases where exclusively LC/MS is applied and in those cases where exclusively gas chromatography is used.

Quantity value

Only validated methods were used for the determination of the assigned values. Investigation of the method and measurement details of the individual results shows that all the relevant input parameters of each technically accepted dataset have been properly calibrated. In some cases for the GC methods other detectors such as ECD were additionally employed. Calibrants of known purity, specified traceability of their assigned values and of different independent commercial origins were used. All values in the technically accepted datasets are therefore traceable to the same reference, namely the SI. The traceability to the SI is also confirmed by the agreement of results within their respective uncertainties through the use of GC and/or LC methods as indicated in the certificate.

8.2 Commutability

Many measurement procedures include one or more steps which select specific (or specific groups of) measurands from the sample for the subsequent whole measurement process. Often the complete identity of these 'intermediate analytes' is not fully known or taken into account. Therefore, it is difficult to mimic all analytically relevant properties of real samples within a CRM. The degree of equivalence in the analytical behaviour of real samples and a CRM with respect to various measurement procedures (methods) is summarised in a concept called 'commutability of a reference material'. There are various definitions that define this concept. For instance, the CLSI Guideline C53-A [12] recommends the use of the following definition for the term *commutability*:

> "The equivalence of the mathematical relationships among the results of different measurement procedures for an RM and for representative samples of the type intended to be measured."

The commutability of a CRM defines its fitness for use and is therefore a crucial characteristic when applying different measurement methods. When the commutability of a CRM is not established, the results from routinely used methods cannot be legitimately compared with the certified value to determine whether a bias does not exist in calibration, nor can the CRM be used as a calibrant.

ERM-BC700 was produced from a naturally grown soya bean material which was spiked with a mixture of pesticides and further manipulated by cryo-milling and mixing. The analytical behaviour of this matrix is expected to be highly similar to routine samples. It should be borne in mind that the methods used in the characterisation are methods routinely applied for measuring pesticides in soya. The agreement of results from different methods demonstrates that the processing did not affect any property relevant for these methods and that ERM-BC700 behaves like a real sample.

9 Instructions for use

9.1 Safety information

The ERM-BC700 is intended for laboratory use only. The usual laboratory safety measures apply.

9.2 Storage conditions

The materials should be stored at -20 ± 5 °C in the dark.

Please note that the European Commission cannot be held responsible for changes that happen during storage of the material at the customer's premises, especially for opened vials.

9.3 Preparation and use of the material

The material consists of an amber glass vial containing 32 g soya bean powder.

Leave the vial to thaw at room temperature. The units shall be shaken by turning upside down by hand for at least 1 min before opening to ensure material re-homogenisation.

9.4 Minimum sample intake

The minimum sample intake representative for all parameters is 2 g.

9.5 Dry mass correction

Dry mass determination shall be carried out on a separate portion of at least 1 g, by drying in an oven at 103 ^{o}C ± 2 ^{o}C until constant mass (separate weighing should not differ by more than 5 mg) is attained. Weighing of the samples for dry mass determination and weighing for the analysis shall be done at the same time to avoid differences due to possible take up of moisture by the material.

9.6 Use of the certified value

The main purpose of these materials is to assess method performance, i.e. for checking accuracy of analytical results/calibration. As any reference material, it can be used for establishing control charts or validation studies.

Use as a calibrant

It is not recommended to use this matrix material as calibrant. If used nevertheless, the uncertainty of the certified value shall be taken into account in the estimation of the measurement uncertainty.

Comparing an analytical result with the certified value

A result is unbiased if the combined standard uncertainty of measurement and certified value covers the difference between the certified value and the measurement result (see also ERM Application Note 1, [13].

When assessing the method performance, the measured values of the CRMs are compared with the certified values. The procedure is summarised here:

- Calculate the absolute difference between mean measured value and the certified value (Δ_{meas}).
- Combine the measurement uncertainty (u_{meas}) with the uncertainty of the certified value (u_{CRM}): $u_{\Delta} = \sqrt{u_{meas}^2 + u_{CRM}^2}$
- Calculate the expanded uncertainty (U_{Δ}) from the combined uncertainty ($u_{\Delta,}$) using an appropriate coverage factor, corresponding to a level of confidence of approximately 95 %

- If $\Delta_{meas} \leq U_{\Delta}$ then no significant difference exists between the measurement result and the certified value, at a confidence level of approximately 95 %.

Use in quality control charts

The materials can be used for quality control charts. Using CRMs for quality control charts has the added value that a trueness assessment is built into the chart.

10 Acknowledgments

The authors would like to acknowledge the support received from the JRC, Directorate F.6 processing team and from Maria Contreras concerning the set-up of the required isochronous studies. Furthermore, the authors would like to thank Horacio Heinzen, Pharmacognosy & Natural products, Faculty of Chemistry, University of the Republic (UdelaR), Montevideo, UY for supplying the soya beans.

Finally, the authors would like to thank Stefanie Trapmann and Reinhard Zeleny (JRC, Directorate F) for reviewing the certification report, as well as the experts of the Reference Material Review Panel, T. Pihlström (National Food Agency, Sweden) and J. de Boer (Vrije Universiteit, Amsterdam, The Netherlands) for their constructive comments.

11 References

1 ISO Guide 34:2009, General requirements for the competence of reference materials producers, International Organization for Standardization, Geneva, Switzerland

2 ISO Guide 35:2006, Reference materials – General and statistical principles for certification, International Organization for Standardization, Geneva, Switzerland

3 ISO/IEC 17025:2017(en) General requirements for the competence of testing and calibration laboratories, International Organization for Standardization, Geneva, Switzerland

4 ISO/IEC Guide 98-3:2008, Guide to the Expression of Uncertainty in Measurement, (GUM 1995), International Organization for Standardization, Geneva, Switzerland

5 Regulation (EC) No 396/2005 of the European Parliament and of the Council of 23 February 2005 on maximum residue levels of pesticides in or on food and feed of plant and animal origin and amendments, Brussels, Belgium.

6 European Commission, Directorate General for Health And Food Safety, Guidance document on analytical quality control and method validation procedures for pesticide residues and analysis in food and feed (SANTE/111813/2017), Brussels, Belgium

7 G.E. O'Donnell, D.B: Hibbert, Treatment of bias in estimating measurement uncertainty, Analyst 130 (2005) 721-729

8 T.P.J. Linsinger, J. Pauwels, A.M.H. van der Veen, H. Schimmel, A. Lamberty, Homogeneity and stability of reference materials, Accred. Qual. Assur. 6 (2001) 20-25

9 EURACHEM/CITAC Guide (2000) Quantifying Uncertainty in Analytical Measurement. www.measurementuncertainty.org/mu/guide/index.html (last accessed on 04/10/2018)

10 A. Lamberty, H. Schimmel, J. Pauwels, The study of the stability of reference materials by isochronous measurements, Fres. J. Anal. Chem. 360 (1998) 359-361

11 T.P.J. Linsinger, J. Pauwels, A. Lamberty, H. Schimmel, A.M.H. van der Veen, L. Siekmann, Estimating the uncertainty of stability for matrix CRMs, Fres. J. Anal. Chem. 370 (2001) 183-188

12 H. Vesper, H. Emons, M. Gnezda, C. P. Jain, W. G. Miller, R. Rej, G. Schumann, J. Tate, L. Thienpont, J. E. Vaks, Characterization and qualification of commutable reference materials for laboratory medicine; approved guideline, CLSI document C53-A, Clinical and Laboratory Standards Institute, Wayne, PA, USA, 2010

13 T.P.J. Linsinger, ERM Application Note 1: Comparison of a measurement result with the certified value, https://ec.europa.eu/jrc/en/reference-materials/application-notes (last accessed on 04/10/2018)

12 Annexes

Annex A: List of target pesticides for certification in ERM-BC700 with some characteristics

Annex B: Results of the homogeneity measurements.

Annex C: Results of the short-term stability measurements

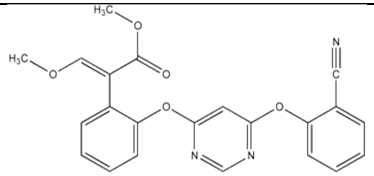
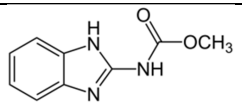
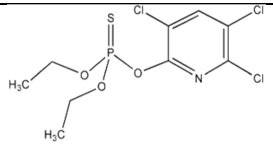
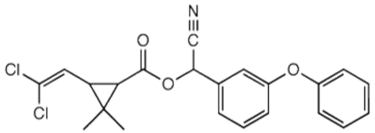
Annex D: Results of the long-term stability measurements

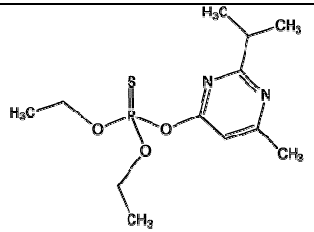
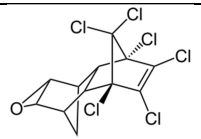
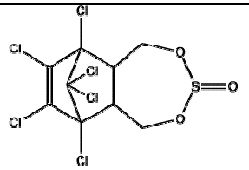
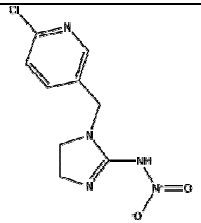
Annex E: Summary of analytical methods used in the characterisation of ERM-BC700

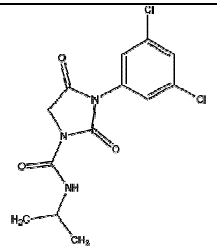
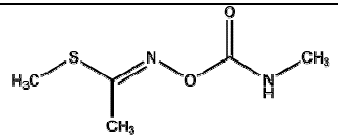
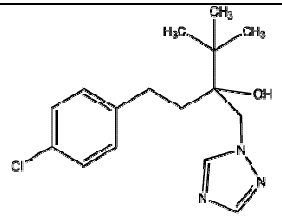
Annex F: Results of the characterisation measurements of ERM-BC700

Annexes

Annex A. List of pesticides for certification in ERM-BC700 with some characteristics

Pesticide	Chemical class	Application	CAS number	Chemical structure	Molecular weight	MRL[1] [mg/kg]	Legislation Reg (EU)[2]
Azoxystrobin	Strobin	Fungicide	131860-33-8		403.39	0.5	2011/559
Carbendazim	Benzimidazole	Fungicide	10605-21-7		191.19	0.2	2011/559
Chlorpyrifos	Organophosphorus	Insecticide, nematicide	2921-88-2		350.57	0.05	2018/686
Cypermethrin (mixture of constituent isomers (sum of))	Pyrethroid	Insecticide	52315-07-8 (undefined stereochemistry)		416.30	0.05	2017/626

Pesticide	Chemical class	Application	CAS number	Chemical structure	Molecular weight	MRL[1] [mg/kg]	Legislation Reg (EU)[2]
Diazinon	Organophosphorus	Insecticide	333-41-5		304.35	0.02	2013/834
Dieldrin	Organochloride	Insecticide	60-57-1		380.91	0.02	2008/839
Endosulfan[3] (sum of alpha and beta isomers)	Organochloride	Insecticide	115-29-7		406.93	0.5	2011/310
Imidachloprid	Neonicotinoid	Insecticide	138261-41-3		255.66	0.05	2014/491

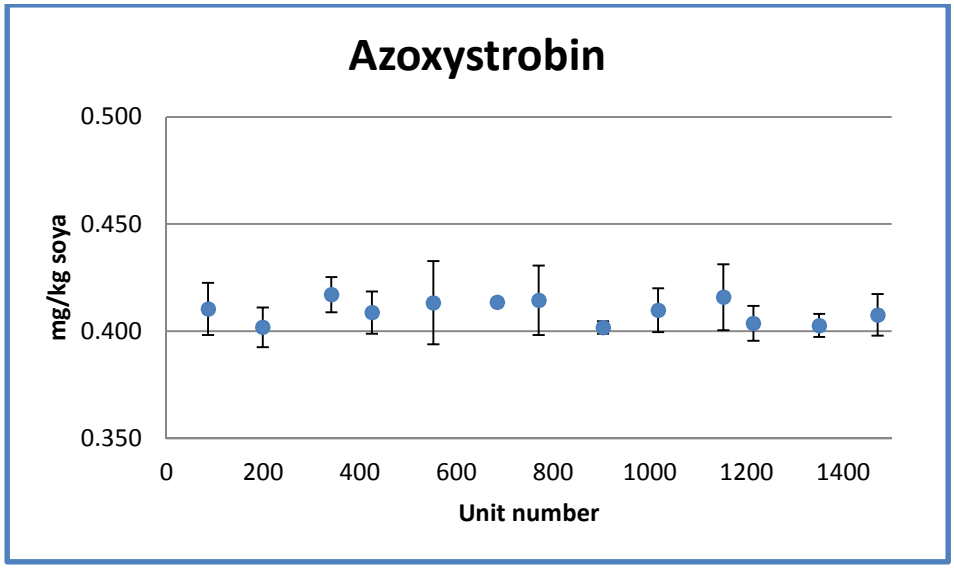
Pesticide	Chemical class	Application	CAS number	Chemical structure	Molecular weight	MRL[1] [mg/kg]	Legislation Reg (EU)[2]
Iprodione	Dicarboximide	Fungicide, nematicide	36734-19-7		330.17	0.01	2015/400
Methomyl	N-Methyl Carbamate	Insecticide	16752-77-5		162.20	0.2	2016/1822
Tebuconazole	Triazole	Fungicide	107534-96-3		307.82	0.15	2017/626

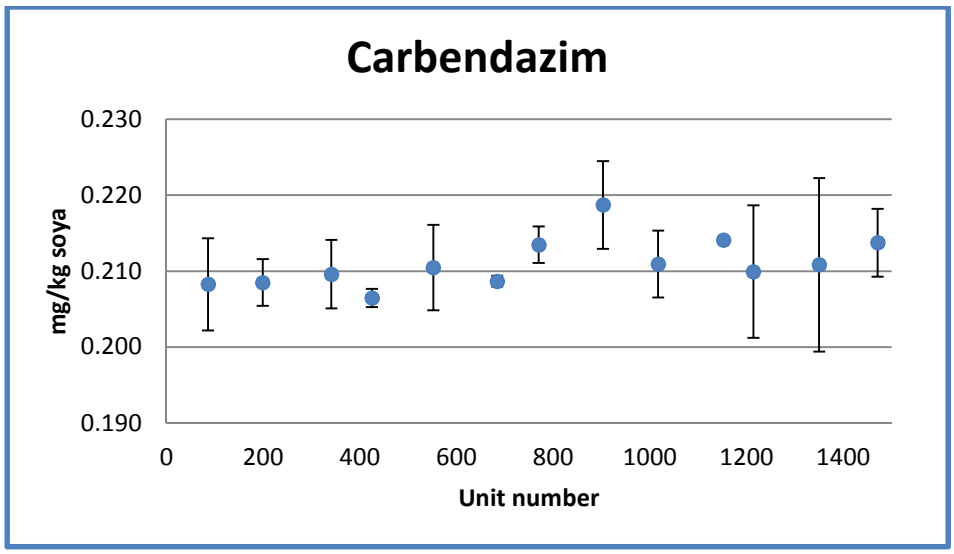
[1]MRL in soya **(http://ec.europa.eu/food/plant/pesticides/eu-pesticides-database/public/?event=homepage&language=EN, last accessed August 2018)**

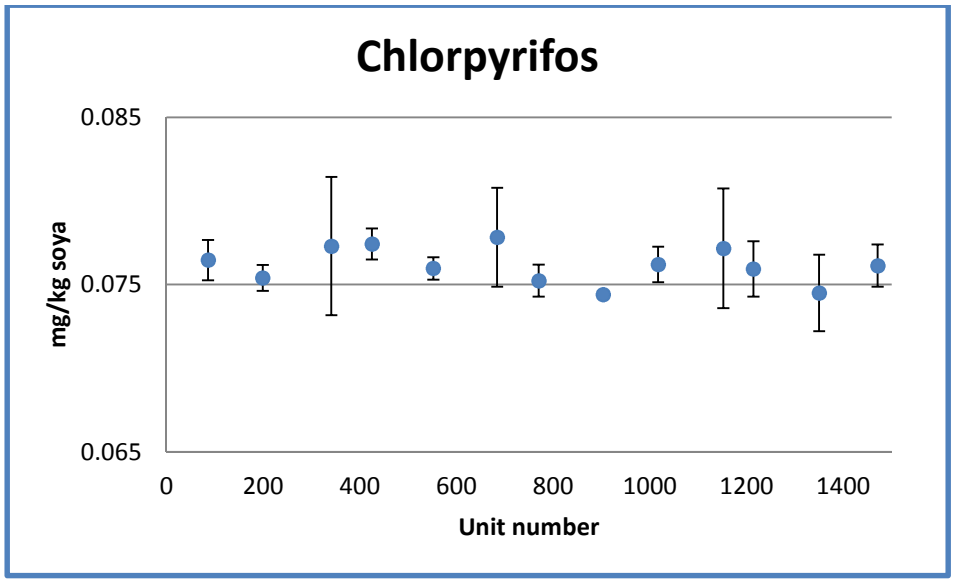
[2]EU Regulation amending relevant annexes of Regulation (EU) 396/2005

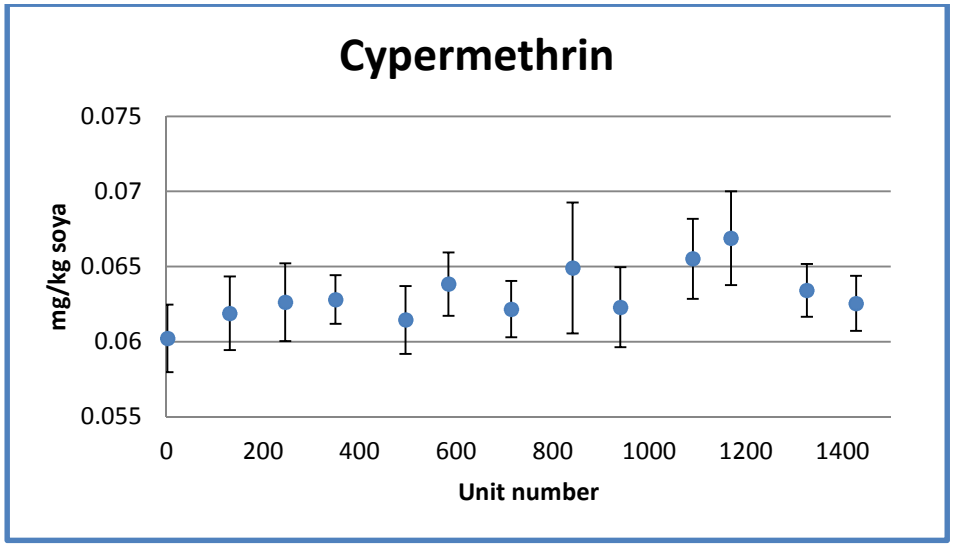
[3]Endosulfan is defined as the sum of α and β-endosulfan and endosulfan-sulphate expressed as endosulfan. In this material, only α- and β-endosulfan are detected.

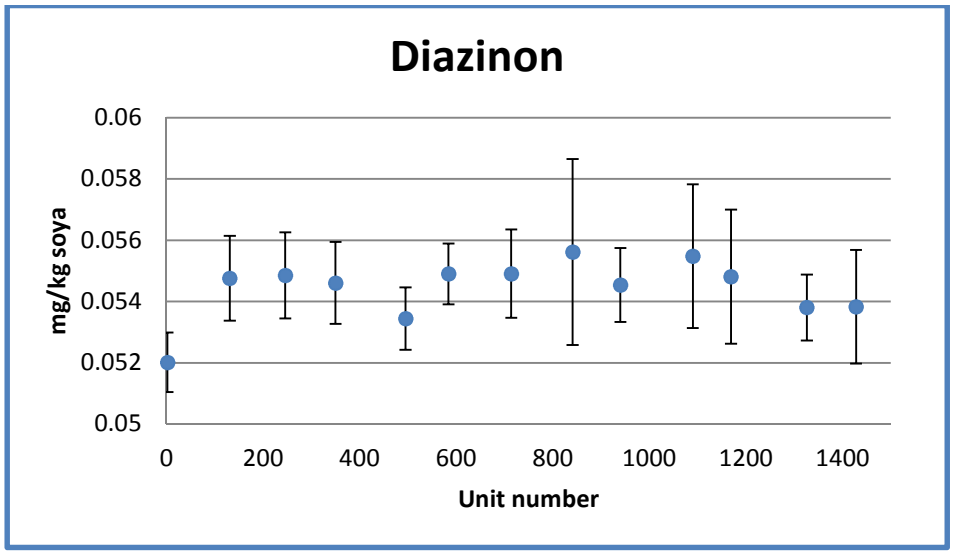
Annex B. Results of the homogeneity measurements. Values are illustrated as mean mass fractions of pesticide obtained from the analysis of 3 subsamples per unit of ERM-BC700.

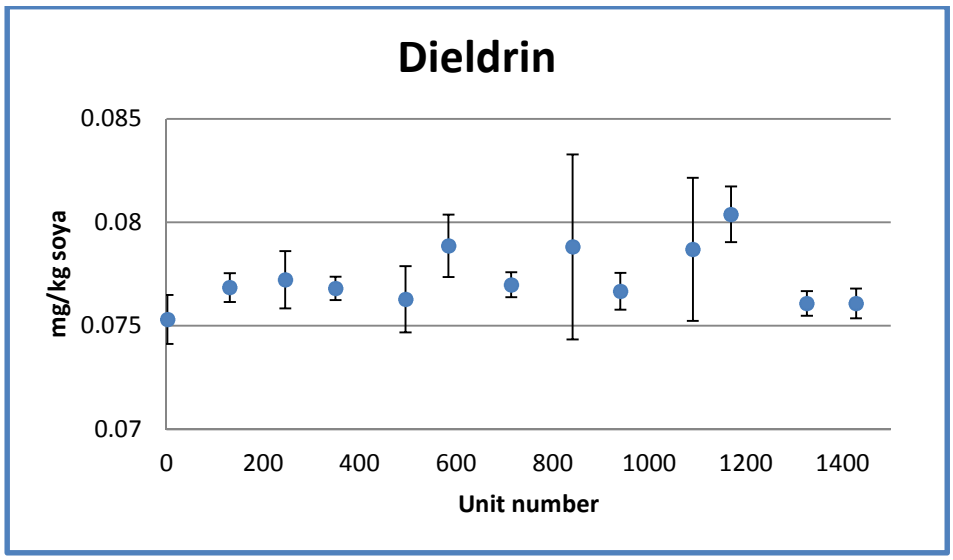


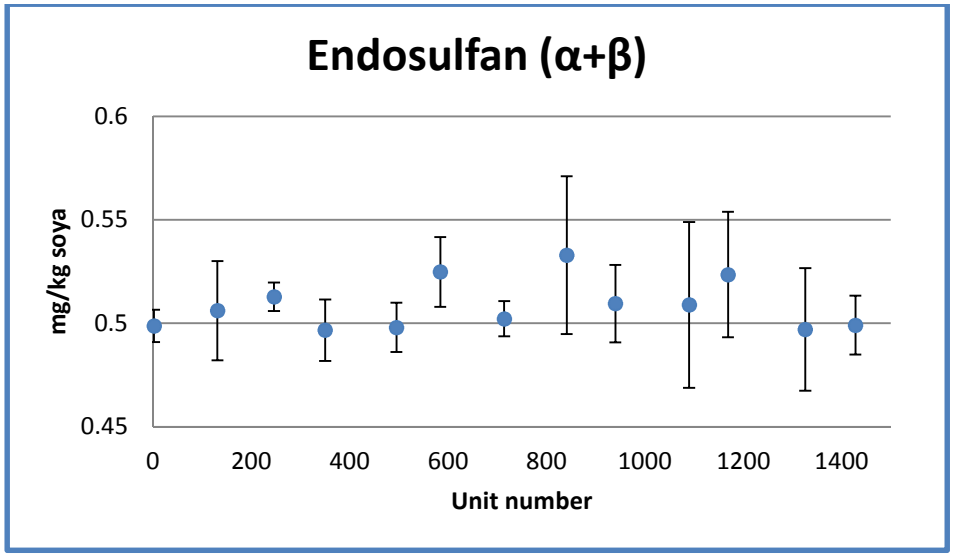


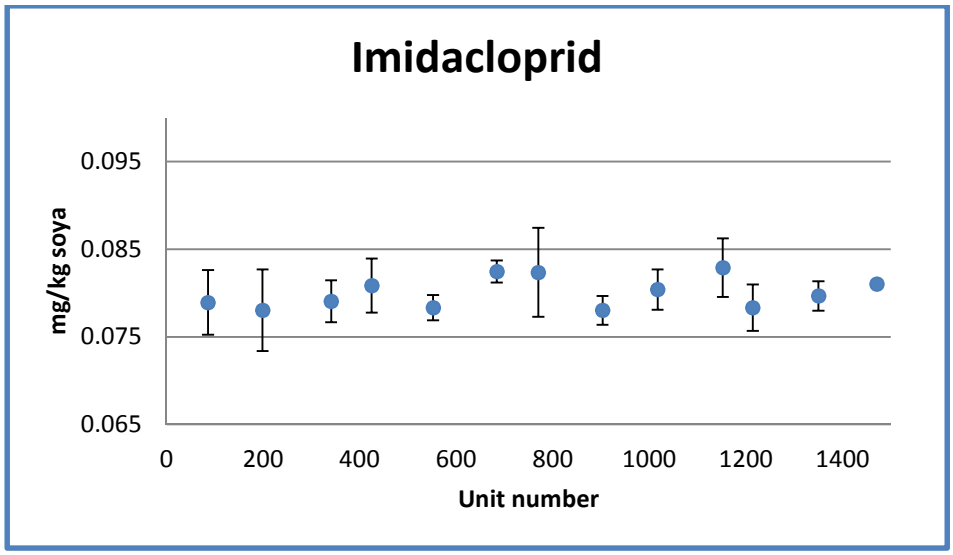


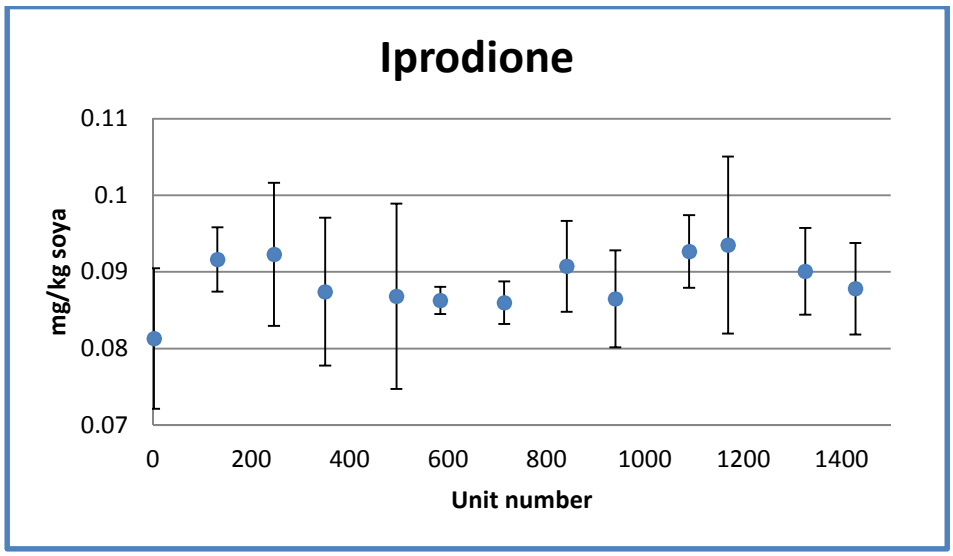

Cypermethrin
0.075
0.07
0.065
0.06
0.055
mg/kg soya
0
200
400
600
800
1000
1200
1400
Unit number

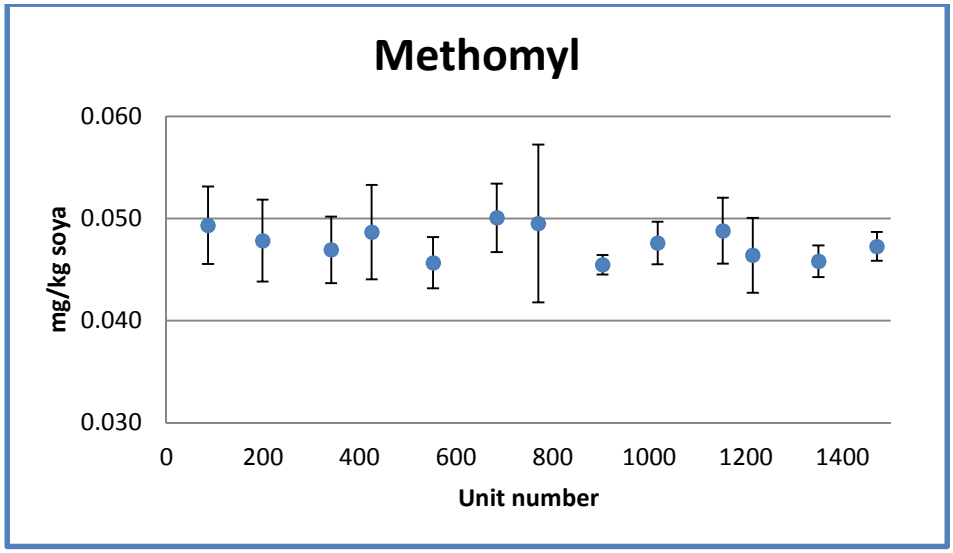

Diazinon
0.06
0.058
0.056
0.054
0.052
0.05
mg/kg soya
0
200
400
600
800
1000
1200
1400
Unit number

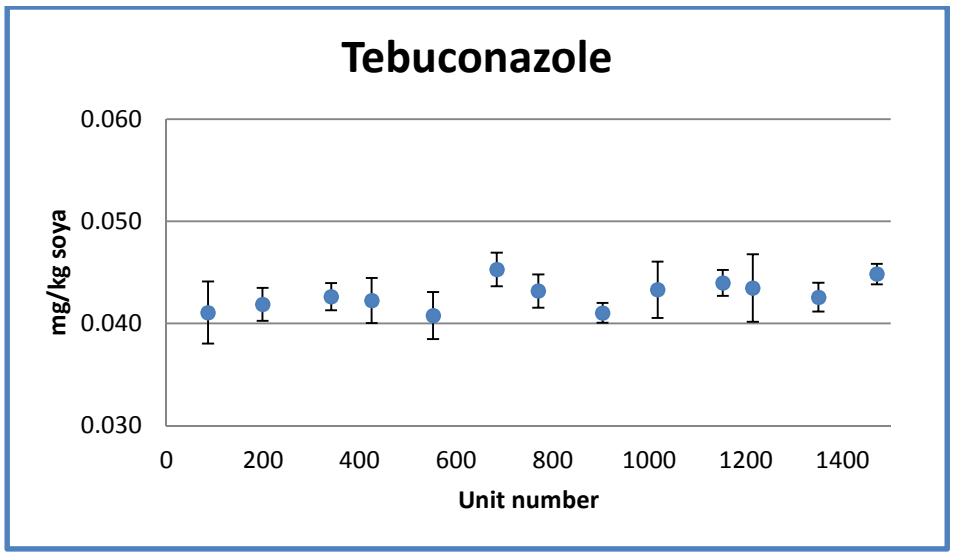

Dieldrin
0.085
0.08
0.075
0.07
mg/kg soya
0
200
400
600
800
1000
1200
1400
Unit number


Endosulfan (α+β)
mg/kg soya
0.6
0.55
0.5
0.45
0
200
400
600
800
1000
1200
1400
Unit number

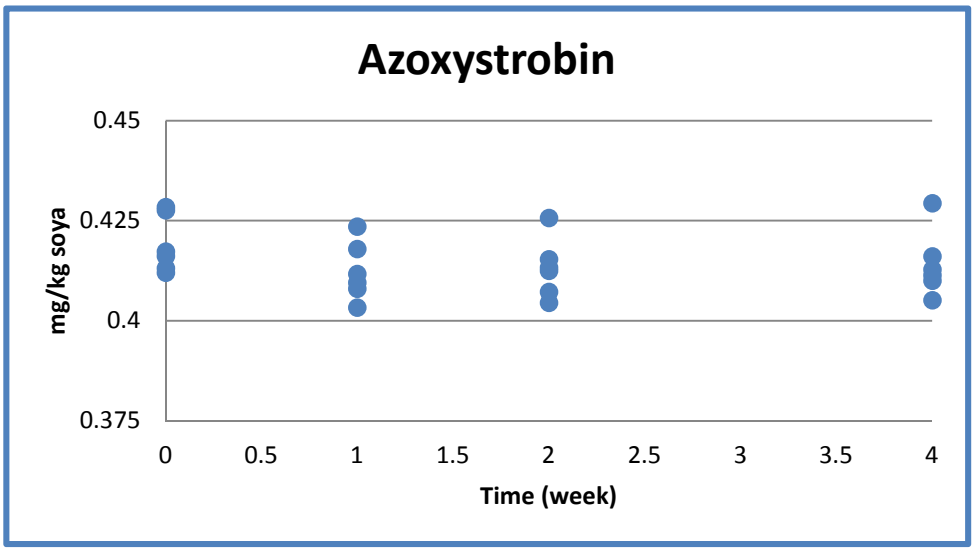

Imidacloprid
mg/kg soya
0.095
0.085
0.075
0.065
0
200
400
600
800
1000
1200
1400
Unit number

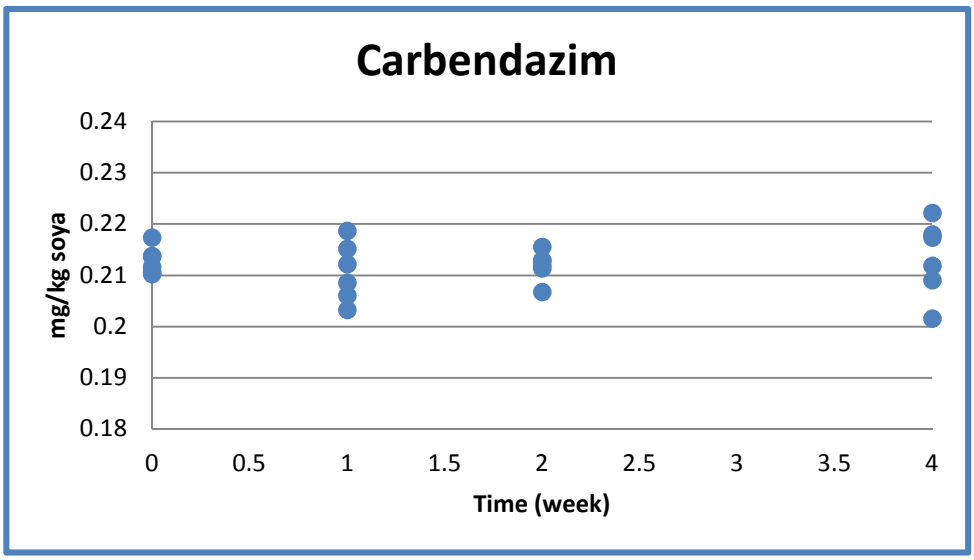

Iprodione
mg/kg soya
0.11
0.1
0.09
0.08
0.07
0
200
400
600
800
1000
1200
1400
Unit number

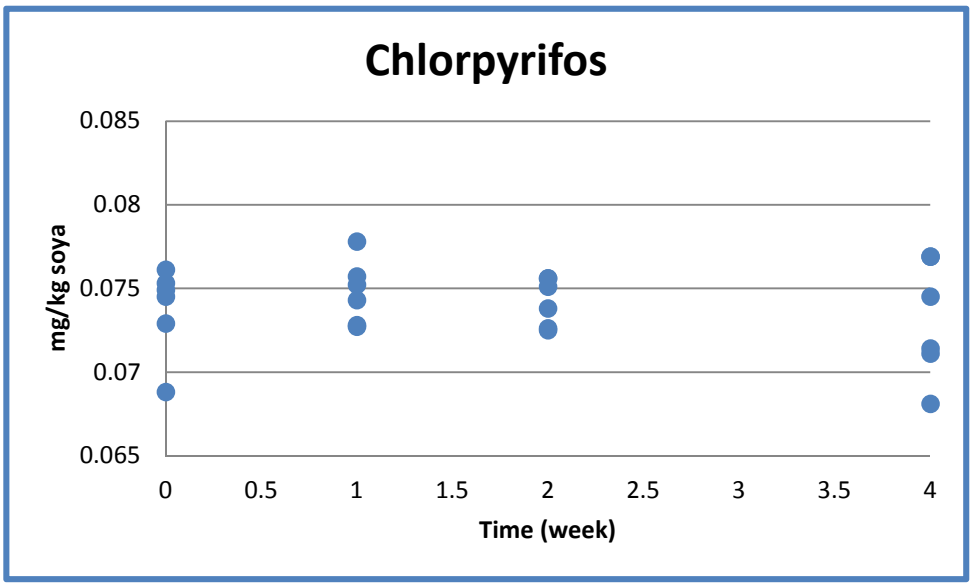

Methomyl
0.060
0.050
0.040
0.030
mg/kg soya
0
200
400
600
800
1000
1200
1400
Unit number

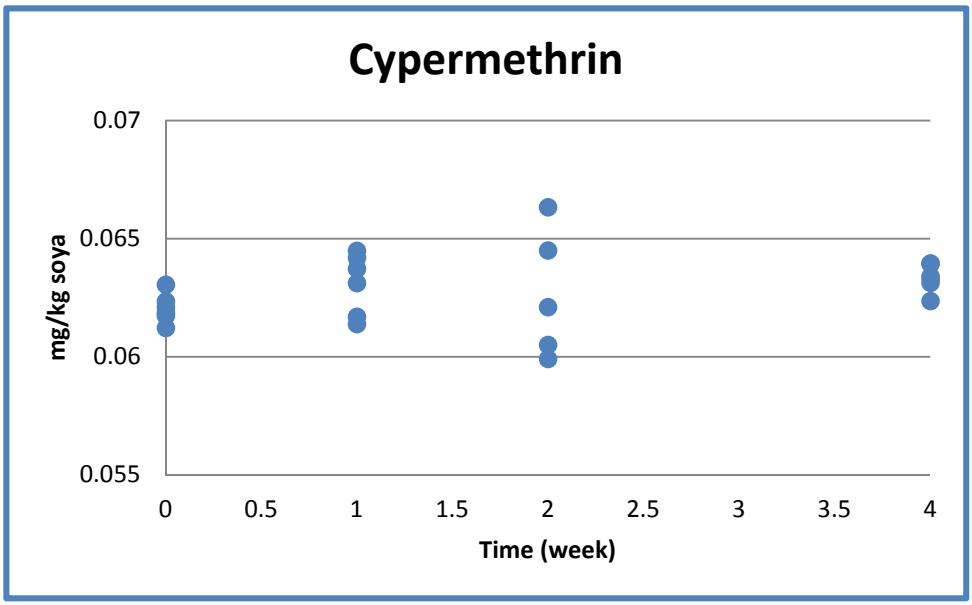

Tebuconazole
0.060
0.050
0.040
0.030
mg/kg soya
0
200
400
600
800
1000
1200
1400
Unit number

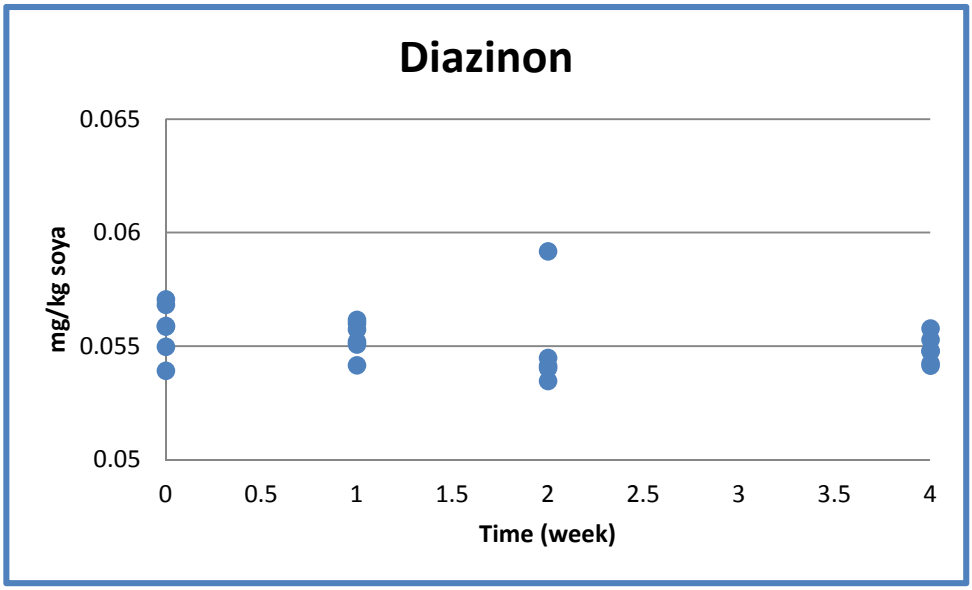
Annex C: **Results of the short-term stability measurements at +4 ^{0}C. Graphs provide individual results (6 replicates per time point) to better illustrate the presence, if any, of outliers.**

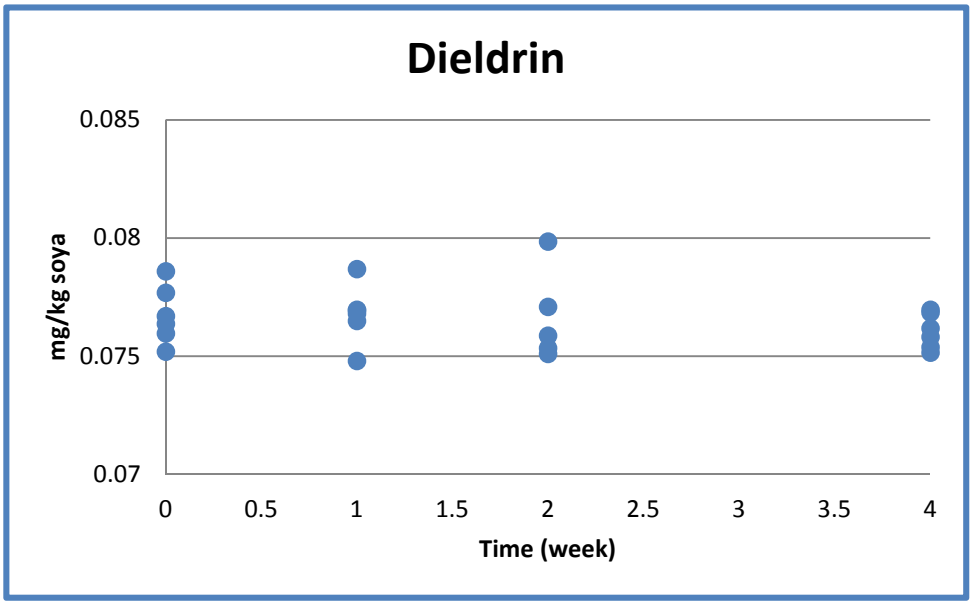


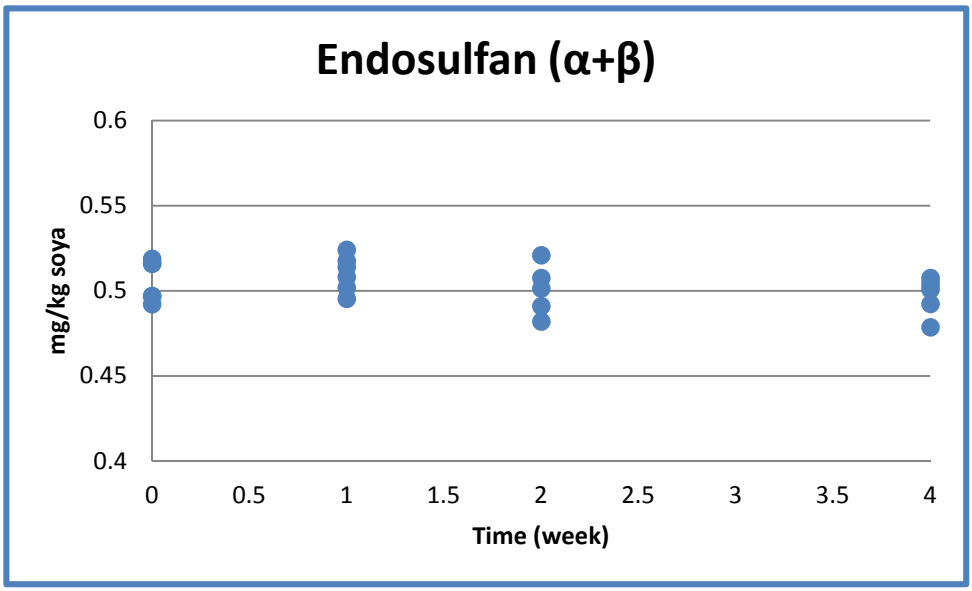


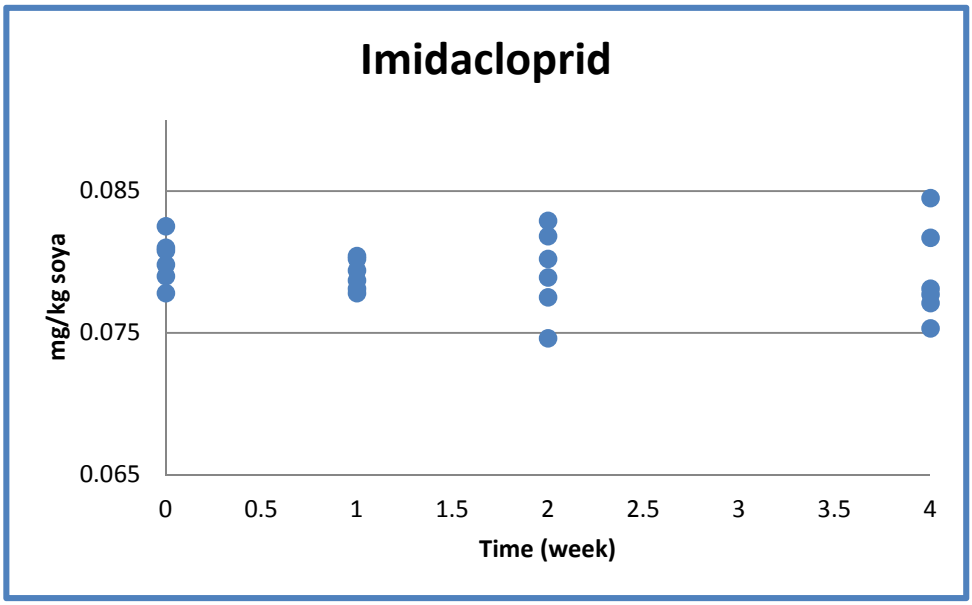


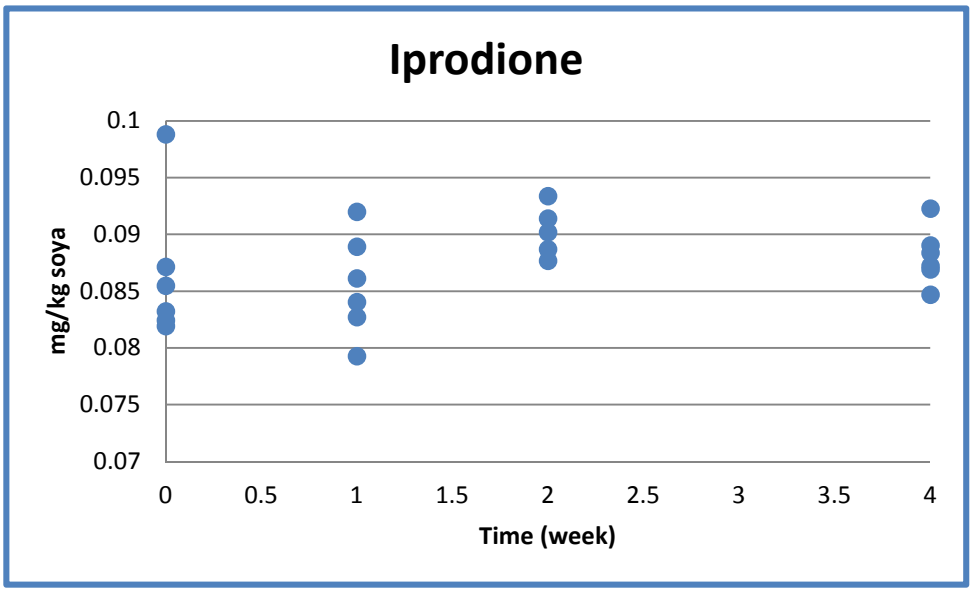

Cypermethrin
0.07
0.065
0.06
0.055
mg/kg soya
0
0.5
1
1.5
2
2.5
3
3.5
4
Time (week)

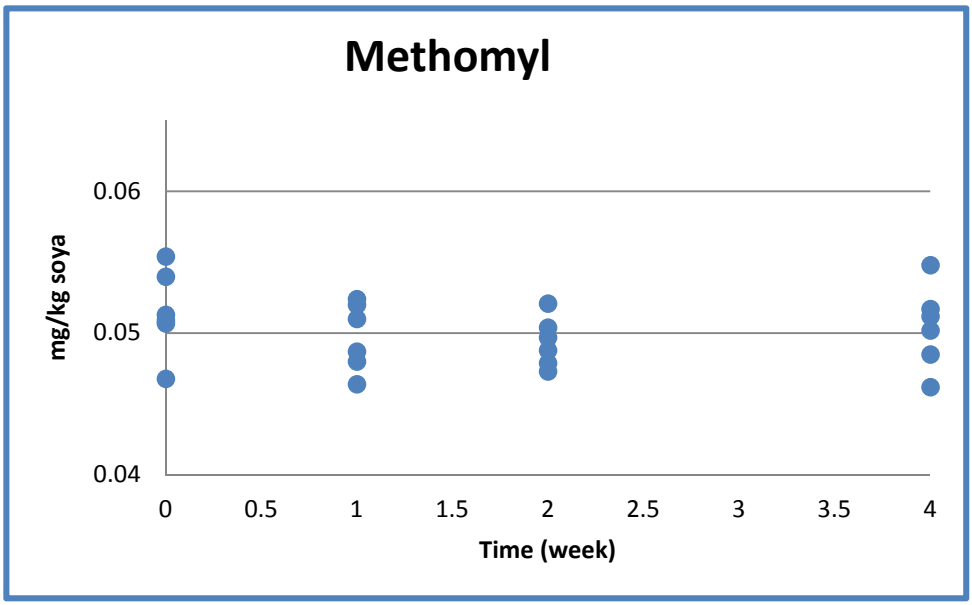

Diazinon
0.065
0.06
0.055
0.05
mg/kg soya
0
0.5
1
1.5
2
2.5
3
3.5
4
Time (week)

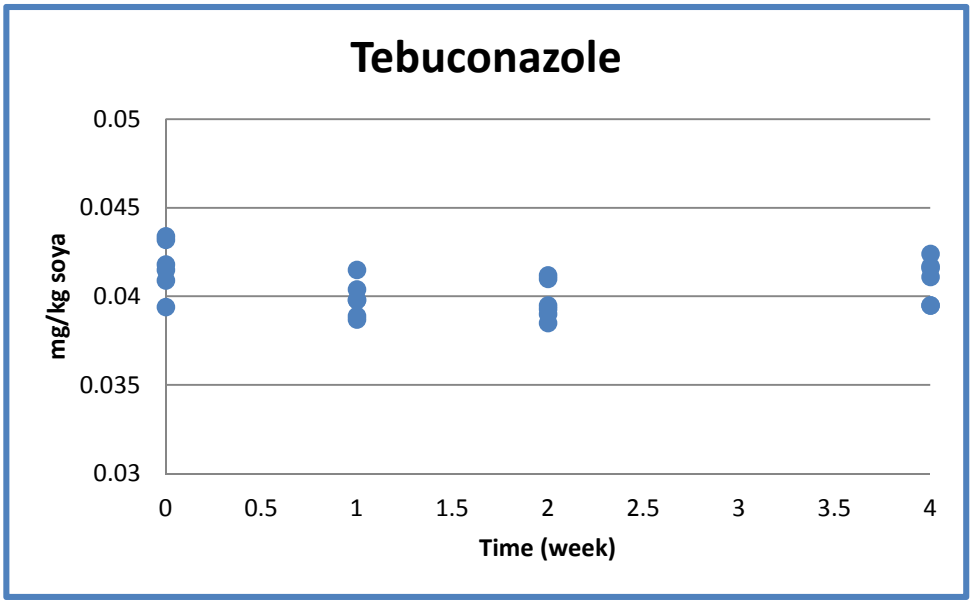

Dieldrin
0.085
0.08
0.075
0.07
mg/kg soya
0
0.5
1
1.5
2
2.5
3
3.5
4
Time (week)


Endosulfan (α+β)
mg/kg soya
0.6
0.55
0.5
0.45
0.4
0
0.5
1
1.5
2
2.5
3
3.5
4
Time (week)

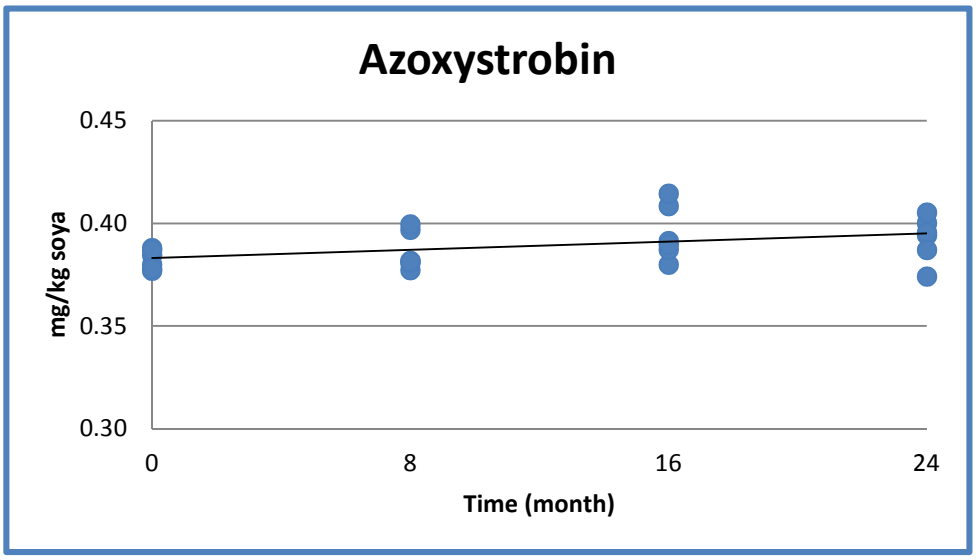

Imidacloprid
mg/kg soya
0.085
0.075
0.065
0
0.5
1
1.5
2
2.5
3
3.5
4
Time (week)

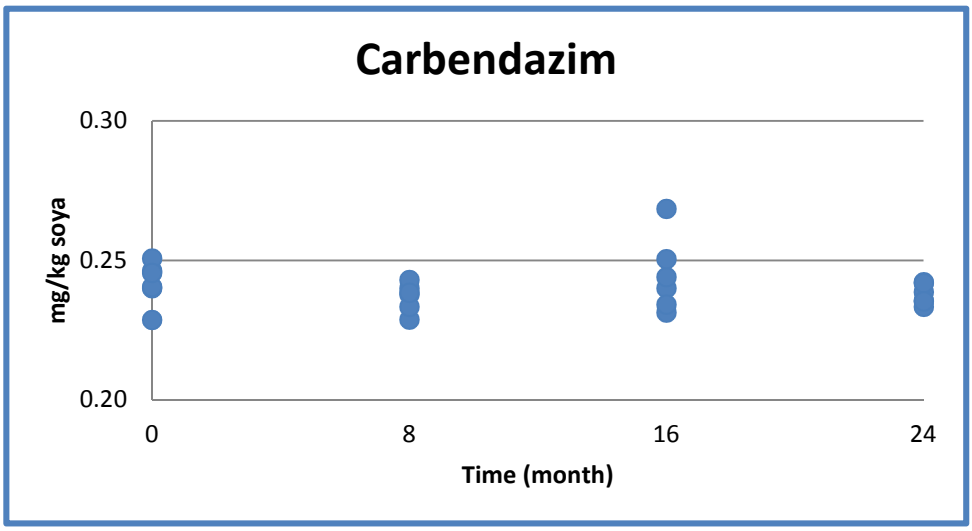

Iprodione
mg/kg soya
0.1
0.095
0.09
0.085
0.08
0.075
0.07
0
0.5
1
1.5
2
2.5
3
3.5
4
Time (week)

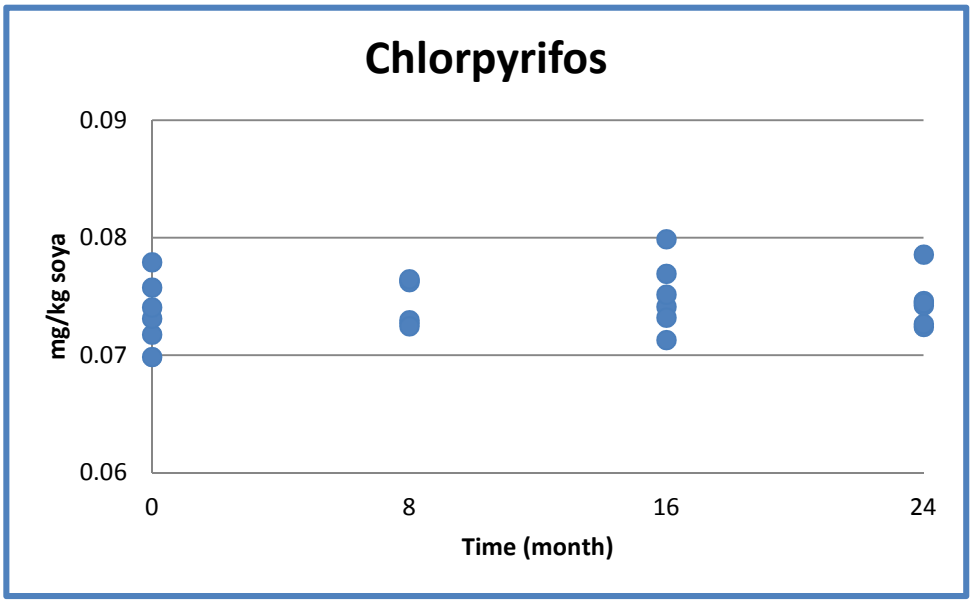

Methomyl
0.06
0.05
0.04
mg/kg soya
0
0.5
1
1.5
2
2.5
3
3.5
4
Time (week)

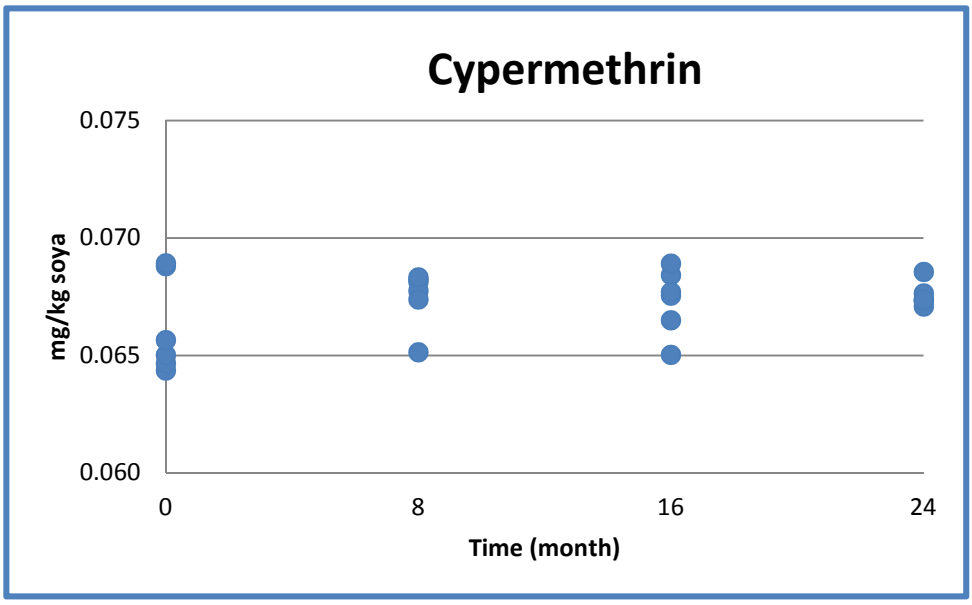

Tebuconazole
0.05
0.045
0.04
0.035
0.03
mg/kg soya
0
0.5
1
1.5
2
2.5
3
3.5
4
Time (week)

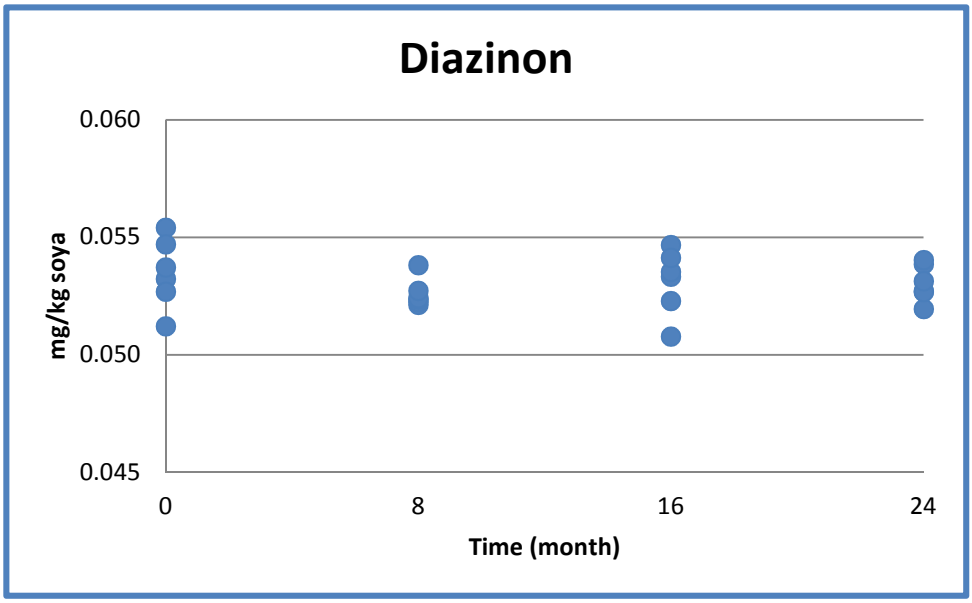
Annex D. **Results of the long-term stability results at -20 °C for 24 months. Graphs provide individual results (6 replicates per time point) to better illustrate the presence, if any, of outliers. A linear regression trendline is plotted whenever a trend is statistically significant.**

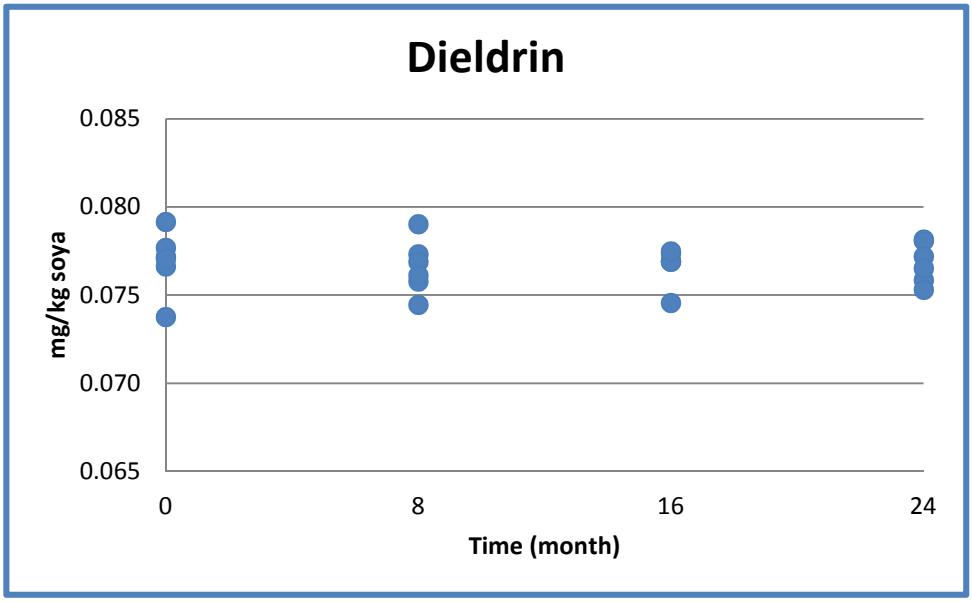


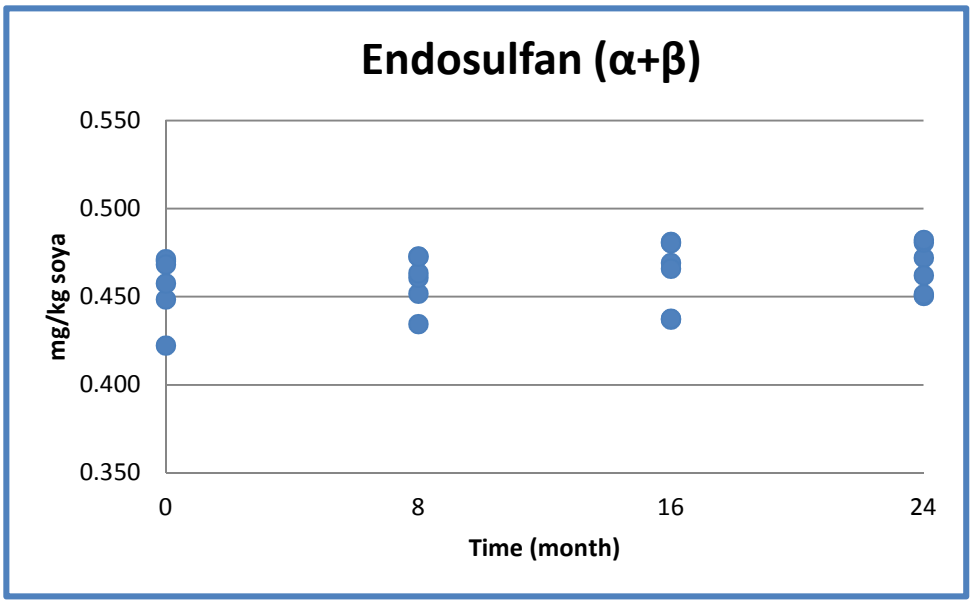


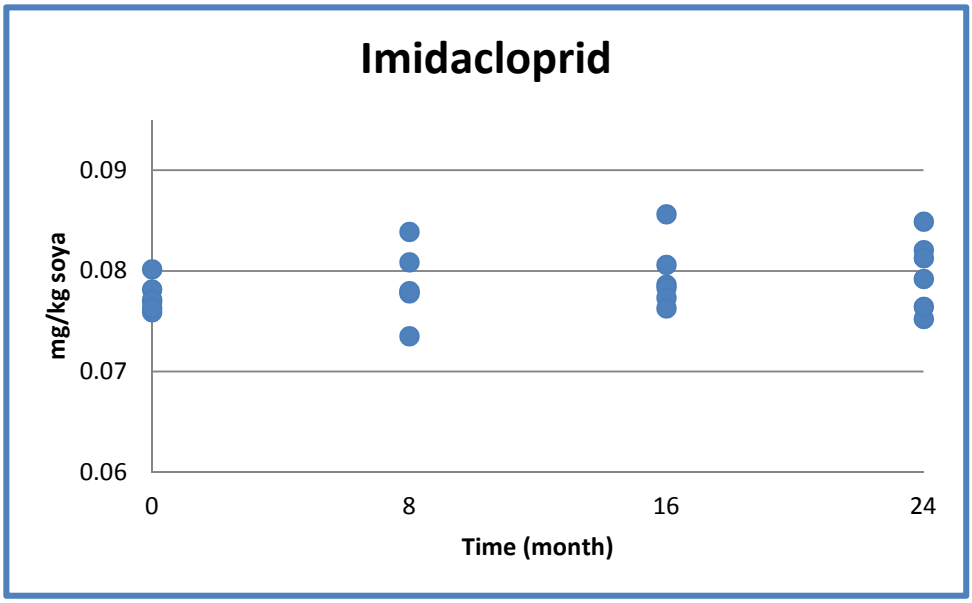


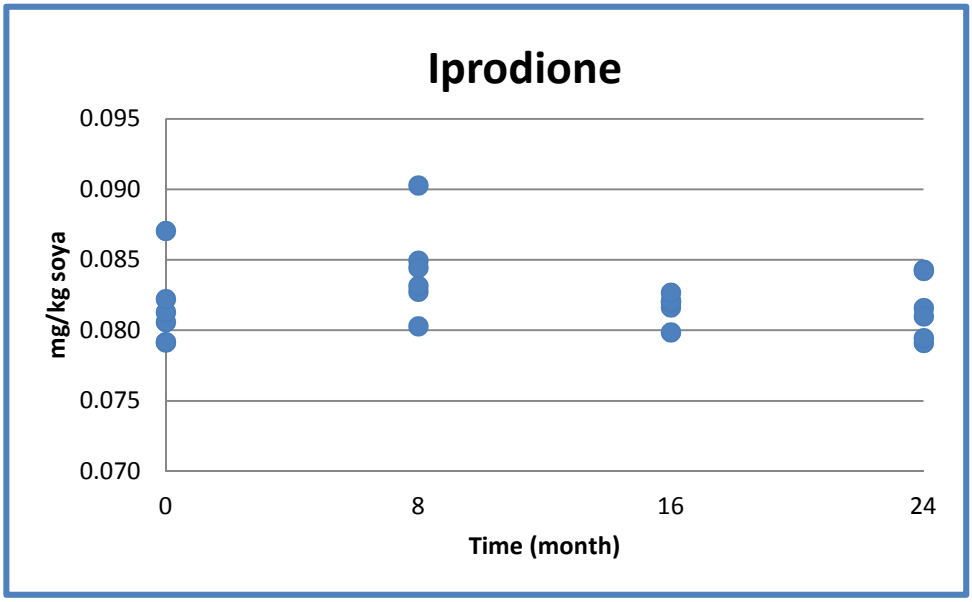

Cypermethrin
0.075
0.070
0.065
0.060
mg/kg soya
0
8
16
24
Time (month)

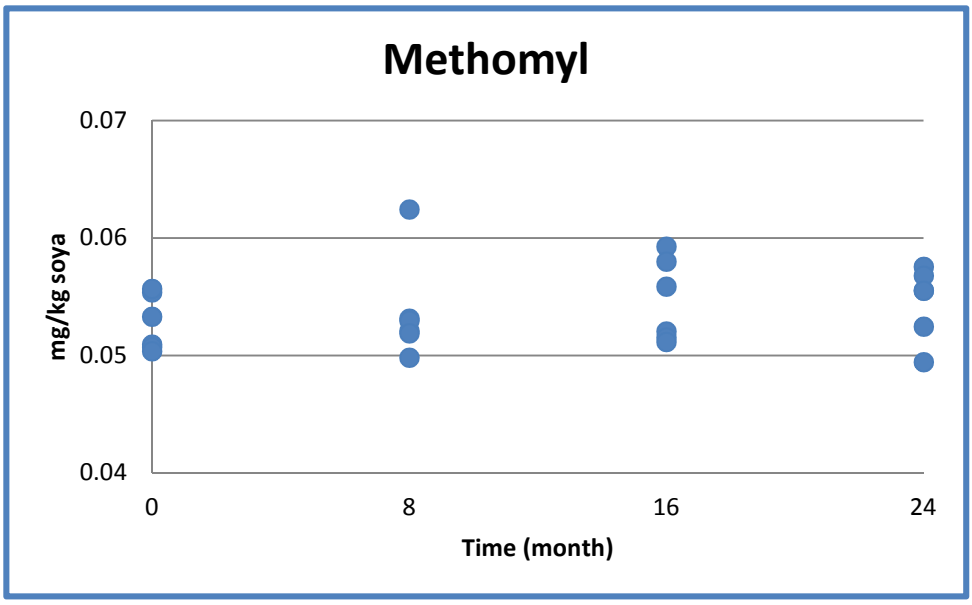

Diazinon
0.060
0.055
0.050
0.045
mg/kg soya
0
8
16
24
Time (month)

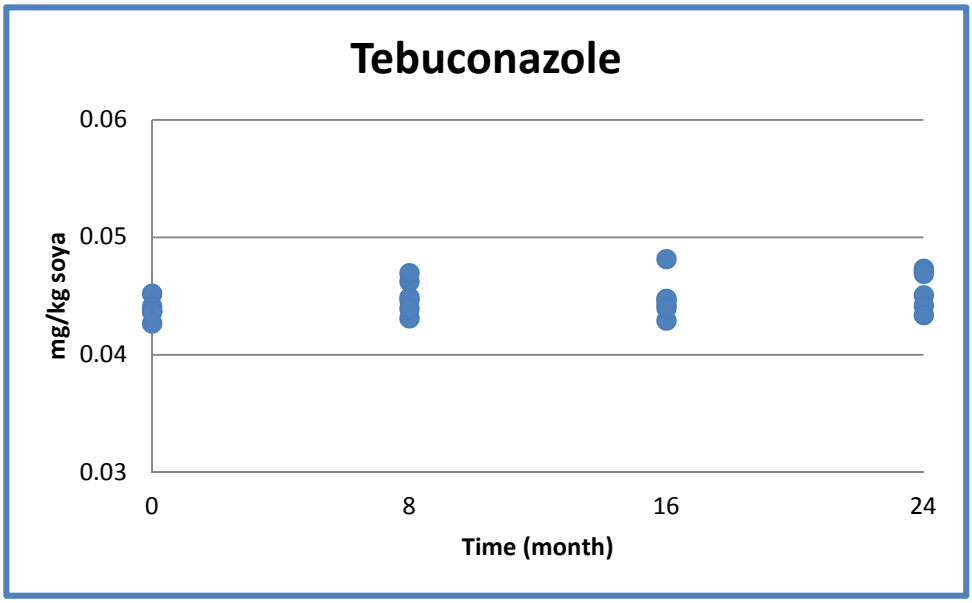

Dieldrin
0.085
0.080
0.075
0.070
0.065
mg/kg soya
0
8
16
24
Time (month)


Endosulfan (α+β)
0.550
0.500
0.450
0.400
0.350
mg/kg soya
0
8
16
24
Time (month)


Imidacloprid
0.09
0.08
0.07
0.06
mg/kg soya
0
8
16
24
Time (month)


Iprodione
0.095
0.090
0.085
0.080
0.075
0.070
mg/kg soya
0
8
16
24
Time (month)


Methomyl
0.07
0.06
0.05
0.04
mg/kg soya
0
8
16
24
Time (month)


Tebuconazole
0.06
0.05
0.04
0.03
mg/kg soya
0
8
16
24
Time (month)

Annex E. Summary of Analytical methods used for characterisation of ERM-BC700

Table E.1 Details of analytical methods, as given by the laboratories where gas chromatography was applied for the determination of pesticides in soya.

LABORATORY CODE	Sample intake (g)	Internal Standard	Extraction	Clean up	Injection technique	Stationary phase and dimensions analytical column	Analytical column	Calibration	Ionisation technique	Mass analyser/detector
01	2	Azoxystrobin D4 Chlorpyrifos D10, Cypermethrin $^{13}C_6$ Diazinon D10 Dieldrin $^{13}C_{12}$ Endosulfan b $^{13}C_9$ Iprodione D7	ASE, EtAc 100%, 1500 psi, 90°C, static 10 min, 1 cycle	GPC 55g BioBead, EtAc/Cyclohexane 1:1, followed by Supelco C18 SPE and Supelco PSA SPE	Slit-splitless	(20 m, 0.25 mm, 0.25 µm)	TG-SQC	Response factor	EI	Triple quadrupole
02	5	TPhP	QuEChERS – citrate buffered (EN15662)	Supel QuE Z-Sep	On column	5% Phenyl 95% dimehylpolysiloxane (30 m, 0.25 mm, 0.25 µm)	HP-5MS(I)	Calibration curve	EI	Triple quadrupole
03	5	TPhP	QuEChERS – citrate buffered (EN15662)	dSPE with PSA and C18e	On column	5% Phenyl 95% dimehylpolysiloxane (30 m, 0.25 mm, 0.25 µm)	HP-5MS UI	Matrix matched	EI	Triple quadrupole
04	2	PCB-31	QuEChERS – citrate buffered	-	PTV	5% Phenyl 95% dimehylpolysiloxane (30 m, 0.25 mm, 0.25 µm)	HP-5MS UI	Matrix matched	EI	Triple quadrupole
05	5	TPhP	10 mL water + 10 mL acetonitrile	freezing out - d-SPE PSA/C18 (25mg PSA/25 mg C18)/mL	Split-splitless	Proprietary (30 m, 0.25 mm, 0.2 µm)	Restek Rtx pesti-cides-2		EI	Triple quadrupole
06	5	-	Ethyl acetate/Cyclohexane 1:1 extraction	PSA/C18 (200+200 mg), filtration	Split-splitless	(5%-Phenyl)-methylpolysiloxane (30m, 0.25 mm, 0.25 µm)	HP-5ms Ultra Inert	Matrix matched	EI	Triple quadrupole
07	5	Cypermethrin D5 Chlorpyrifos D10 Diazinon D10 Endosulfan a D4 Endosulfan b D4 Dieldrin C13	Quechers Technique. 5 g sample + 15 mL Water + 10 mL acetonitrile	Quechers PSA clean-up	Split-splitless	5 % diphenyl/95% dimethyl polysiloxane (20 m, 0.18 mm, 0.18 µm)	Rxi-5Sil MS	Matrix matched	EI	Triple quadrupole
08	5	-	QuEChERS - Citrate buffered (EN 151662)	Dispersive-SPE (PSA/MgSO4)	split-splitless	5% Phenyl 95% dimehylpolysiloxane (15m, 0.25 mm)	HP-5ms	Matrix matched	EI	Triple quadrupole

09	7.5	-	Miniluke Method	Sample homogenised with 45mls solvent(15 acetone, 15 Dichloromethane, 15 Petroleum Ether) 15g anhydrous Sodium Sulphate added, centrifuged. 30mls of sample taken and concentrated down to 5mls in Ethyl Acetate.	split-splitless	5% Phenyl 95% dimehylpolysiloxane (30 m, 0.25 mm, 0.25 µm)	HP-5MSI	Calibration curve	EI	Triple quadrupole
10	2	PCB108	According to QuChERS method (CEN 15662); extraction of 2.0 g of sample after addition of 10 ,0 g of distilled water with 10 mL acetonitrile for 1 min (mechanical shaking)	Phase separation by addition of a mixture of 4 g MgSO4, 1 g NaCl, 1 g Trisodium citrate dihydrate, 0.5 g Disodium hydrogen citrate sesquihydrate and centrifugation at 3500 g for 5 min; DSPE of supernatant by addition of 200 mg MgSO4 and 25 mg PSA/mL extract, 1 min mechanical shaking and centrifugation at 3500 g for 5 min	Split-splitless	95 % dimethylpolysiloxane, 5 % phenylsiloxane (30 m, 0.25 mm, 0.25 µm)	J&W DB5-MS	Matrix matched	EI	Triple quadrupole
11	5	-	5 g sample + 10 mL (water; 1 H)+ 10 mL (acidified acetonitrile (1%) with acetic acid); evaporation of 6 mL and reconstituted with 1 mL (isooctane:toluene 90:10 v/v)	None	Split-splitless	(50%-phenyl)-methylpolysiloxane (30 m, 0.53 mm, 0.25 µm)	HP-50	Matrix matched		µECD
12	5	TPhP	The QuEChERS method: 5g sample/5 H2O/10 mL Acetonitrile 1% Acetic Acid plus 1ppm Tphp. 1h freezing. 5 g MgSO4/AcONa (4:1). Shake 1' and centrifuge 4'/4000 rpm.	4mL of acetonitrile extract with 1,0g MgSO4/0,6g PSA / 0,4g C18. Shake and centrifuge 4'/4000 rpm.	Split-splitless	Crossbond, similar to 5% diphenyl/95% dimethyl polysiloxane (30 m, 0.25 mm, 0.25 µm)	Restek Rxi-5Sil MS	Calibration curve	IE	Triple quadrupole

Table E.2: m/z transitions used for quantitative (in bold) and qualitative purposes for the different target compounds (gas chromatography)

LAB CODE	Azoxystrobin	Chlorpyrifos	Cypermethrin	Diazinon	Dieldrin	α-Endosulfan	β-Endosulfan	Iprodione	Tebuconazole	Other
1	**344->329** 388->345 **D4 348->332** D4 348->156	**315.9->259.9** 313.9->257.7 **D10 324.1->259.8** D10 326.0->261.8	**181.1->151.9** 181.1->152.6 **$^{13}C_6$ 187.2->157.8** $^{13}C_6$ 215.2->115.8	**179.1->136.9** 304.1->179.9 **D10 183.3->123** D10 183.3->123.8	263->193 279->206 **$^{13}C_{12}$ 270->200** $^{13}C_{12}$272->200	**338.8->195** 338.8->197	**338.8->195** 338.8->197 **$^{13}C_9$ 347.9->131.7** $^{13}C_9$ 348->167	**314.1->244.9** 316.1->246.6 **D7 320.0->192.1** D7 318.1->218.0		
2		**313.8->258.0** 314.0->286.0		**304.0->179.0** 179.1->137.2	**263.0->193.0** 263.0->191	**238.8->204.0** 229.0->194.0	**238.8->204.0** 229.0->194.0			
3		**314 -> 258** 314 -> 286	**163 -> 127** 180,8 -> 151,8 180,8 -> 126,8		**263,2 -> 193,2** 263,2 -> 191,2 279 -> 243	**241 -> 206** 195 -> 125 339 -> 160	**241 -> 206** 195 -> 125 339 -> 160	**314 -> 56** 314 -> 245 314 -> 271		TPhP **326 -> 233** 326 -> 169
4	**344->329** 388->345	**314->258** 314->286	**181->127** 163->127	**179->137** 304->179	**263->193** 263->191	**241->206** 195->159	**241->206** 195->159	**314->245** 314->271	**250->125** 252->127	
5		**170->169** 314->258	**181->152** 163->127		**277->241** 263->193	**195->159** 195->125	**207->172** 195->125	**314->245** 314->271		
6		**314.0->258.0** 314.0->286.0	**163.0->127.0** 181.0->152.0		**263.0->193.0** 277.0->241.0	**241.0->206.0** 241.0->170.0	**207.0->172.0** 241.0->206.0	**314.0->56.0** 314.0->245.0		Endosulfan sulphate **272.0->237.0** 274.0->239.0
7		**314->258** 314->286 **D10 324->260** D10 324->292	**181->152** 181->127 **D5 185,2->156** D5 185,2->130,7	**304->179** 304->137 **D10 315->170** D10 315->137	**263->193** 263->228 **C13 270->200** C13 270->235	**339->159,3** 339->266,8 **D4 347->166** D4 347->275 D4 347->202	**339->159,3** 339->159,3 **D4 347->166** D4 347->275 D4 347->202			
8		**196.9 -> 169.1** 198.9 -> 171.0 313.8 -> 257.8	**163.0 -> 127.0** 163.0 -> 91.0 164.9 -> 91.0	**199.1 -> 93.0** 137.1 -> 84.0 137.1 -> 54.0	**262.9 -> 193.0** 262.9 -> 191.0 277.0 -> 241.0	**194.9 -> 159.0** 194.9 -> 125.0 194.9 -> 160.0	**206.9 -> 172.0** 194.9 -> 158.9 194.9 -> 124.9	**313.8 -> 55.9** 243.9 -> 187.0 187.0 -> 124.0		
9			**181.1->152.1** 181.1->127.1	**304.0->179.1** 304.0->137.1	**263.0->193.0** 263.0->191.0	**240.8->206.0** 195.0->159.0	**241.0->206.0** 195.0->159.0	**314.0->245.1** 314.0->271.0		
10			**163.0 -> 127.1** 163.0 -> 91.1 209.0 -> 141.1	**179.1 -> 121.1** 179.1 -> 137.2 199.1 -> 135.1	**262.9 -> 192.9** 262.9 -> 190.9 277.0 -> 241.0	**240.9 -> 205.9** 194.9 -> 159.0 240.9 -> 136.0	**206.9 -> 172.0** 195.0 -> 125.0 195.0 -> 159.0	**314.0 -> 245.0** 216.0 -> 187.0 314.0 -> 56.0		PCB 108 **326 -> 256** 328 -> 256
11										
12	**344.00->329.00** 344.00->156.00	**314.00->258.00** 314.00->286.00	**163.00->127.00** 181.00->127.00	**304.00->179.00** 304.00->164.00	**277.00->241.00** 277.00->206.00	**241.00->206.00** 195.00->160.00 241.00->170.00	**241.00->206.00** 195.00->160.00 241.00->170.00	**314.00->245.00** 314.00->271.00	**250.00->153.00** 250.00->163.00	TPhP **325.00->169.00** 325.00->231.00
14				**137->84** 304->179						

Annex E. Analytical methods used for characterisation of ERM-BC700

Table E.3 Details of analytical methods, as given by the laboratories where liquid chromatography was applied for the determination of pesticides in soya.

LABORATORY CODE	Sample intake (g)	Internal Standard	Extraction	Clean up	Stationary phase and dimensions analytical column	Analytical column	Calibration	Ionisation technique	Mass analyser/detector
01	1	Azoxystrobin D4, Carbendazim D4, Chlorpyrifos D10, Diazinon D10, Tebuconazole D6	LLE with 10 ml solvent mixture (Acetonitrile:water 75:25 v/v)	extract filtered (cellulose), filtrate collected in volumetric flask (20 ml), (gravi-metrically control), an aliquot is filtered by syringe filter	C18 (100 mm, 2.1 mm, 1.7 µm)	Waters Acquity UPLC BEH C18	Calibration curve	ESI	Triple quadrupole
02	5	-	QuEChERS Citrate buffered (EN 15662)	Supel QuE Z-sep	C18 (100mm, 2.1 mm)	Zorbax Eclipse Plus C18	Calibration curve	ESI	Triple quadrupole
03	5	D4-Carbendazim, TPhP	QuEChERS citrate buffered (EN15662)	dSPE with PSA and C18e	Endcapped C18 (50 mm, 2mm, 1.8 µm)	Macherey-Nagel EC 50/2 Nucleodur C18 Gravity	Matrix matched	ESI	Triple quadrupole
04	2	Sulfotep	QuEChERS citrate buffered	-	C18 (100 mm, 2.1 mm, 1.8 µm)	Zorbax Eclipse Plus C18	Matrix matched	ESI	Triple quadrupole
05	5	-	10 mL water + 10 mL Acetonitrile	freezing out - d-SPE PSA/C18 (25mg PSA/25 mg C18)/mL	C18 (100 mm, 3.0 mm)	Phenomenex Kinetex		ESI	Triple quadrupole
06	5	-	10 ml H2O+10 ml acified EtOAc	-	C18 (150 mm, 2.1 mm, 1.8 µm)	Waters HSS T3	Matrix matched	ESI	Triple quadrupole
07	5	Azoxystrobin D4 Carbendazim D3 Imidacloprid D4 Iprodione D7 Methomyl D3 Tebuconazole D9	Quechers Technique. 5 g sample + 15 mL Water + 10 mL acetonitrile	Quechers PSA clean-up	C18 (100 mm, 2.1 mm)	Intensity Solo HPLC column	Matrix matched	ESI	Triple quadrupole
08	5	-	QuEChERS - Citrate buffered (EN 151662)	-	EC-18 (100 mm, 2.1	Agilent Poroshell 120	Matrix matched	ESI	Triple quadrupole

					mm, 2.7 µm)				
09	7.5	-	Miniluke Method	Sample homogenised with 45mls solvent(15 acetone, 15 Dichloro-methane, 15 Petro-leum Ether) 15g anhydrous Sodium Sulphate added, centrifuged. 30mls of sample taken and concentrated down to 5mls in Ethyl Acetate. Sample filtered. This is the GC extract (1g/ml). Dilute 1:20 in Methanol for LC fraction.	C18 100A	Kinetex 2.6µm C18 100A	Calibration curve	ESI	Triple quadrupole
10	2	TPhP	According to QuChERS method (CEN 15662); extraction of 2.0 g of sample after addition of 10 ,0 g of distilled water with 10 mL acetonitrile for 1 min (mechanical shaking)	Phase separation by addition of a mixture of 4 g MgSO4, 1 g NaCl, 1 g Trisodium citrate dihydrate, 0.5 g Disodium hydrogen citrate sesquihydrate and centrifugation at 3500 g for 5 min; DSPE of supernatant by addition of 200 mg MgSO4 and 25 mg PSA/mL extract, 1 min mechanical shaking and centrifugation at 3500 g for 5 min	Phenyl (RP phase) (10 0 mm, 2.1 mm, 1.7 µm)	Waters BEH phenyl	Matrix matched	ESI	Triple quadrupole
11	2	-	2g of sample + 10 mL Water; 1 H. + 10mL acidified acetonitrile (1% acetic acid)+(4.0g MgSO4 + 1.0g NaCl +1.0g trisodium citrate dihydrate +0.5g disodium hydrogencitrate sesquihydrate)	-	RP 100A (100x2.00m m, 2.5 µm)	phenomenex Synergi 2.5u,Fusion	Standard addition	ESI	Triple quadrupole
12	5	TPhP	The QuEChERS method: 5g sample/5 H2O/10 mL Acetonitrile 1% Acetic Acid plus 1ppm Tphp. 1h freezing. 5 g MgSO4/AcONa (4:1). Shake 1' and centrifuge 4'/4000 rpm.	4mL of acetonitrile extract with 1,0g MgSO4/0,6g PSA / 0,4g C18. Shake and centrifuge 4'/4000 rpm.	C18 Selectivity (100 mm, 2.1 mm)	Thermo Hypersil GOLD	Calibration curve	ESI	Triple quadrupole

Table E.4: m/z transitions used for quantitative (in bold) and qualitative purposes for the different target compounds (liquid chromatography)

LAB CODE	Azoxystrobin	Carbendazim	Chlorpyrifos	Cypermethrin	Diazinon	Imidacloprid	Iprodione	Methomyl	Tebuconazole	Other
1	**404->329** 404->344 404->172 **D4 408->333**	**192 -> 160** 192 -> 132 192 -> 105 **D4 196 -> 164**	**349.9 -> 197.9** 351.9 -> 199.9 349.9 -> 124.9 **D10 360 -> 131**		**305 -> 169** 305 -> 153 305 -> 97 **D10 315 -> 170**				**308 -> 70** 310 -> 70 308 -> 125 **D6 314 -> 72**	
2	**404.0->372.0** 404.0->344.1	**192.1->160.0** 192.1->132.0		**433.2->191.2** 433.2->127.0 435.1->193.1		**256.1->175.1** 256.1->209.0	**330.0->245.0** 330.0->56.0	**163.1->88.1** 163.1->106.0	**308.2->70.1** 308.2->124.8	
3	**404 -> 372** 404 -> 344	**192,1 -> 160** 192,1 -> 132			**305,1 -> 169** 305,1 -> 96,9	**256,1 -> 209** 256,1 -> 175,1		**163,1 -> 106** 163,1 ->88,1	**308,2 -> 125** 308,2 -> 70,1	D4->Carbendazim **196,1 -> 164** 196,1 -> 136,1 Triphenylphosphate **327 -> 77** 327 -> 152
4		**192->160** 192->132				**256->209** 256->175		**163->88** 163->106		
5	**404->372** 404->344	**192->160** 192->132			**305->169** 305->153	**256->209** 256->175		**163->88** 163->106	**308->70** 308->125	
6	**404.0->372.0** 404.0->344.0	**192.1->160.0** 192.1->132.0			**305.0->169.0** 305.0->153.0	**256.1->175.1** 256.1->209.0		**163.1->88.1** 163.1->106.0	**308.2->70.1** 308.2->124.9	
7	**404->344** 404->328 **D4 408->348** D4 408->332	**192->160** 192->132 **D3 195,2->160** D3 195,2->132				**256->209** 256->175 **D4 260->213** D4 260->179	**330,2->245** 332,2->247 **D7 337,2->245** D7 339,2->247	**163->88** 163->106 **D3 166->88,2** D3 166->106,2	**308,2->70** 308,2->125 **D9 317,3->70** D9 317,3->125	
8	**404.0 -> 372.0** 404.0 -> 344.0	**192.0 -> 160.0** 192.0 -> 132.0				**256.0 -> 175.0** 256.0 -> 209.0		**163.0 -> 88.0** 163.0 -> 106.0	**308.0 -> 70.0** 308.0 -> 125.0	
9	**404.1->372.1** 404.1->344.1	**192.1->160.1** 192.1->132.1	**350.0->96.9** 350.0->197.8			**256.1->209.1** 256.1->175.0		**163.0->88.0** 163.0->106.0	**308.2->70.0** 308.2->125.0	
10	**404 -> 372** 404 -> 344	**192.1 -> 160.1** 192.1 -> 132.1	**350 -> 97** 350 -> 198			**256.06 -> 175.1** 256.06 -> 209.1		**162.9 -> 87.8** 162.9 -> 105.9	**307.93 -> 69.8** 307.93 -> 124.74	TPhP **327 -> 77**
11	**404.2->372.3** 404.2->329.0	**192.2->160.1** 192.2->132.0	**351.9->200** 351.9->97.1 349.9->197.8		**305.1->169** 305.1->153 305.1->96.9	**256.1->209.0** 256.1->175.0		**163.1->87.9** 163.1->105.9	**308.1->70.0** 308.1->125.0	
12		**192,00->160,05** 192,00->132,10				**256,20->209,10** 256,20->175,15		**163,20->88,10** 163,20->106,15		TPhP **325.00->169.00** 325.00->231.00
13	**404->372** 404->344		**350->198** 350->97		**305->169** 305->153		**330->245** 330->288		**308->70** 308->125	

Annex F: Results from the characterisation of ERM-BC700

Table F.1. Mass fractions of azoxystrobin in soya as reported by participant laboratories.

LABORATORY CODE	Replicate 1 [mg/kg]	Replicate 2 [mg/kg]	Replicate 3 [mg/kg]	Replicate 4 [mg/kg]	Replicate 5 [mg/kg]	Replicate 6 [mg/kg]	Mean [mg/kg]	S [mg/kg]
L01-LC-MS/MS	0.464	0.452	0.471	0.469	0.473	0.473	**0.467**	**0.008**
L02-LC-MS/MS	0.510	0.517	0.500	0.503	0.517	0.500	**0.508**	**0.008**
L03-LC-MS/MS	0.365	0.366	0.359	0.374	0.361	0.363	**0.365**	**0.005**
L04-GC-MS/MS	0.418	0.402	0.359	0.442	0.442	0.473	**0.423**	**0.040**
L05-LC-MS/MS	0.451	0.43	0.404	0.511	0.635	0.549	**0.497**	**0.086**
L06-LC-MS/MS	0.48	0.48	0.49	0.50	0.49	0.50	**0.490**	**0.009**
L07-LC-MS/MS	0.32	0.34	0.31	0.35	0.37	0.36	**0.342**	**0.023**
L08-LC-MS/MS	0.60	0.57	0.57	0.54	0.50	0.53	**0.552**	**0.035**
L09-LC-MS/MS	0.27	0.29	0.35	0.37	0.27	0.40	**0.325**	**0.056**
L10-LC-MS/MS	0.432	0.445	0.45	0.422	0.439	0.422	**0.435**	**0.012**
L11-LC-MS/MS	0.6556	0.6716	0.6663	0.6795	0.6634	0.685	**0.670**	**0.011**
L12-GC-MS/MS	0.436	0.401	0.407	0.515	0.516	0.521	**0.466**	**0.058**
L13-LC-MS/MS	0.445	0.467	0.446	0.460	0.429	0.446	**0.449**	**0.013**
L14-GC-MS/MS	0.388	0.398	0.390	0.395	0.396	0.406	**0.396**	**0.006**

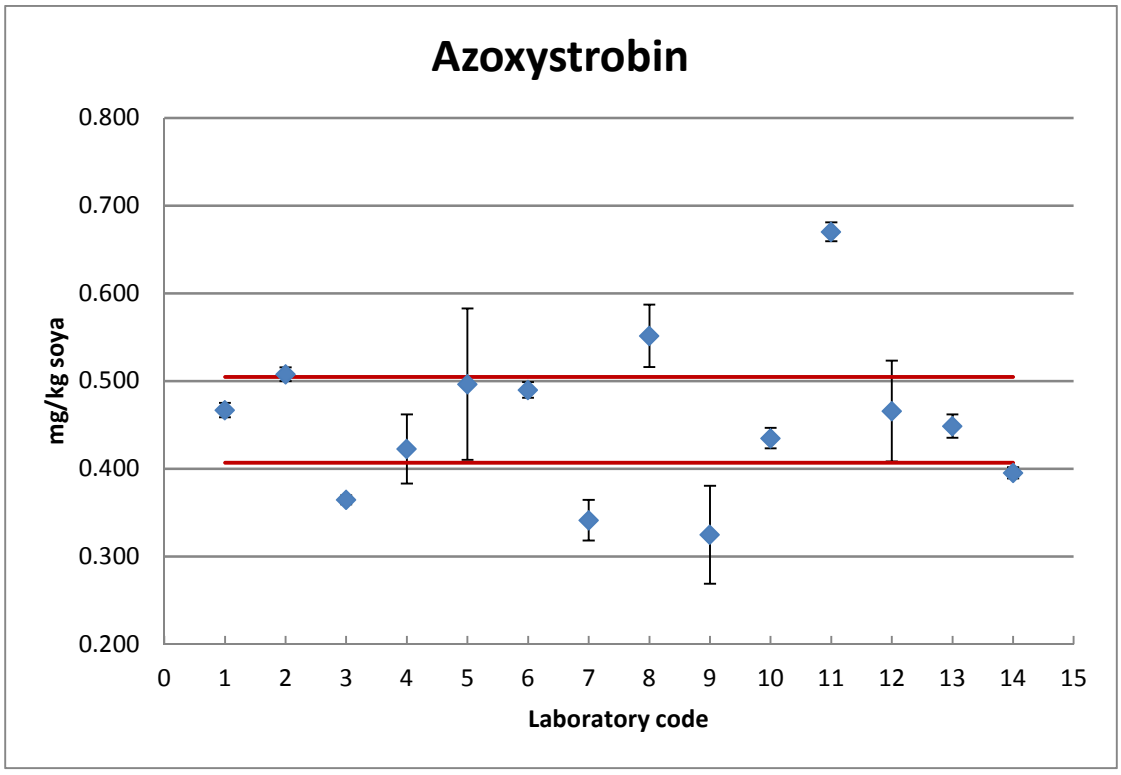


Figure F.1. **L**aboratory means with their standard deviations represented as error bars. Red lines correspond to the mean of laboratory means ± U_{CRM}

Table F.2. Mass fractions of carbendazim in soya as reported by participant laboratories.

LABORATORY CODE	Replicate 1 [mg/kg]	Replicate 2 [mg/kg]	Replicate 3 [mg/kg]	Replicate 4 [mg/kg]	Replicate 5 [mg/kg]	Replicate 6 [mg/kg]	Mean [mg/kg]	S [mg/kg]
L01-LC-MS/MS	0.246	0.249	0.242	0.242	0.243	0.240	**0.244**	**0.003**
L02- LC-MS/MS	0.182	0.182	0.177	0.178	0.175	0.175	**0.178**	**0.003**
L03-LC-MS/MS	0.135	0.140	0.134	0.142	0.134	0.14	**0.138**	**0.004**
L04-LC-MS/MS	0.196	0.209	0.197	0.212	0.206	0.201	**0.204**	**0.007**
L05-LC-MS/MS	0.199	0.213	0.203	0.222	0.273	0.249	**0.227**	**0.029**
L06-LC-MS/MS	0.22	0.22	0.22	0.22	0.22	0.22	**0.220**	**0.000**
L07-LC-MS/MS	0.21	0.20	0.18	0.19	0.21	0.20	**0.198**	**0.012**
L08-LC-MS/MS	0.19	0.19	0.19	0.20	0.19	0.18	**0.190**	**0.006**
L09-LC-MS/MS	0.16	0.15	0.18	0.19	0.16	0.19	**0.172**	**0.017**
L10-LC-MS/MS	0.184	0.18	0.183	0.185	0.177	0.174	**0.181**	**0.004**
L11-LC-MS/MS	0.2171	0.2232	0.2066	0.1898	0.208	0.2152	**0.210**	**0.012**
L12-LC-MS/MS	0.218	0.218	0.249	0.180	0.185	0.176	**0.204**	**0.029**

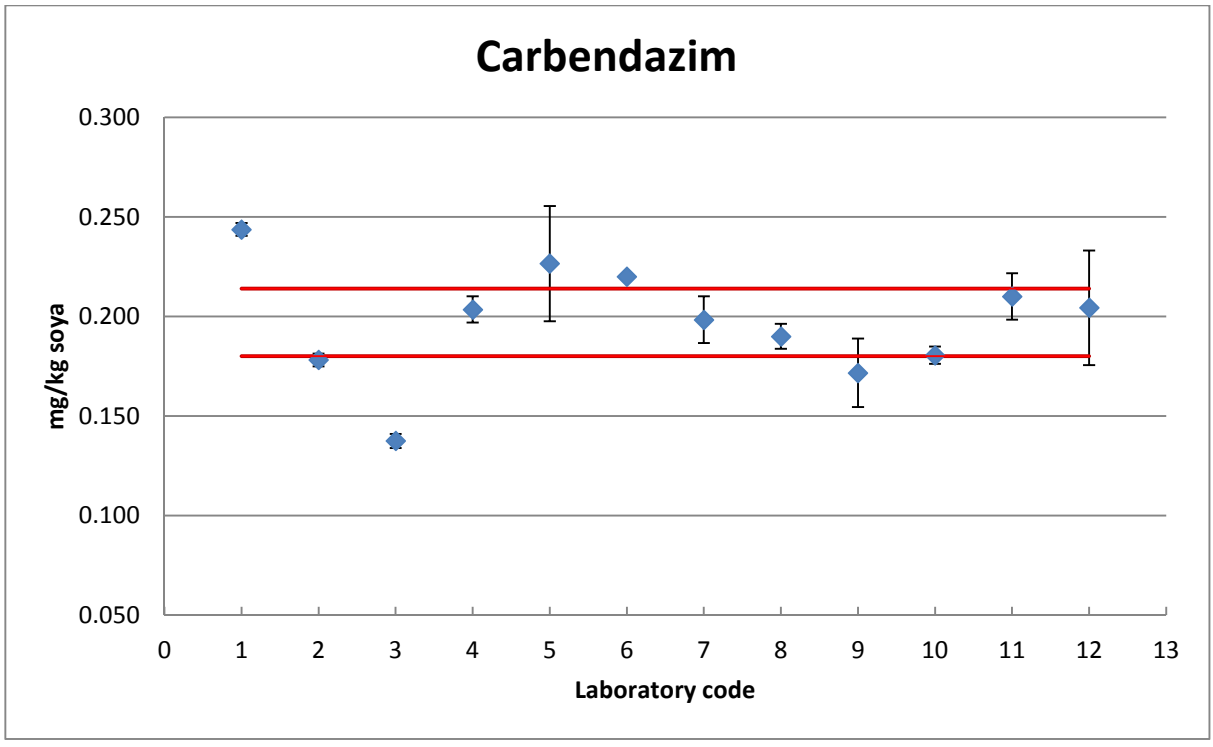


Figure F.2. **L**aboratory means with their standard deviations represented as error bars. Red lines correspond to the mean of laboratory means ± U_{CRM}

Table F.3. Mass fractions of chlorpyrifos in soya as reported by participant laboratories.

LABORATORY CODE	Replicate 1 [mg/kg]	Replicate 2 [mg/kg]	Replicate 3 [mg/kg]	Replicate 4 [mg/kg]	Replicate 5 [mg/kg]	Replicate 6 [mg/kg]	Mean [mg/kg]	S [mg/kg]
L01-LC-MS/MS	0.0804	0.0781	0.0803	0.0833	0.0794	0.0806	**0.080**	**0.002**
L02-GC-MS/MS	0.0613	0.0644	0.0632	0.0563	0.0576	0.0576	**0.060**	**0.003**
L03-GC-MS/MS	0.051	0.046	0.053	0.053	0.047	0.047	**0.050**	**0.003**
L04-GC-MS/MS	0.070	0.069	0.067	0.072	0.075	0.073	**0.071**	**0.003**
L05-GC-MS/MS	0.077	0.078	0.077	0.080	0.090	0.083	**0.081**	**0.005**
L06-GC-MS/MS	0.067	0.067	0.067	0.068	0.067	0.064	**0.067**	**0.001**
L07-GC-MS/MS	0.061	0.061	0.062	0.073	0.073	0.074	**0.067**	**0.007**
L08-GC-MS/MS	0.069	0.068	0.069	0.068	0.064	0.062	**0.067**	**0.003**
L09-LC-MS/MS	0.048	0.054	0.055	0.059	0.052	0.055	**0.054**	**0.004**
L10-LC-MS/MS	0.062	0.060	0.064	0.059	0.060	0.058	**0.061**	**0.002**
L11-LC-MS/MS	0.06578	0.07106	0.06479	0.0671	0.0649	0.066	**0.067**	**0.002**
L13-LC-MS/MS	0.065	0.070	0.067	0.068	0.068	0.058	**0.066**	**0.004**
L14-GC-MS/MS	0.075	0.076	0.075	0.077	0.076	0.076	**0.076**	**0.001**

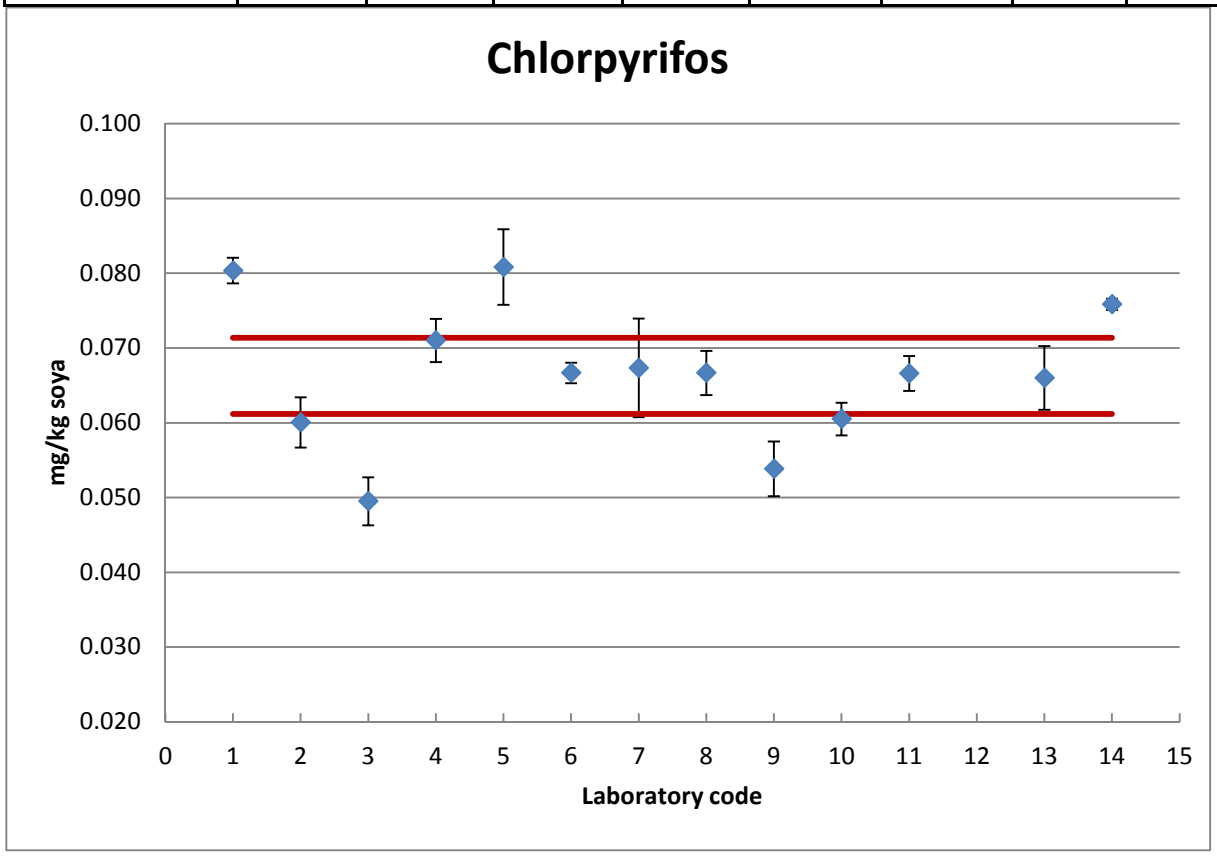


Figure F.3. **L**aboratory means with their standard deviations represented as error bars. Red lines correspond to the mean of laboratory means ± U_{CRM}

Table F.4. Mass fractions of cypermethrin in soya as reported by participant laboratories.

LABORATORY CODE	Replicate 1 [mg/kg]	Replicate 2 [mg/kg]	Replicate 3 [mg/kg]	Replicate 4 [mg/kg]	Replicate 5 [mg/kg]	Replicate 6 [mg/kg]	Mean [mg/kg]	S [mg/kg]
L01-GC-MS/MS	0.071	0.070	0.070	0.071	0.071	0.072	**0.071**	**0.001**
L02-LC-MS/MS	0.0449	0.0479	0.0453	0.0452	0.0423	0.0420	**0.045**	**0.002**
L03-GC-MS/MS	0.054	0.054	0.056	0.054	0.055	0.052	**0.054**	**0.001**
L04-GC-MS/MS	0.061	0.060	0.060	0.060	0.062	0.061	**0.061**	**0.001**
L05-GC-MS/MS	0.059	0.060	0.060	0.054	0.062	0.061	**0.059**	**0.003**
L06-GC-MS/MS	0.065	0.063	0.064	0.069	0.065	0.067	**0.066**	**0.002**
L07-GC-MS/MS	0.059	0.055	0.056	0.065	0.061	0.072	**0.061**	**0.006**
L08-GC-MS/MS	0.044	0.043	0.044	0.045	0.043	0.044	**0.044**	**0.001**
L10-GC-MS/MS	0.035	0.036	0.037	0.035	0.036	0.034	**0.036**	**0.001**
L11-GC-ECD	0.0253	0.0253	0.0286	0.0259	0.034	0.0223	**0.027**	**0.004**

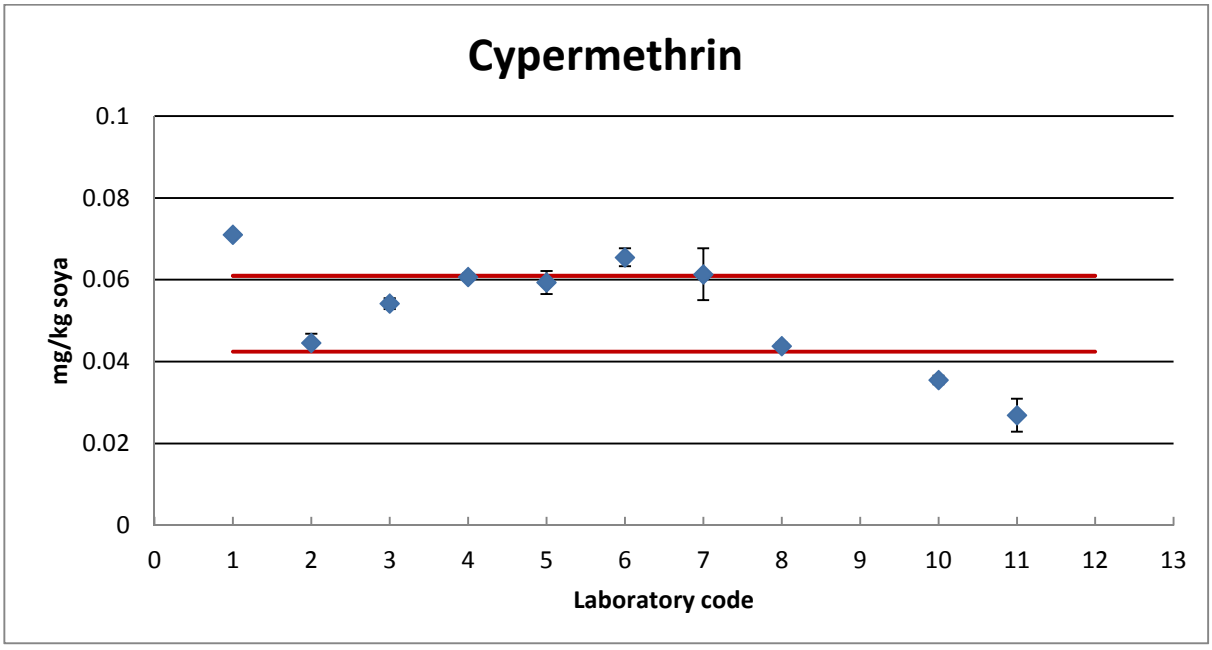


Figure F.4. **L**aboratory means with their standard deviations represented as error bars. Red lines correspond to the mean of laboratory means ± U_{CRM}

Table F.5. Mass fractions of diazinon in soya as reported by participant laboratories.

LABORATORY CODE	Replicate 1 [mg/kg]	Replicate 2 [mg/kg]	Replicate 3 [mg/kg]	Replicate 4 [mg/kg]	Replicate 5 [mg/kg]	Replicate 6 [mg/kg]	Mean [mg/kg]	S [mg/kg]
L01-GC-MS/MS	0.060	0.059	0.060	0.060	0.059	0.061	**0.060**	**0.001**
L02- GC-MS/MS	0.0686	0.0732	0.0752	0.0617	0.0688	0.0667	**0.069**	**0.005**
L03-LC-MS/MS	0.060	0.057	0.056	0.058	0.057	0.058	**0.058**	**0.001**
L04-GC-MS/MS	0.067	0.064	0.060	0.070	0.067	0.076	**0.067**	**0.005**
L05-LC-MS/MS	0.060	0.061	0.059	0.083	0.083	0.088	**0.072**	**0.014**
L06-LC-MS/MS	0.071	0.071	0.072	0.071	0.069	0.071	**0.071**	**0.001**
L07-GC-MS/MS	0.055	0.051	0.055	0.06	0.059	0.064	**0.057**	**0.005**
L08-GC-MS/MS	0.074	0.069	0.074	0.074	0.068	0.067	**0.071**	**0.003**
L10-GC-MS/MS	0.048	0.050	0.050	0.048	0.047	0.049	**0.049**	**0.001**
L11-LC-MS/MS	0.07172	0.07656	0.06897	0.08085	0.08085	0.0814	**0.077**	**0.005**
L12-LC-MS/MS	0.068	0.072	0.075	0.087	0.09	0.086	**0.080**	**0.009**
L13-LC-MS/MS	0.067	0.064	0.062	0.061	0.064	0.060	**0.063**	**0.003**
L14-GC-MS/MS	0.073	0.081	0.074	0.080	0.086	0.088	**0.080**	**0.006**
L15-LC-MS/MS	0.076	0.0732	0.0732	0.0697	0.0719	0.0742	**0.073**	**0.002**

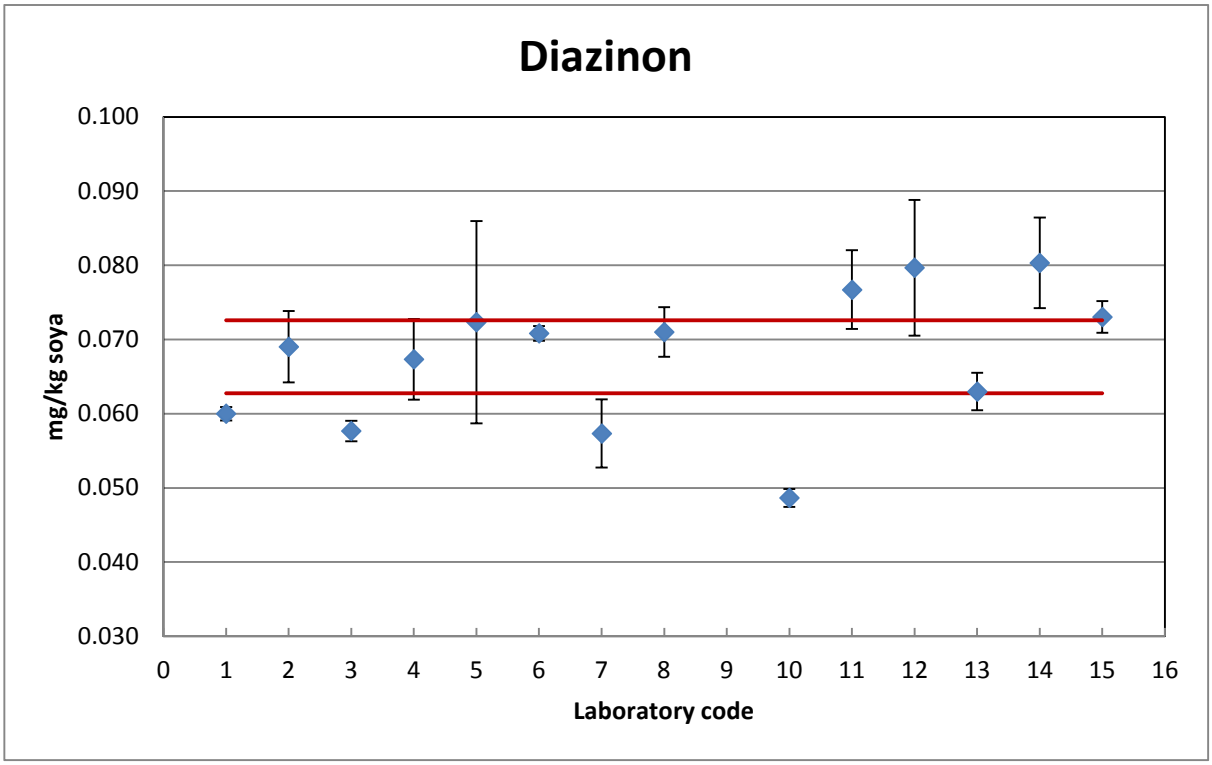


Figure F.5. **L**aboratory means with their standard deviations represented as error bars. Red lines correspond to the mean of laboratory means ± U_{CRM}

Table F.6. Mass fractions of dieldrin in soya as reported by participant laboratories.

LABORATORY CODE	Replicate 1 [mg/kg]	Replicate 2 [mg/kg]	Replicate 3 [mg/kg]	Replicate 4 [mg/kg]	Replicate 5 [mg/kg]	Replicate 6 [mg/kg]	Mean [mg/kg]	S [mg/kg]
L01-GC-MS/MS	0.086	0.086	0.086	0.085	0.086	0.086	**0.086**	**0.001**
L02-GC-MS/MS	0.0701	0.0734	0.0721	0.0629	0.0659	0.0629	**0.068**	**0.005**
L03-GC-MS/MS	0.054	0.059	0.063	0.060	0.053	0.058	**0.058**	**0.004**
L04-GC-MS/MS	0.096	0.084	0.080	0.081	0.083	0.084	**0.085**	**0.006**
L05-GC-MS/MS	0.088	0.084	0.093	0.075	0.076	0.086	**0.084**	**0.007**
L06-GC-MS/MS	0.084	0.082	0.082	0.085	0.083	0.081	**0.083**	**0.002**
L07-GC-MS/MS	0.061	0.063	0.060	0.074	0.071	0.073	**0.067**	**0.006**
L08-GC-MS/MS	0.073	0.074	0.074	0.074	0.073	0.072	**0.073**	**0.001**
L10-GC-MS/MS	0.063	0.063	0.063	0.056	0.059	0.055	**0.060**	**0.004**
L11-GC-ECD	0.0924	0.0885	0.087	0.0647	0.0874	0.0668	**0.081**	**0.012**
L12-GC-MS/MS	0.082	0.080	0.085	0.077	0.092	0.065	**0.080**	**0.009**

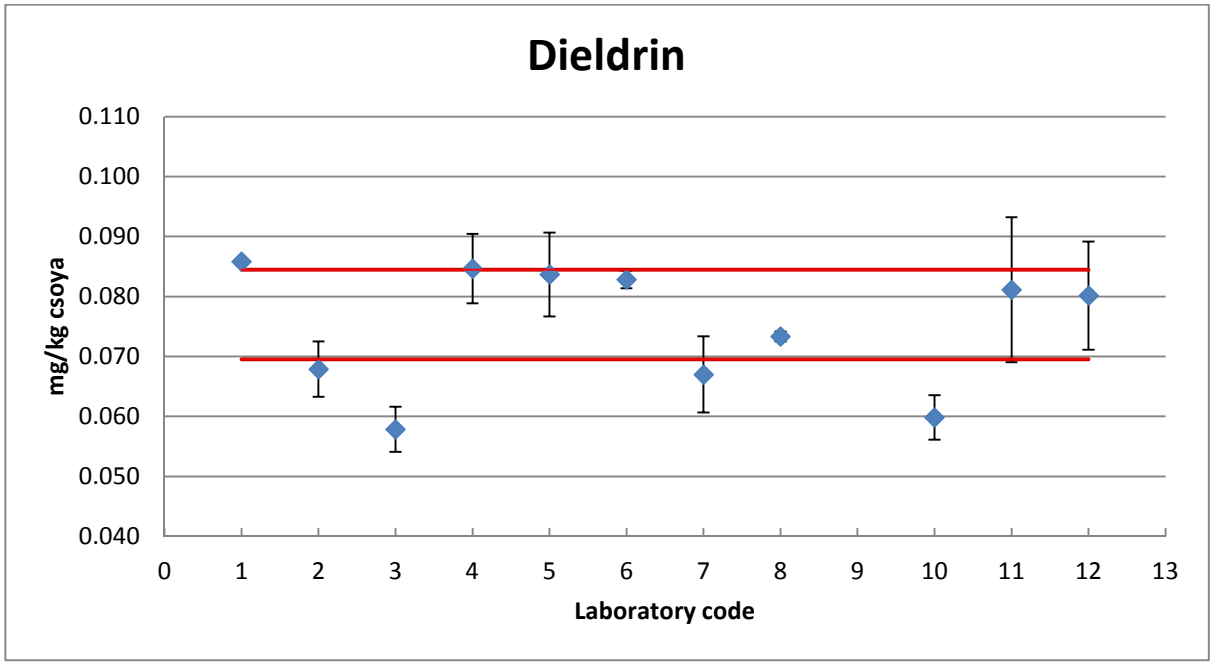


Figure F.6. **L**aboratory means with their standard deviations represented as error bars. Red lines correspond to the mean of laboratory means ± U_{CRM}

Table F.7. Mass fractions of endosulfan-(α+β) in soya as reported by participant laboratories.

LABORATORY CODE	Replicate 1 [mg/kg]	Replicate 2 [mg/kg]	Replicate 3 [mg/kg]	Replicate 4 [mg/kg]	Replicate 5 [mg/kg]	Replicate 6 [mg/kg]	Mean [mg/kg]	S [mg/kg]
L01-GC-MS/MS	0.582	0.580	0.557	0.536	0.562	0.539	**0.559**	**0.020**
L02-GC-MS/MS	0.510	0.525	0.512	0.461	0.467	0.457	**0.489**	**0.030**
L03-GC-MS/MS	0.445	0.414	0.461	0.460	0.397	0.436	**0.436**	**0.026**
L04-GC-MS/MS	0.549	0.556	0.534	0.562	0.594	0.628	**0.571**	**0.035**
L05-GC-MS/MS	0.583	0.630	0.620	0.589	0.655	0.663	**0.623**	**0.033**
L06-GC-MS/MS	0.48	0.47	0.48	0.49	0.47	0.46	**0.475**	**0.011**
L07-GC-MS/MS	0.46	0.43	0.44	0.48	0.48	0.52	**0.468**	**0.033**
L08-GC-MS/MS	0.51	0.51	0.51	0.51	0.51	0.49	**0.507**	**0.008**
L09-GC-MS/MS	0.42	0.38	0.40	0.46	0.40	0.41	**0.412**	**0.027**
L10-GC-MS/MS	0.393	0.403	0.398	0.363	0.373	0.363	**0.382**	**0.018**
L12-GC-MS/MS	0.472	0.452	0.470	0.418	0.410	0.402	**0.437**	**0.031**

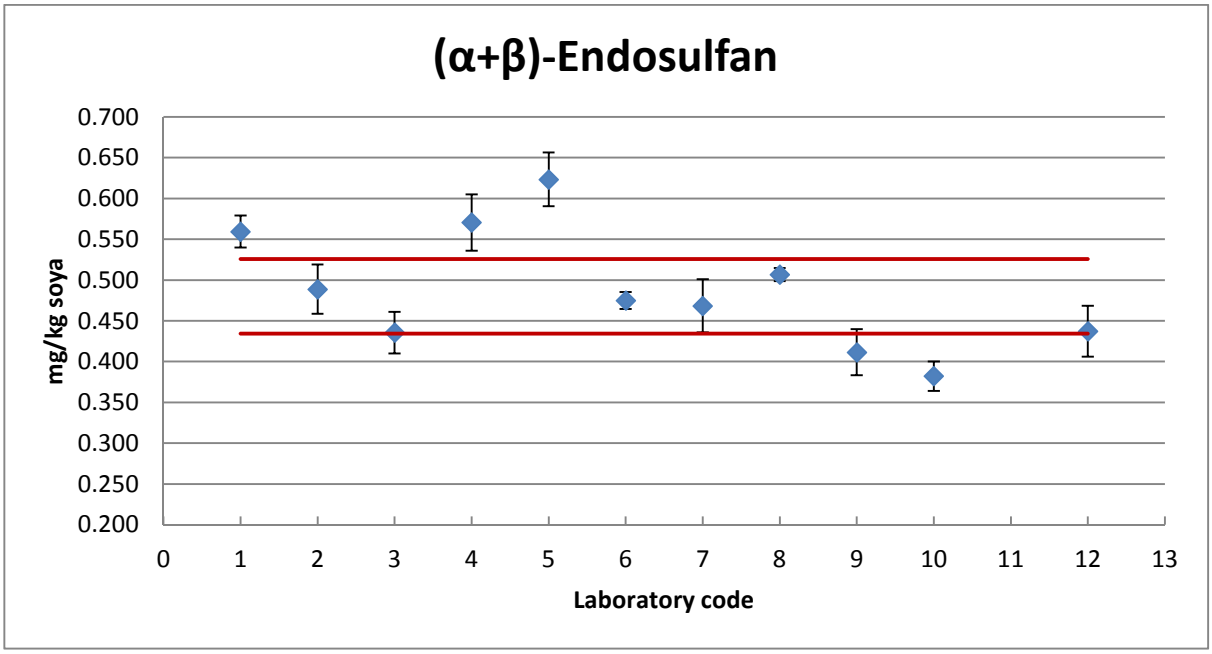


Figure F.7. **L**aboratory means with their standard deviations represented as error bars. Red lines correspond to the mean of laboratory means ± U_{CRM}

Table F.8. Mass fractions of imidacloprid in soya as reported by participant laboratories.

LABORATORY CODE	Replicate 1 [mg/kg]	Replicate 2 [mg/kg]	Replicate 3 [mg/kg]	Replicate 4 [mg/kg]	Replicate 5 [mg/kg]	Replicate 6 [mg/kg]	Mean [mg/kg]	S [mg/kg]
L02-LC-MS/MS	0.0748	0.0759	0.0712	0.0712	0.0759	0.0718	**0.074**	**0.002**
L03-LC-MS/MS	0.065	0.066	0.065	0.068	0.065	0.068	**0.066**	**0.002**
L04-LC-MS/MS	0.069	0.068	0.061	0.075	0.075	0.078	**0.071**	**0.006**
L05-LC-MS/MS	0.082	0.075	0.074	0.079	0.093	0.082	**0.081**	**0.007**
L06-LC-MS/MS	0.076	0.077	0.078	0.071	0.072	0.071	**0.074**	**0.003**
L07-LC-MS/MS	0.063	0.062	0.054	0.068	0.07	0.071	**0.065**	**0.006**
L08-LC-MS/MS	0.11	0.10	0.10	0.11	0.11	0.10	**0.105**	**0.006**
L09-LC-MS/MS	0.073	0.069	0.074	0.081	0.063	0.069	**0.072**	**0.006**
L10-LC-MS/MS	0.068	0.072	0.075	0.070	0.074	0.071	**0.072**	**0.003**
L11-LC-MS/MS	0.0869	0.0852	0.0813	0.0863	0.0819	0.0836	**0.084**	**0.002**
L12-LC-MS/MS	0.059	0.059	0.060	0.055	0.060	0.059	**0.059**	**0.002**

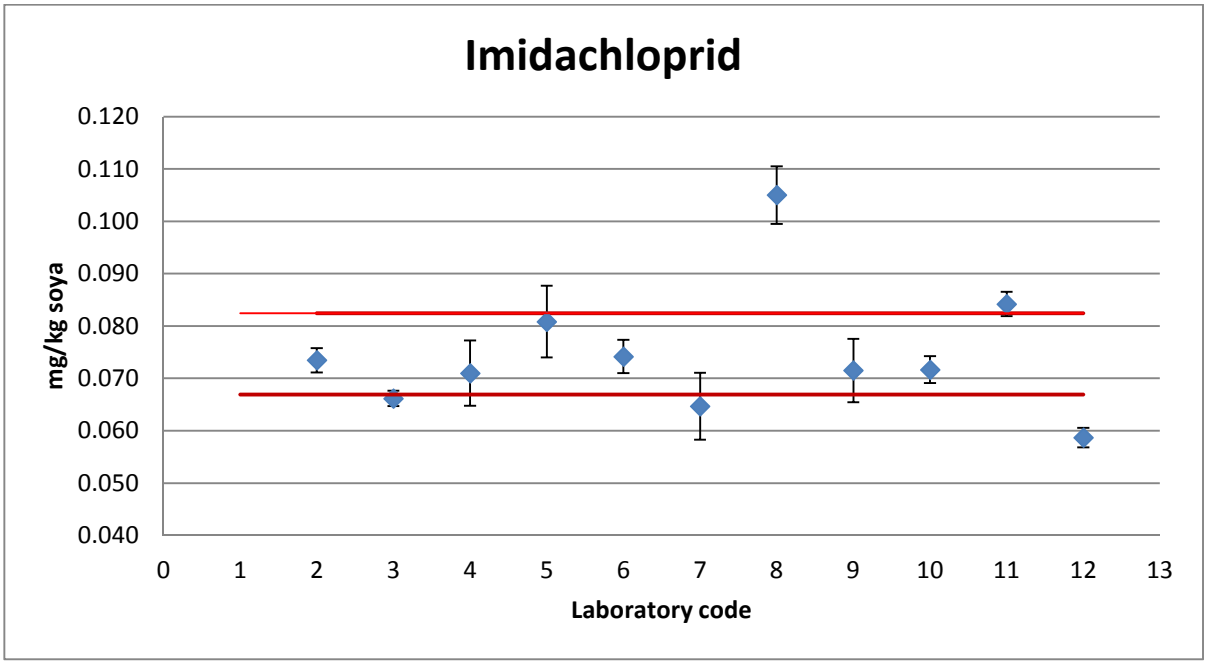


Figure F.8. **L**aboratory means with their standard deviations represented as error bars. Red lines correspond to the mean of laboratory means ± U_{CRM}

Table F.9. Mass fractions of iprodione in soya as reported by participant laboratories.

LABORATORY CODE	Replicate 1 [mg/kg]	Replicate 2 [mg/kg]	Replicate 3 [mg/kg]	Replicate 4 [mg/kg]	Replicate 5 [mg/kg]	Replicate 6 [mg/kg]	Mean [mg/kg]	S [mg/kg]
L01-GC-MS/MS	0.101	0.102	0.104	0.103	0.106	0.106	**0.104**	**0.002**
L02-LC-MS/MS	0.123	0.126	0.118	0.122	0.131	0.123	**0.124**	**0.004**
L03-GC-MS/MS	0.080	0.081	0.086	0.087	0.077	0.076	**0.081**	**0.005**
L04-GC-MS/MS	0.108	0.107	0.100	0.117	0.121	0.127	**0.113**	**0.010**
L05-GC-MS/MS	0.132	0.143	0.144	0.153	0.170	0.145	**0.148**	**0.013**
L06-GC-MS/MS	0.091	0.089	0.094	0.099	0.092	0.096	**0.094**	**0.004**
L07-LC-MS/MS	0.093	0.098	0.086	0.089	0.100	0.095	**0.094**	**0.005**
L08-GC-MS/MS	0.12	0.11	0.11	0.12	0.11	0.11	**0.113**	**0.005**
L09-GC-MS/MS	0.13	0.12	0.15	0.11	0.090	0.093	**0.116**	**0.023**
L10-GC-MS/MS	0.064	0.071	0.066	0.065	0.057	0.066	**0.065**	**0.005**
L11-GC-ECD	0.0736	0.0677	0.0742	0.0989	0.0987	0.1105	**0.087**	**0.018**
L12-GC-MS/MS	0.091	0.104	0.100	0.116	0.107	0.102	**0.103**	**0.008**
L13-LC-MS/MS	0.117	0.111	0.112	0.118	0.107	0.118	**0.114**	**0.005**

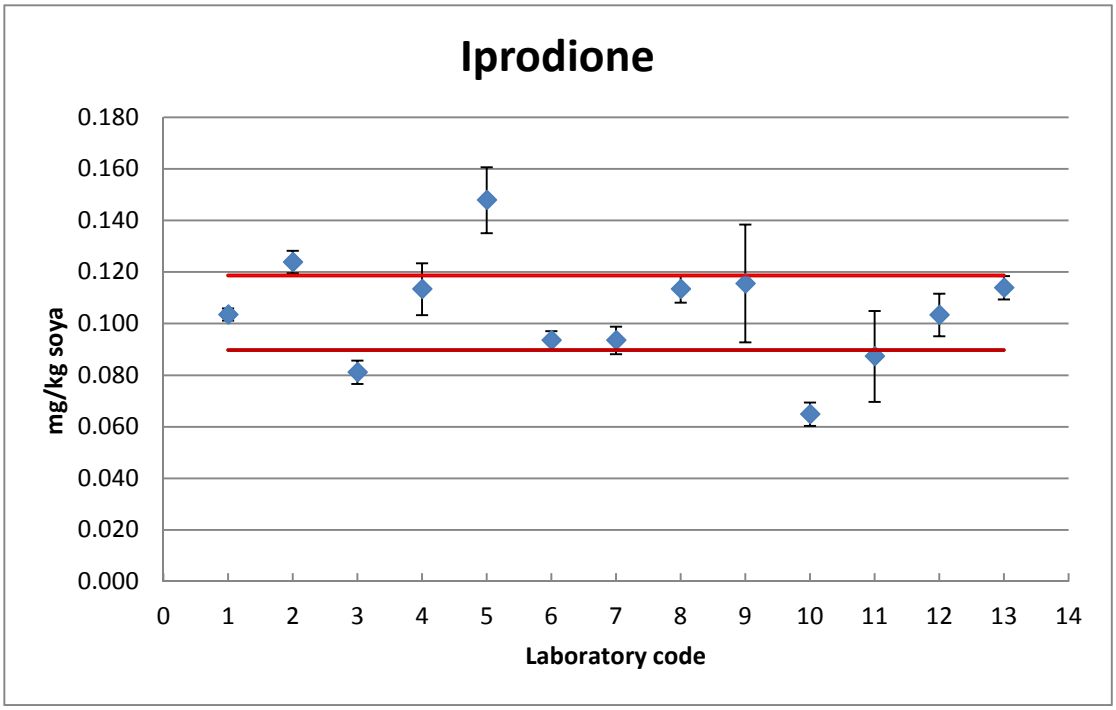


Figure F.9. **L**aboratory means with their standard deviations represented as error bars. Red lines correspond to the mean of laboratory means ± U_{CRM}

Table F.10. Mass fractions of methomyl in soya as reported by participant laboratories.

LABORATORY CODE	Replicate 1 [mg/kg]	Replicate 2 [mg/kg]	Replicate 3 [mg/kg]	Replicate 4 [mg/kg]	Replicate 5 [mg/kg]	Replicate 6 [mg/kg]	Mean [mg/kg]	S [mg/kg]
L02-LC-MS/MS	0.0507	0.0517	0.0475	0.0480	0.051	0.0479	**0.050**	**0.002**
L03-LC-MS/MS	0.036	0.037	0.037	0.038	0.037	0.038	**0.037**	**0.001**
L04-LC-MS/MS	0.043	0.043	0.041	0.039	0.039	0.039	**0.041**	**0.002**
L05-LC-MS/MS	0.061	0.062	0.051	0.055	0.073	0.064	**0.061**	**0.008**
L06-LC-MS/MS	0.046	0.050	0.047	0.052	0.051	0.049	**0.049**	**0.002**
L07-LC-MS/MS	0.044	0.047	0.043	0.049	0.051	0.050	**0.047**	**0.003**
L08-LC-MS/MS	0.041	0.044	0.042	0.039	0.041	0.041	**0.041**	**0.002**
L10-LC-MS/MS	0.048	0.052	0.049	0.046	0.048	0.046	**0.048**	**0.002**
L11-LC/MS/MS	0.0502	0.052	0.0465	0.0481	0.0504	0.0537	**0.050**	**0.003**
L12-LC-MS/MS	0.037	0.036	0.036	0.038	0.041	0.042	**0.038**	**0.003**

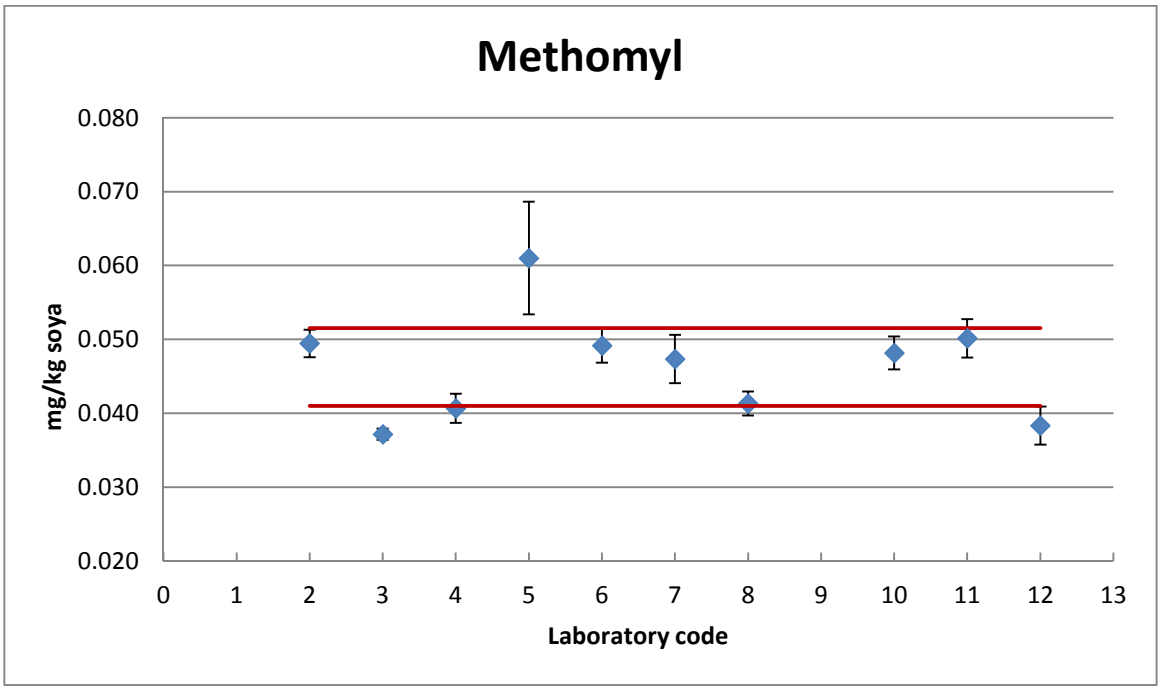


Figure F.10. **L**aboratory means with their standard deviations represented as error bars. Red lines correspond to the mean of laboratory means ± U_{CRM}

Table F.11. Mass fractions of tebuconazole in soya as reported by participant laboratories.

LABORATORY CODE	Replicate 1 [mg/kg]	Replicate 2 [mg/kg]	Replicate 3 [mg/kg]	Replicate 4 [mg/kg]	Replicate 5 [mg/kg]	Replicate 6 [mg/kg]	Mean [mg/kg]	S [mg/kg]
L01-LC-MS/MS	0.0497	0.0486	0.0481	0.0504	0.0508	0.0521	**0.050**	**0.002**
L02-LC-MS/MS	0.0474	0.0474	0.046	0.0438	0.0478	0.0468	**0.047**	**0.002**
L03-LC-MS/MS	0.034	0.033	0.033	0.034	0.032	0.034	**0.033**	**0.001**
L04-GC-MS/MS	0.046	0.047	0.042	0.047	0.045	0.049	**0.046**	**0.002**
L05-LC-MS/MS	0.048	0.054	0.055	0.06	0.072	0.069	**0.060**	**0.009**
L06-LC-MS/MS	0.045	0.049	0.048	0.046	0.047	0.045	**0.049**	**0.002**
L07-LC-MS/MS	0.041	0.045	0.045	0.047	0.052	0.048	**0.046**	**0.004**
L08-LC-MS/MS	0.053	0.052	0.051	0.056	0.053	0.052	**0.053**	**0.002**
L09-LC-MS/MS	0.033	0.033	0.040	0.038	0.035	0.035	**0.036**	**0.003**
L10-LC-MS/MS	0.046	0.045	0.045	0.042	0.042	0.044	**0.044**	**0.002**
L11-LC-MS/MS	0.0496	0.0513	0.0493	0.0553	0.0587	0.0570	**0.054**	**0.004**
L12-GC-MS/MS	0.049	0.056	0.059	0.063	0.067	0.063	**0.060**	**0.006**
L13-LC-MS/MS	0.045	0.046	0.043	0.043	0.041	0.044	**0.044**	**0.002**

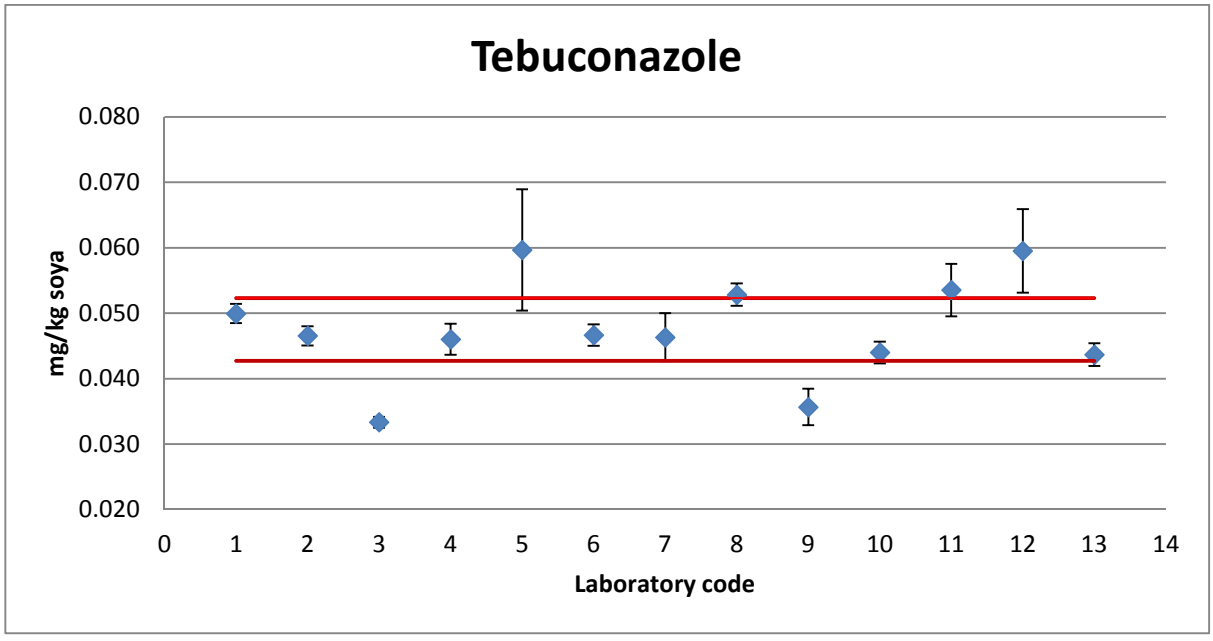


Figure F.11. **L**aboratory means with their standard deviations represented as error bars. Red lines correspond to the mean of laboratory means ± U_{CRM}

European Commission

EUR 29625 EN – Joint Research Centre – Directorate F – Health, Consumers and Reference Materials
Title: **CERTIFICATION REPORT: The certification of the mass fraction of pesticides in soya: ERM®-BC700**
Author(s): B. Sejerøe-Olsen, P. Shegunova, S. Harbeck, J. Seghers, A.R. Fernández-Alba, M. Dabrio
Luxembourg: Publications Office of the European Union
2019 – 62 pp. – 21.0 x 29.7 cm
EUR – Scientific and Technical Research series – ISSN 1831-9424
ISBN 978-92-79-98884-4
doi: 10.2760/064105

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KJ-NA- 29625-EN-N

Publications Office

doi: 10.2760/064105

ISBN: 978-92-79-98884-4