European Commission

Renewal Assessment Report of the Inclusion of the Active Substance in Annex I of the Regulation (EC) 1107/2009

Oxamyl

Volume 3 (CA)
ANNEX B.5 Methods of analysis

Rapporteur Member State: Italy
Co-Rapporteur Member State: France

December 2017

VERSION HISTORY

Date	Data points containing amendments or additions	Document identifier or version number
May 2016	First RAR of Italy	
December 2017	Revised RAR after comment CoRMS (France)	highlighted

TABLE OF CONTENTS

TABLE OF TABLES

B.5 METHODS OF ANALYSIS

Studies submitted to the EU for the first time in this submission is highlighted in yellow.

B.5.1 Methods used for the generation of pre-approval data

B.5.1.1 Methods for the analysis of the active substance as manufactured

B.5.1.1.1 Methods for the analysis of the active substance

Studies submitted to the EU for the first time in this submission.

B.5.1.1.1/01

Reference: B.5.1.1.1/01	**Report**	Pandey, S., McNally, M.E.P. (2015f); Determination of oxamyl (DPX-D1410) in technical grade oxamyl and end-use products **DuPont Report No.:** DuPont-42001 **GLP:** No

B.5.1.1.1/02

Reference: B.5.1.1.1/02	**Report**	Pandey, S. (2015b); Validation of the analytical method for determination of oxamyl (DPX-D1410) in technical grade oxamyl and oxamyl end-use products by reverse phase high performance liquid chromatography (RPLC) and ultra performance liquid chromatography (UPLC) **DuPont Report No.:** DuPont-36605 and DuPont-36605 Confidential attachment **GLP:** Yes

Description of the method

The method for assay of oxamyl as manufactured involves dissolution by ultrasonication of oxamyl in a solution of 5% methanol and 95% water adjusted to pH 3.0 with H_3PO_4. A known amount of acetanilide internal standard was added to each standard or sample. Samples were filtered before analysis. Analysis was done by reversed-phase liquid chromatography (HPLC) and ultra-high pressure liquid chromatography (UPLC), with quantitation by ultraviolet absorbance at 240 nm. The column used was a 1.8-µm particle size, 4.6 mm × 100 mm Agilent Zorbax-XDB C18 column for UPLC and 5.0 µm particle size, 4.6 mm × 150 mm Agilent Zorbax-Aqua C18 column for HPLC. The mobile phase was a gradient mixture of acetonitrile and water that had been adjusted to pH 3.0. The weight percent of oxamyl in each sample was determined by comparing peak area ratios of oxamyl/acetanilide with a calibration curve generated from the analysis of standard solutions.

Specificity, linearity, accuracy, repeatability

Information reported below concerns the method for the determination of the active substance in oxamyl as manufactured Oxamyl 42%, technical concentrate, TK and solid oxamyl as the pure standard, the Technical Grade Active Ingredient, TGAI

Specificity

The method was evaluated for interferences from expected manufacturing impurities, none of the known impurities exist above 0.5% in the technical active ingredient. Thus, none of the known impurities expected to be present in technical grade oxamyl co-elute with oxamyl or the internal standard, acetanilide at the 3% level. Since the EU requires that any interference present does not contribute more than ± 3% to the total quantity determined, this method satisfies the EU criteria for specificity.

Details regarding the specificity of the method are claimed as confidential and are included in the Oxamyl EU Renewal Dossier, Document J, Part 1, DuPont-40926 EU. As the disclosure of this information would also disclose the impurity profile of the technical product, we request them to be treated as confidential information according to Article 63 of Regulation EC 1107/2009, Council Directive 2003/4/EC, and similar regulations in other countries.

Linearity

The linearity of the method proposed for the determination of the active substance, oxamyl, as manufactured was demonstrated. Linearity has been demonstrated for both HPLC and UPLC, by the analyses of seven standard solutions within the nominal ranges,0.2–1.40 mg/mL for oxamyl. The resulting least square linear equations and correlation coefficients are as follows.

Table 1 Least square linear equations and correlation coefficients

System	Slope	y-intercept	Correlation coefficient (R^2)
HPLC	32.198	0.0042	0.9999
UPLC	32.004	0.0066	0.9999

Accuracy

For HPLC, the accuracy of this method for the analysis of oxamyl, Technical Grade Active Ingredient, TGAI, as manufactured samples was evaluated by analysing standard material as surrogate samples with duplicate determinations at three concentrations, in the range of 75–125% of the nominal sample active concentration. The same duplicate determinations at three concentrations in the range of 75–125% of the nominal sample concentration for the technical concentrate, TK, (Oxamyl 42%) were conducted. The average percent recovery obtained was 100.57% with a standard deviation of 0.8%, and 99.19% with a standard deviation of 1.1% for the TGAI and the TK, respectively. Therefore, the accuracy of this method is adequate for HPLC.

For UPLC, the accuracy of this method for the analysis of oxamyl, TGAI, as manufactured samples was evaluated by analysing standard material as surrogate samples with duplicate determinations at three concentrations, in the range of 75–125% of the nominal sample active concentration. The same duplicate determinations at three concentrations in the range of 75–125% of the nominal sample concentration for the technical concentrate, TK, (Oxamyl 42%) were conducted. The average percent recovery obtained was 100.55% with a standard deviation of 0.84% and 99.31% with a standard deviation of 1.08% for the TGAI and the TK, respectively. Therefore, the accuracy of this method is also adequate for UPLC.

Repeatability

Repeatability testing of the assay method was determined by calculating the standard deviation of the average percent oxamyl obtained from the analysis of eight replicate test portions of the same sample of oxamyl TGAI and TK as manufactured. The results were calculated for one analyst on one day.

For HPLC, the relative standard deviation was 0.43% and 0.55% for the oxamyl, TGAI and TK, as manufactured, respectively. The maximum allowable relative standard deviation calculated from the modified Horwitz equation was 1.34 % for the TGAI and 1.53 % for TK. Therefore, the HPLC method fulfils the EU repeatability criteria.

For UPLC, the relative standard deviation was 0.46% and 0.43% for the oxamyl, TGAI and TK, as manufactured, respectively. The maximum allowable relative standard deviation calculated from the modified Horwitz equation was also 1.34 % for the TGAI and 1.53 % for TK. Therefore, the UPLC method fulfils the EU repeatability criteria.

There were no outliers during this testing.

Applicability of existing CIPAC methods

There is a CIPAC method for oxamyl, it is FAO specification 342/TK (April, 2008). The analytical method for determination of oxamyl (including identity tests) is based on reversed-phase HPLC with UV detection at 240 nm and internal standardization with acetanilide. The method was adopted by CIPAC with provisional status in 2006 and full CIPAC method status in 2007. DuPont and CIPAC have noted that the method can also be used with external standardization. The method is applicable for technical concentrate, granules, and soluble concentrates.

RMS comments and conclusion

IT: the methods presented are considered acceptable

B.5.1.1.2 Methods for the determination of significant and/or relevant impurities in the active substance as manufactured

Analytical methods for the determination of quantitatively significant impurities (>1 g/kg) in oxamyl as manufactured are can be found in the Oxamyl EU Renewal Dossier Volume 4. As their disclosure would also disclose the impurity profile of the technical product, we request them to be treated as confidential information according to Regulation (EC) 1107/2009.

B.5.1.1.3 Methods for the determination of additives (e.g. stabilizers) in the active substance as manufactured

There are no additives considered of toxicological or environmental significance in oxamyl as manufactured that would justify submission of analytical methods.

B.5.1.2 Methods for risk assessment

B.5.1.2.1 Description of methods for determination of residues in soil used in support of environmental fate studies

Studies submitted to the EU for the first time in this submission.

B.5.1.2.1/01

Reference: B.5.1.2.1/01	**Report**	Nanita, S.C. (2009); Analytical method for the determination of oxamyl and its oxime metabolite in soil using LC/MS analysis **DuPont Report No.:** DuPont-2392, Revision No. 1 **GLP:** No

B.5.1.2.1/02

Reference: B.5.1.2.1/02	**Report**	Stry, J.J. (2000); Independent laboratory validation of method number DuPont-2392, "Analytical method for the determination of oxamyl and its oxime metabolite in soil using LC/MS analysis" **DuPont Report No.:** DuPont-3738 **GLP:** Yes

B.5.1.2.1/03

Reference: B.5.1.2.1/03	**Report**	Zietz, E. (2002); Field soil dissipation of oxamyl nematicide and insecticide applied as Vydate 10 L by means of drip irrigation to cucurbits in a greenhouse in Italy - Season 2000 **DuPont Report No**.: DuPont-4800 **GLP:** Yes

Description of the method

Oxamyl and its oxime metabolite are extracted from soil aliquots (approximately 13 g fresh weight) and into organic solvents using the Dionex Accelerated Solvent Extraction ASE® 200 or ASE. Aliquots of the soil extracts are concentrated and diluted for analysis by LC/MS. The instrumental method is liquid chromatographic gradient elution interfaced to a Mass Selective Detector (MSD) using Selected-Ion-Monitoring (SIM). Mode of analysis is Atmospheric Pressure Ionization-electrospray (API-ES) with positive ion mass spectrometric detection of ions having mass/charge (m/z) ratios of 237 (M+NH4+) for oxamyl and 163 (MH+) for its oxime metabolite.

Recovery findings

The fortification data reported in the method proposed for monitoring oxamyl and metabolite residues in soil are summarised in Table 2. Validation data are also presented based on additional method DuPont-4800. During the validation the extraction method remained unchanged, but the MS conditions were modified to include MS/MS detection. Validation was performed using soil from the study location. These data are summarized in Table 3. The daughter ion (m/z 71.70) of the ammonium (NH4+) adduct ion of oxamyl (m/z 237) and daughter ion (m/z 71.70) of the protonated IN-A2213 ion (m/z 163) were determined using electrospray ionization.

Table 2 Validation data for analytical methods for the determination of oxamyl and its metabolite IN-A2213 in soil

Reference	Matrix	Number of tests	Fortification level (mg/kg)[a,b]	Average recovery (%)	Standard deviation	% Relative standard deviation
Oxamyl						
DuPont-2392, Revision No. 1	Soil Edgecombe, NC, USA	11 3 3 Total = 17	0.01 0.03 0.10	90 90 95 Mean = 92	6 2 8 Mean = 5.3	7 2 10 Mean = 6.3
IN-A2213						
DuPont-2392, Revision No. 1	Soil Edgecombe, NC, USA	11 3 3 Total = 17	0.01 0.03 0.10	96 90 93 Mean = 93	7 2 9 Mean = 6	7 3 11 Mean = 7

[a] Fortifications were performed with analyte reference standard solutions
[b] Limit of quantification, defined by a signal-to-noise ratio of approximately 10 and the lowest validated fortification level

Table 3 Validation data for analytical methods for the determination of oxamyl and its metabolite IN-A2213 in soil

Reference	Matrix	Number of tests	Fortification level (mg/kg)[a,b]	Average recovery (%)	Standard deviation	% Relative standard deviation
Oxamyl (237 → 71.70)						
DuPont-4800	Soil	8 4 4 Total = 16	0.005 0.10 2.30	95 90 94 Mean = 93	4.2 6.3 3.3 Mean = 4.6	4.4 3.3 3.5 Mean = 3.7
IN-A2213 (163 → 71.70)						
DuPont-4800	Soil	8 4 4 Total = 16	0.005 0.10 2.30	94 90 92 Mean = 92	1.4 5.6 2.6 Mean = 3.2	1.5 6.2 2.9 Mean = 3.5

[a] Fortifications were performed with analyte reference standard solutions
[b] Limit of quantification, defined by a signal-to-noise ratio of approximately 10 and the lowest validated fortification level

Linearity

Good linearity was observed in the range 0.125 to 20.0 ng/mL for oxamyl and its metabolite, IN-A2213. All calculations were performed using standards prepared in solvent.

Specificity

The limit of detection of the method proposed for monitoring oxamyl and its metabolite, IN-A2213, residues in soil is 0.001 mg/kg. Unfortified control soil samples were carried through the same sample extraction and analysis procedures to establish the background response in the vicinity of each analyte. No significant interference peaks in the vicinity of either analyte were detected in these control matrix samples.

Limit of quantification

The limit of quantification of the method for residues of oxamyl and its metabolite, IN-A2213, in soil is 0.010 mg/kg for DuPont-2392, Revision No. 1 and DuPont-3738.

The limit of quantification of the method for residues of oxamyl and its metabolite, IN-A2213, in soil is 0.005 mg/kg based on most recent study submitted.

Repeatability

Repeatability of the method is addressed by the data in Table 4, which were obtained over multiple days. Relative standard deviations of less than 20% were obtained for fortifications made at the quantification limit for each matrix, as well as at higher levels. Therefore, the repeatability of this method would seem to be adequate for the purposes of detecting oxamyl and metabolite residues in soil.

Reproducibility

An independent laboratory validation (DuPont-3738) was conducted and the results are summarized in Table 4.

Table 4 Validation data for analytical methods for the determination of oxamyl and its metabolite IN-A2213 in soil

Reference	Matrix	Number of tests	Fortification level (mg/kg)[a,b]	Average recovery (%)	Standard deviation	% Relative standard deviation
Oxamyl						
DuPont-3738	Soil	5 5 Total = 10	0.01 0.10	91 105 Mean = 98	6 5 Mean = 6	7 5 Mean = 6
IN-A2213						
DuPont-3738	Soil	5 5 Total = 10	0.01 0.10	100 89 Mean = 95	2 4 Mean = 3	2 5 Mean = 4

[a] Fortifications were performed with analyte reference standard solutions
[b] Limit of quantification, defined by a signal-to-noise ratio of approximately 10 and the lowest validated fortification level

Extraction efficiency

Extraction efficiency for the solvents used during the ASE extraction was verified for oxamyl and IN-A2213 during the course of freezer storage stability studies conducted as part of DuPont-2815,. Samples were fortified with oxamyl and IN-A2213 (separately) at a level of 0.1 mg/kg and stored at -20°C. The recovery results obtained are shown in Table 5. It is apparent that no loss of extraction efficiency was observed for oxamyl or IN-A2213, which shows that the extraction procedure was capable of releasing aged residues from soil.

Table 5 Recovery data from the storage stability study of oxamyl and IN-A2213 in Netherlands soil

		0 days	21 days	7 months
Oxamyl	Control	ND[a]	ND	ND
	Fresh Fortification	104	114	108
	Aged Fortification 1	-	98	96
	Aged Fortification 2	-	99	104
IN-A2213	Control	ND	ND	ND
	Fresh Fortification	101	109	91
	Aged Fortification 1	-	88	82
	Aged Fortification 2	-	92	81

[a] N.D = not detected

Confirmatory method

The method used to generate these data is not proposed as an environmental monitoring method. A confirmatory procedure is not required for data collection methods.

Overall suitability for data collection

This method is suitable to generate data used for environmental risk assessment. The instrumentation required to perform both the analysis is available in most well equipped analytical laboratories.

RMS comments and conclusion

B.5.1.2.2 Description of methods for determination of residues in water used in support of environmental fate studies

The methods used for analysing water samples in support of environmental fate studies were performed in conjunction with soil methods and are summarized in this document. Additionally, the proposed monitoring method summarized in Point B.5.2.4 could be used for the determination of oxamyl residues in water.

B.5.1.2.3 Description of methods for determination of residues in air used in support of environmental fate studies

The environmental fate studies did not require analysis of air samples. The proposed environmental monitoring method summarised in Point B.5.2.5 in this document could be used to support the environmental fate studies.

B.5.1.2.4 Description of methods for determination of residues in soil used in support of efficacy studies

The efficacy trials conducted did not require the analysis of soil samples. Therefore a method is not submitted. The proposed environmental monitoring method is summarised in Point B.5.2.3 in this document.

B.5.1.2.5 Description of methods for determination of residues in water used in support of efficacy studies

The efficacy trials conducted did not require the analysis of water samples. Therefore a method is not submitted. The proposed environmental monitoring method is summarised in Point B.5.2.4 in this document.

B.5.1.2.6 Methods for the analysis of products of animal origin and feed used in support of toxicological studies

Toxicology studies did not require a method to analyse residues of oxamyl in products of animal origin or feed. However, the enforcement method proposed in Point B.5.2.2 of this document may be used for this purpose.

B.5.1.2.7 Methods for the analysis of body fluids used to support in support of toxicological studies

Toxicology studies did not require a method to analyse residues of oxamyl in body fluids. The enforcement method proposed in Point B.5.2.6 of this document may be used for this purpose.

B.5.1.2.8 Description of methods for determination of residues in air used to support in support of toxicological studies

Toxicology studies did not require a method to analyse residues of oxamyl in air. The enforcement method proposed in Point B.5.2.5 of this document may be used for this purpose.

B.5.1.2.9 Methods for the analysis of additional matrices used in support of operator, worker, resident and bystander exposure studies

The analytical method used for the analysis of oxamyl in a worker exposure study (DuPont-2311) has been previously reviewed within the toxicological section and has been determined to be acceptable. Therefore, it does not need to be included in this section.

B.5.1.2.10 Methods for the analysis of body fluids used to support operator, worker, resident and bystander exposure studies

The proposed monitoring method summarized in Point B.5.2.6 for the analysis of body fluids can be used in support of the of operator, worker, resident and bystander exposure studies.

B.5.1.2.11 Description of methods for determination of residues in air used in support of operator, worker, resident and bystander exposure studies

The analytical method used for the analysis of oxamyl in a worker exposure study (DuPont-2311) has been previously reviewed within the toxicological section and has been determined to be acceptable. Therefore, it does not need to be included in this section.

B.5.1.2.12 Methods for the analysis of additional matrices used in support of operator, worker, resident and bystander exposure studies

No additional analytical methods were developed to collect operator, worker, resident, or bystander exposure data.

B.5.1.2.13 Methods for the analysis of plants, plant products and processed food commodities to support residue trials

Study submitted in the EU Dossier in 2001 and included in the first EU approval review.

B.5.1.2.13/01

Reference: --	**Report**	Françon, B., Jetzer, M., Matthey, C. (2000); Method validation for the determination of oxamyl and its oxime metabolite in different crops **DuPont Report No.:** DuPont-3702 **GLP:** Yes

Principle of the method:

The sample (3g) is mixed with "Hydromatrix"(3g) and silica gel (3g) and the homogenate is placed in an "Accelerated Solvent Extraction"(ASE) where the sample was extracted using acetone. The acetone extract was then cleaned up further using an SPE "ENVITM-Carb cartridge" (0.5g) and a "Silica Mega Bond Elut (1g). The cleaned up extract is evaporated to low volume (~0.5ml), made up to volume (2ml) with water /acetonitrile (92:8) and is then analysed by HPLC using column switching and a uv detector at 245nm. The HPLC columns used in the course of the analysis were a Zorbax SB-phenyl [5-□m particle size, 4.6 mm id and 150mm long] and a Discovery, RP Amide C16 [5-µm particle size, 4.6 mm id and 150 mm long]. The mobile phase was a gradient phase of water:acetonitrile [92:8 Oxamyl, 98:2 Oxime].

Validation data: Linearity: The linearity of the system was assessed for Oxamyl using a 4 point calibration curve in the range 0.02 to 0.5 mg/l. The correlation co-efficient was determined to be 0.99968.

The linearity of the system was assessed for Oxamyl oxime using a 4 point calibration curve in the range 0.02 to 0.5 mg/l. The correlation co-efficient was determined to be 0.998578.

Recovery studies were carried out using lettuce, potato, melon and sugar beet. The results of the recovery study are presented below in Table 6.

Table 6 Recovery studies for Oxamyl and Oxamyl oxime in a range of crops

Crop matrix	Fortification level (mg/kg)	Number of replicates	Oxamyl		Oxamyl Oxime	
			Mean recovery %	RSD %	Mean recovery %	RSD %
Lettuce	0.02	8	90	13.3%	96	11.5%

leaf.	**0.2**	**5**	**74**	**10.8%**	**79**	**7.6%**
Potato tuber	**0.02**	**5**	**82**	**20.7%**	**Not validated**	**Not validated**
	0.2	**8**	**92**	**10.9%**	**Not validated**	**Not validated**
Melon pulp.	**0.02**	**8**	**79**	**12.7%**	**84**	**16.7%**
	0.2	**5**	**80**	**5%**	**76**	**11.8%**
Melon peel.	**0.02**	**7**	**86**	**17.4%**	**98**	**8.2%**
	0.2	**7**	**80**	**16.3%**	**75**	**6.7%**
Sugar beet root.	**0.02**	**5**	**86**	**18.6%**	**93**	**11.8%**
	0.2	**5**	**77**	**19.5%**	**76**	**15.8%**
Sugar beet top+ leaves.	**0.02**	**5**	**80**	**15%**	**Not validated**	**Not validated**
	0.2	**5**	**89**	**6.7%**	**Not validated**	**Not validated**
Note: "Not validated" indicates that it was not possible to quantify the spiked residue due to matrix interference.						

Sample chromatograms were presented for each of the crops studied and for the fortification levels studied.

Conclusion:

The method of analysis has been validated for a range of crops for the analysis of oxamyl and its oxime. Due to the interferences present in potato tubers and sugar beet leaves/tops which prevent the analysis of the spiked oxamyl oxime it is clear that the method is not applicable across a wide range of crops and will require further validation data if it is used in the laboratory. It is also considered that whereas the method is fully validated for the determination of Oxamyl in the range of crops studied that the extension of the method for the analysis of other crops would also require additional validation data. This comment is made due to the poor confirmatory nature of a single wavelength UV detector. This is confirmed in section B.7.6.4.1. where difficulties were encountered with the analysis of orange peel using this method due to matrix interferences which required the use of an alternative analytical method, B.5.2.1.4..

The monitoring method for determination of residues in plants, plant products and processed food commodities, DuPont-3702, originally submitted under EU Rev8 Point IIA 4.3.1.2, was conducted under guidelines Directive 91/414/EEC and SANCO 8064/VI/97-rev 4 (1998). A review of this study indicates that it partially meets the current guideline SANCO/825/00 rev 8.1; deviations include lack of confirmatory data and fortifications that do not meet the required LOQ. However, due to the precision and recovery statistics for this method, reconduct is unlikely to yield a significantly different result. This method is relied upon for this submission.

RMS comments and conclusion: conclusions presented are considered acceptable

Studies submitted to the EU for the first time in this submission.

B.5.1.2.13/01

Reference: CA 4.1.2/15	**Report**	Doran, A.M., Cairns, S.D., McGuire, G.M., Vance, C.J. (2003); Validation of an analytical method for the determination of oxamyl in potatoes using LC-MS **DuPont Report No.:** DuPont-11125 **GLP:** Yes

B.5.1.2.13/02

Reference: CA 4.1.2/14	**Report**	Chapleo, S., Johnson, J. (2014); The metabolism of ^{14}C-oxamyl in tomato plants **DuPont Report No.:** DuPont-32188 **GLP:** Yes

Description of the method

Oxamyl was extracted from potato samples using a mixture of organic solvents followed by solid phase extraction clean-up using an aminopropyl cartridge. Following extraction and centrifugation, a 2 mL aliquot was blown to dryness under a stream of oxygen free nitrogen. The sample was then reconstituted in organic solvent and passed through an aminopropyl SPE tube. The sample was once again blown to dryness and reconstituted in mobile phase. Oxamyl residues were detected and quantified by HPLC by electrospray mass spectrometry detection (LC-MS) in positive mode (ESI +). 237.3 m/z was used as ion mass for determination.

Method DuPont 11125 - The samples were extracted using acetone (30 mL), dichloromethane (30mL) and petroleum ether (30mL), the extract was purified on Bond Elut cartridge, it was eluted with dichloromethane. The extract was evaporated to dryness and resuspend in acetonitrile/water (65:35, v/v) before to inject.

Method DuPont-32188 - the samples were extracted by methanol and methanol/water (1:1, v:v).

Recovery findings

The fortification data reported in the DuPont-11125 are summarised in Table 7. The results listed below were obtained using standards prepared in solvent. The average recovery specified in the decision-making criteria is 70–120%, with a standard deviation of ≤20%. Therefore, the recovery of this method is adequate for the purposes of residue data collection and enforcement of MRLs.

Table 7 Validation data for analytical methods for the determination of oxamyl in food of plant origin

Matrix	Fortification level (mg/kg)[a,b]	Number of tests	Average recovery (%)	Standard deviation	% Relative standard deviation	Reference
Potatoes	0.01 0.10	5 5 Total = 10	86 90 Mean = 91	2.3 2.5 Mean = 2.4	2.7 2.8 Mean = 2.8	DuPont-11125

[a] Fortifications were performed with analyte reference standard solutions

[b] Limit of quantification, defined by a signal-to-noise ratio of approximately 10 and the lowest validated fortification level

Linearity

Good linearity was observed in the range of 0.20 to 50 ng/mL for oxamyl, (8 solutions, 1 time injected).

Specificity

The limit of determination for oxamyl in DuPont-11125 was 0.007 mg/kg for the potato samples tested. Analysis of control samples resulted in no detectable apparent residues of oxamyl, and the response in the area of the oxamyl peak always corresponded to less than 20% of the limit of determination. It can therefore be concluded that few, if any, apparent residues or false positive values would arise.

Limit of quantification

The limit of quantification of the method proposed for monitoring oxamyl residues is 0.010 mg/kg for the potato samples tested.

Repeatability

Repeatability of the method is addressed by the data in Table 7. Relative standard deviations of less than 20% were obtained for fortifications made at the quantification limit, as well as at higher levels. Therefore, the repeatability of this method is adequate for the purposes of residue data collection and enforcement of MRLs.

Reproducibility

The method used to generate these data is not proposed as a monitoring method for food and feed of plant origin. Independent laboratory validations are not required for data collection methods.

Extraction efficiency

Extraction efficiency of this method was investigated in DuPont-32188 by extracting radiolabelled residue samples of tomatoes. The residue profiles obtained were compared with those obtained for the same samples in the plant metabolism study. The results of this study showed that the residue method is able to extract most of the residue present in the crop. On average, the residue method extraction used in DuPont-11125 extracted 67% of the radiolabelled oxamyl that was extracted using the metabolism method.

Confirmatory method

The method used to generate these data is not proposed as a monitoring method for food and feed of plant origin. A confirmatory procedure is not required for data collection methods.

Overall suitability for data collection

This method is suitable for residue data collection of oxamyl in potatoes. The instrumentation required to perform the analysis method is available in most well equipped analytical laboratories. All of the sample preparation equipment is commercially available.

Conclusions

The residue method for the determination of oxamyl residues in potatoes involves simple extraction, clean-up, and analytical determination by HPLC/MS detection. A limit of quantification of 0.010 mg/kg can be achieved consistently. This method is suitable for the collection of oxamyl residue data in potatoes.

Studies submitted to the EU for the first time in this submission.

B.5.1.2.13/03

Reference: CA 4.1.2/13	**Report**	Cairns, S.D., Davidson, J. (2006); Validation of an analytical method for the determination of oxamyl in green, dried and fermented tobacco leaves **DuPont Report No.:** DuPont-17601 **GLP:** Yes

Description of the method

Residues of oxamyl were extracted from green, dried, and fermented tobacco leaves using acetonitrile. Lipids were removed using hexane, and the samples were then cleaned up by solid phase extraction using an aminopropyl (NH_2) cartridge. Oxamyl residues were detected and quantified by liquid chromatography with mass spectrometry detection (LC-MS) in positive mode (ESI +). 237.3 m/z was used as ion mass for determination. The method was validated with respect to system suitability, system linearity, assay accuracy, assay precision, and assay specificity.

Recovery findings

The fortification data reported in the method for the determination of oxamyl residues in green, dried, and fermented tobacco samples are summarised in Table 8. The results listed below were obtained using standards prepared in solvent. The average recovery specified in the decision-making criteria is 70–120%, with a standard

deviation of ≤20%. Therefore, the recovery of this method is adequate for the purposes of residue data collection.

Table 8 Validation data for analytical methods for the determination of oxamyl in green, dried, and fermented tobacco leaves

Matrix	Fortification level (mg/kg)[a,b]	Number of tests	Average recovery (%)	Standard deviation	% Relative standard deviation	Reference
Green Tobacco Leaves	0.01 0.10	5 5 Total = 10	96 93 Mean = 95	4 6 Mean = 5	4 6 Mean = 5	DuPont-17601
Dried Tobacco Leaves	0.01 0.10	5[c] 5 Total = 10	100 97 Mean = 99	6 5 Mean = 6	6 5 Mean = 6	
Fermented Tobacco Leaves	0.01 0.10	5 5 Total = 10	92 82 Mean = 87	6 1 Mean = 4	7 1 Mean = 4	

[a] Fortifications were performed with analyte reference standard solutions

[b] Limit of quantification, defined by a signal-to-noise ratio of approximately 10 and the lowest validated fortification level

[c] One sample omitted as an outlier. Exclusion of one replicate from five is permissible according to SANCO/3029/99 rev. 4 when a clear outlier is identified.

Linearity

Good linearity was observed in the range of 0.20 to 50 ng/mL for oxamyl (8 solutions, 1 time injected).

Specificity

The limit of quantification of the method for the determination of oxamyl residues in green, dried, and fermented tobacco leaves is 0.010 mg/kg for the samples tested. The analysis of fortified control extracts demonstrated that there was no significant matrix enhancement or suppression. The mean percentage deviation from an equivalent solvent standard was found to be less than ±15% for green, dried, and fermented tobacco leaves. It can therefore be concluded that few, if any, apparent residues or false positive values would arise.

Limit of quantification

The limit of quantification of the method for the determination of oxamyl residues in green, dried, and fermented tobacco leaves is 0.010 mg/kg for the samples tested.

Repeatability

Repeatability of the method is addressed by the data in Table 8. Relative standard deviations of less than 20% were obtained for fortifications made at the quantification limit for each matrix, as well as at higher levels. Therefore, the repeatability of this method is adequate for the purposes of residue data collection.

Reproducibility

The method used to generate these data is not proposed as an environmental monitoring method. Independent laboratory validations are not required for data collection methods.

Extraction efficiency

The extraction efficiency of this method was investigated in DuPont-44316, summarized under Point 5.2.1 of this document, by extracting tomato samples with incurred, radiolabelled residues. Tomato foliage and fruit were extracted using the method found in DuPont-17601 and compared to the metabolism method. On average, DuPont-17601 extracted 111% of the oxamyl residue found in tomato foliage and 123% of the oxamyl residue found in tomato fruit. This data indicates that the method provides acceptable extraction efficiency for the analysis of oxamyl residue.

Confirmatory method

The method used to generate these data is not proposed as an environmental monitoring method. A confirmatory procedure is not required for data collection methods.

Overall suitability for Data Collection

This method is suitable for residue data collection of oxamyl in dried, green, and fermented tobacco leaves. The instrumentation required to perform the analysis method is available in most well equipped analytical laboratories. All of the sample preparation equipment is commercially available.

Conclusions

The residue method for the determination of oxamyl residues in dried, green, and fermented tobacco involves simple extraction, clean-up, and analytical determination by HPLC/MS detection. A limit of quantification of 0.010 mg/kg can be achieved consistently. This method is suitable for the collection of oxamyl residue data in dried, green and fermented tobacco leaves.

Studies submitted to the EU for the first time in this submission.

B.5.1.2.13/04

Reference: CA 4.1.2/12	**Report**	Cairns, S. (2012); Method validation for the analysis of oxamyl (DPX-D1410) in representative crop commodities using LC-MS/MS **DuPont Report No.:** DuPont-33191 **GLP:** Yes

B.5.1.2.13/05

Reference: CA 4.1.2/14	**Report**	Chapleo, S., Johnson, J. (2014); The metabolism of ^{14}C-oxamyl in tomato plants **DuPont Report No.:** DuPont-32188 **GLP:** Yes

Description of the method

Oxamyl was extracted from dry, watery, acidic, and oily crops by shaking with a mixture of organic solvents, followed by sample clean-up by solid-phase extraction (SPE) using an aminopropyl cartridge. The final extracts were quantified for oxamyl residues by liquid chromatography with tandem mass spectrometry employing electrospray ionization in the positive ion mode. Two ion transitions were monitored for oxamyl, one serving as the quantitative ion transition and the other as the confirmatory transition.

Recovery findings

The fortification data reported in the method proposed for monitoring oxamyl residues in crop samples are summarised in Table 9. The results listed below were obtained using standards prepared in solvent. The average recovery specified in the decision-making criteria is 70–120%, with a standard deviation of ≤20%. Therefore, the recovery of this method is adequate for the purposes of residue data collection and enforcement of MRLs.

Table 9 Validation data for analytical methods for the determination of oxamyl in food of plant origin

Matrix	Fortification level (mg/kg)[a,b]	Number of tests	Average recovery (%)	Standard deviation	% Relative standard deviation	Reference
Oxamyl (237 → 72)						
Wheat Grain	0.01 0.10	5[c] 5 Total = 10	82 106 Mean = 94	8 10 Mean = 9	10 10 Mean = 10	DuPont-33191
Linseed	0.01 0.10	5 5 Total = 10	100 100 Mean = 100	10 4 Mean = 7	10 4 Mean = 7	
Cucumber	0.01 0.10	5 5 Total = 10	83 82 Mean = 83	6 3 Mean = 5	8 3 Mean = 6	
Orange	0.005 0.10	5 5 Total = 10	83 82 Mean = 83	4 3 Mean = 4	5 4 Mean = 5	

[a] Fortifications were performed with analyte reference standard solutions

[b] Limit of quantification, defined by a signal-to-noise ratio of approximately 10 and the lowest validated fortification level

[c] One sample omitted as an outlier. Exclusion of one replicate from five is permissible according to SANCO/3029/99 rev. 4 when a clear outlier is identified.

Linearity

Good linearity was observed in both the quantitative and confirmatory ion transitions for oxamyl over the range of 0.050 to 5.0 ng/mL. The standards used for quantifying oxamyl residue were prepared in solvent.

Specificity

The limit of quantification of the method for the determination of oxamyl residues in the dry, water, and oily crop samples tested is 0.010 mg/kg. The limit of quantification of the method for the determination of oxamyl residues in the acidic crop samples is 0.005 mg/kg. The analysis of wheat grain, linseed, cucumber, and orange control samples was shown to be specific. There were no significant interfering substances exceeding 30% of the LOQ at the retention time of oxamyl. Simultaneous method confirmation was conducted by acquiring a second MS/MS transition; this confirmed that quantification by the primary (quantitative) transition was selective and not effected by any other compound.

Limit of quantification

The method was validated at concentrations in the range of 0.010 to 0.10 mg/kg for oxamyl in wheat grain, linseed, and cucumbers, and the range of 0.005 to 0.10 mg/kg for oxamyl in oranges.

Repeatability

Repeatability of the method is addressed by the data in Table 9. Relative standard deviations of less than 20% were obtained for fortifications made at the quantification limit for each matrix, as well as at higher levels. Therefore, the repeatability of this method is adequate for the purposes of residue data collection.

Reproducibility

The method used to generate these data is not proposed as a monitoring method for food and feed of plant origin. Independent laboratory validations are not required for data collection methods.

Extraction efficiency

Extraction efficiency of this method was investigated in DuPont-32188 by extracting radiolabelled residue samples of tomatoes. The residue profiles obtained were compared with those obtained for the same samples in the plant metabolism study. The results of this study showed that the residue method is able to extract most of the residue present in the crop. On average, the residue method extraction used in DuPont-33191 extracted 67% of the radiolabelled oxamyl that was extracted using the metabolism method.

Confirmatory method

Confirmation of results was obtained using secondary LC/MS/MS ion transitions collected at the same time as the quantitative transitions. The recovery data obtained using the confirmatory procedure are summarised in Table 10.

Table 10 Validation data for analytical methods for the determination of oxamyl in food of plant origin

Matrix	Fortification level (mg/kg)[a,b]	Number of tests	Average recovery (%)	Standard deviation	% Relative standard deviation	Reference
Oxamyl (237 → 90)						
Wheat Grain	0.01 0.10	5[c] 5 Total = 10	95 106 Mean = 101	7 10 Mean = 9	8 9 Mean = 9	DuPont-33191
Linseed	0.01 0.10	5 5 Total = 10	101 100 Mean = 101	11 4 Mean = 8	11 4 Mean = 8	
Cucumber	0.01 0.10	5 5 Total = 10	75 81 Mean = 78	9 4 Mean = 8	12 5 Mean = 9	
Orange	0.005 0.10	5 5 Total = 10	83 82 Mean = 83	9 3 Mean = 6	11 4 Mean = 8	

[a] Fortifications were performed with analyte reference standard solutions

[b] Limit of quantification, defined by a signal-to-noise ratio of approximately 10 and the lowest validated fortification level

[c] One sample omitted as an outlier. Exclusion of one replicate from five is permissible according to SANCO/3029/99 rev. 4 when a clear outlier is identified.

Overall suitability for Data Collection

This method is suitable for residue data collection of oxamyl in the four EU crop groups (dry, watery, acidic, and oily). The instrumentation required to perform the analysis method is available in most well equipped analytical laboratories. All of the sample preparation equipment is commercially available.

Conclusions

The residue method for the determination of oxamyl residues in dry, watery, acidic, and oily crop samples involves a simple extraction, clean-up, and analytical determination by HPLC/MS/MS detection. A limit of quantification of 0.010 mg/kg can be achieved consistently for the dry, watery, and oily crops and a limit of quantification of 0.005 mg/kg can be achieved consistently for acidic crops. This method is suitable for the collection of oxamyl residue data in the four EU crop groups.

RMS comments and conclusion: the methods presented are considered acceptable.

B.5.1.2.14 Methods for the analysis of products of animal origin and feed used to support an animal feeding study

The proposed environmental monitoring method summarised in Point B.5.2.2 in this document can be used to analyse oxamyl in products of animal origin and feed. Studies requiring the analysis of oxamyl did not require a data collection method, and therefore, a data collection method is not included in this submission.

B.5.1.2.15 Description of methods for determination of residues in soil used in support of ecotoxicology studies

Studies submitted to the EU for the first time in this submission.

B.5.1.2.15/01

Reference: CA 4.1.2/31	**Report**	Schwarz, A., Eichler, M. (2014); Oxamyl (DPX-D1410) 10SL: Field study on residues in arthropods, earthworms and seedlings (wildlife food items) **DuPont Report No.:** DuPont-40221 **GLP:** Yes

B.5.1.2.15/02

Reference: CA 4.1.2/12	**Report**	Cairns, S. (2012); Method validation for the analysis of oxamyl (DPX-D1410) in representative crop commodities using LC-MS/MS **DuPont Report No.:** DuPont-33191 **GLP:** Yes

B.5.1.2.15/03

Reference: CA 4.1.2/14	**Report**	Chapleo, S., Johnson, J. (2014); The metabolism of ^{14}C-oxamyl in tomato plants **DuPont Report No.:** DuPont-32188 **GLP:** Yes

Description of the method

Analysis of arthropods and earthworms

Oxamyl was extracted from arthropod and earthworm samples using acetonitrile and a QuECHERS style cleanup. After partitioning, an aliquot of the supernatant was transferred into a 15 mL SampliQ EN dispersive SPE tube and vortexed. For arthropods, 1 mL of this extract was taken and evaporated to dryness under a gentle stream of nitrogen at room temperature. The residue was taken up in an acetonitrile/water mixture (50/50 v/v) containing 0.1% formic acid, vortexed, sonicated, and filtered for LC-MS/MS analysis. For worms, the extract was diluted and filtered prior to analysis.

Analysis of weeds

Oxamyl was extracted from weed samples using the extraction procedure outlined in DuPont-33191, previously summarised in Point B 5.1.2.13 in this document, with a modified cleanup. An aliquot of the extract was pipetted into a 15 mL tube and evaporated to dryness under a gentle stream of nitrogen at room temperature. The residue was taken up in 2 mL dichloromethane and passed through a NH2-Bond Elut SPE cartridge and then evaporated to dryness at room temperature. The residue was taken up in an acetonitrile/water mixture (50/50 v/v) containing 0.1% formic acid, diluted with the same acetonitrile/water mixture, and transferred into an autosampler vial for LC-MS/MS analysis.

Recovery findings

The fortification data reported in the method used to analyse oxamyl residues in arthropods, earthworms, and weeds are summarised in Table 11.

Table 11 Validation data for analytical methods for the determination of oxamyl in arthropods, earthworms, and weeds

Reference	Matrix	Number of tests	Fortification level (mg/kg)[a,b]	Average recovery (%)	Standard deviation	% Relative standard deviation

Oxamyl (237 → 90)						
DuPont-40221	Arthropods	5 5 Total = 10	0.01 0.10	99 82 Mean = 91	7 3 Mean = 5	7 3 Mean = 5
	Earthworms	5 5 2 Total = 12	0.01 0.10 10.0	92 94 96 Mean = 94	3 2 1 Mean = 2	3 2 1 Mean = 2
	Weeds	5 5 2 Total = 12	0.01 0.10 10.0	82 80 91 Mean = 83	15 6 3 Mean = 8	18 8 3 Mean = 11

[a] Fortifications were performed with analyte reference standard solutions
[b] Limit of quantification, defined by a signal-to-noise ratio of approximately 10 and the lowest validated fortification level

Linearity

Arthropods

Good linearity was observed in the range of 0.1 to 5.0 ng/mL for oxamyl. All calculations were performed using standards prepared in solvent.

Earthworms

Good linearity was observed in the range of 0.05 to 5.0 ng/mL for oxamyl. All calculations were performed using standards prepared in solvent.

Weeds

Good linearity was observed in the range of 0.05 to 5.0 ng/mL for oxamyl. All calculations were performed using standards prepared in solvent.

Specificity

The limit of determination of the method used to monitor oxamyl residues in arthropods, earthworms, and weeds is 0.01 mg/kg. Analysis of control samples from each sample type resulted in no detectable apparent residues of oxamyl; the response in the area of the oxamyl peak always corresponded to less than 20% of the limit of determination. It can therefore be concluded that few, if any, apparent residues or blank values would arise.

Limit of quantification

The limit of quantification of the method for oxamyl residues in all matrices is 0.01 mg/kg.

Repeatability

Repeatability of the method is addressed by the data in Table 11. Relative standard deviations of less than 20% were obtained for fortifications made at the quantification limit for each matrix, as well as at higher levels. Therefore, the repeatability of this method would seem to be adequate for the purposes of detecting oxamyl residues in arthropods, earthworms, and weeds.

Reproducibility

An independent laboratory validation of the analytical method contained in DuPont-40221 was not conducted. This method is proposed as a data collection method.

Extraction efficiency

Extraction efficiency of the method used to analyse arthropods and earthworm samples was not demonstrated for these matrices; however, it was evaluated through the analysis of tomato and foliage extracts during DuPont-44316 and summarized in Point CA 4.2.1 in this document.

Extraction efficiency of the method used to analyse weeds samples was evaluated by analysis of incurred residue samples during DuPont-32188 and summarized in Point CA 4.1.2.13.

Confirmatory method

Confirmatory methods are not required for data collection methods.

Overall suitability for regulatory purposes

This method is suitable for the collection of residue data in arthropods, earthworms, and weeds. The instrumentation required to perform the analysis methods is available in most well equipped analytical laboratories and all of the sample preparation equipment is commercially available. The method does not require the use of untreated commodity to correct for recoveries.

Study submitted to the EU for the first time in this submission.

B.5.1.2.15/04

CA 4.1.2/25	**Report**	Meinerling, M. (2014); Oxamyl (DPX-D1410) technical (98% w/w): Accumulation and elimination in earthworms (*Eisenia fetida*) in artificial soil **DuPont Report No.:** DuPont-38477 **GLP:** Yes

B.5.1.2.15/05

CA 4.1.2/24	**Report**	McClory, J.P. (2004); Analytical method for the determination of oxamyl and IN-A2213 metabolite in soil using LC/MS/MS **DuPont Report No.:** DuPont-7191, Revision No. 1 **GLP:** No

Description of the method

Analysis of Soil Samples

DuPont-38477 utilizes the extraction found in DuPont-7191, Revision No. 1 to determine residues of oxamyl and IN-A2213 in soil samples. During In DuPont-7191, Revision No. 1 soil samples were extracted with a preheated (50°C) solution of formic acid in methanol/acetonitrile, through mechanical shaking. An aliquot of extract was taken and the solvent was exchanged to 10/90 methanol/0.1% formic acid in 10 mM ammonium acetate. The sample was then analyzed for oxamyl and IN-A2213 metabolite using reversed phase high-performance liquid chromatography (HPLC) with mass spectrometry/mass spectrometry (MS/MS) detection. Positive ion electrospray ionization was used, and one parent → daughter ion transition was monitored for each analyte. During DuPont-38477, the samples were extracted in the same manner, but were subsequently diluted rather than being subjected to a solvent exchange.

The Limit of Quantitation (LOQ) was 0.005 mg/kg (ppm). The Limit of Detection (LOD) was estimated to be 0.001-0.003 mg/kg (ppm). The analytical method generated acceptable recoveries for samples fortified with oxamyl and IN-A2213 at 1X, 20X and 440X the LOQ. The recovery data obtained for oxamyl is summarized as follows:

Analysis of worms

Oxamyl was extracted from weed samples using the extraction procedure outlined in DuPont-40221, previously summarized in Point CA 4.1.2.15 of this document. Oxamyl was extracted from arthropod and earthworm samples using acetonitrile and a QuECHERS style cleanup. After partitioning, an aliquot of the supernatant was transferred into a 15 mL SampliQ EN dispersive SPE tube and vortexed. The extract was diluted and filtered prior to analysis.

Recovery findings

The fortification data reported in the method used to analyse oxamyl residues in earthworms and soil are summarised in Table 12.

Table 12 Validation data for analytical methods for the determination of oxamyl in earthworms and soil

Reference	Matrix	Number of tests	Fortification level (mg/kg)[a,b]	Average recovery (%)	Standard deviation	% Relative standard deviation
Oxamyl (237 → 90)						
DuPont-7191, Revision No. 1	Netherlands Soil	12 6 6 Total = 24	0.005 0.10 2.20	93 93 90 Mean = 92	6 7 10 Mean = 7	6 8 11 Mean = 8
	Spanish Soil	8 4 4 Total = 16	0.005 0.10 2.20	96 90 103 Mean = 96	12 13 11 Mean = 12	12 14 11 Mean = 12
Oxamyl (237 → 72)						
DuPont-38477	Soil	10 7 Total = 17	0.10 1.0	107 100 Mean = 104	7.9 8.2 Mean = 8	7.3 8.2 Mean = 8
	Earthworms	5 20 10 Total = 35	0.005 0.01 0.1	101 93 91 Mean = 94	2.5 11 8.5 Mean = 9.1	2.5 12 9.4 Mean = 9.9
IN-A2213 (163→73)						
DuPont-7191, Revision No. 1	Netherlands Soil	12 6 6 Total = 24	0.005 0.10 2.20	88 92 88 Mean = 89	6 5 9 Mean = 7	7 5 10 Mean = 7
	Spanish Soil	8 4 4 Total = 16	0.005 0.10 2.20	103 97 106 Mean = 102	7 4 13 Mean = 8	7 4 12 Mean = 8

[a] Fortifications were performed with analyte reference standard solutions
[b] Limit of quantification, defined by a signal-to-noise ratio of approximately 10 and the lowest validated fortification level

Linearity

Soil

During DuPont-7191, Revision No. 1, good linearity was observed in the range of 0.0014 to 0.22 ng/mL for oxamyl and IN-A2213. All calculations were performed using standards prepared in solvent.

During DuPont-38477, good linearity was observed in the range of 2.5 to 50 ng/mL for oxamyl. All calculations were performed using standards prepared in solvent.

Earthworms

Good linearity was observed in the range of 0.5 to 10.0 ng/mL for oxamyl. All calculations were performed using standards prepared in solvent.

Specificity

During DuPont-7191, Revision No. 1, the limit of determination of the methods used to monitor oxamyl residues in earthworms and soil was 0.001-0.003 mg/kg. Analysis of control samples from each sample type resulted in no detectable apparent residues of oxamyl or IN-A2213; the response in the area of the oxamyl and IN-A2213 peaks always corresponded to less than 20% of the limit of determination. It can therefore be concluded that few, if any, apparent residues or blank values would arise.

During DuPont-38477, the limit of determination of the methods used to monitor oxamyl residues in earthworms and soil was 0.001 mg/kg. Analysis of control samples from each sample type resulted in no detectable apparent residues of oxamyl; the response in the area of the oxamyl peak always corresponded to less than 20% of the limit of determination. It can therefore be concluded that few, if any, apparent residues or blank values would arise.

Limit of quantification

During DuPont-7191, Revision No. 1, the limit of quantification of the method for oxamyl and IN-A2213 residues in all matrices is 0.005 mg/kg.

During DuPont-38447, the limit of quantification of the method used to analyse soil samples was 0.1 mg/kg (the lowest fortification made with acceptable recovery). The limit of quantification of the method used to analyse earthworm samples was 0.01 mg/kg.

Repeatability

Repeatability of the method is addressed by the data in Table 12. Relative standard deviations of less than 20% were obtained for fortifications made at the quantification limit for each matrix, as well as at higher levels. Therefore, the repeatability of this method would seem to be adequate for the purposes of detecting oxamyl residues in arthropods, earthworms, and weeds.

Reproducibility

An independent laboratory validation of the analytical methods contained in DuPont-38477 were not conducted. These methods are proposed as data collection methods.

Extraction efficiency

Extraction efficiency for earthworm samples was not demonstrated in this study; however, extraction efficiency was evaluated through the analysis of tomato and foliage extracts during DuPont-44316 and summarized in Point CA 4.2.1 in this document.

Extraction efficiency of the method used to determine soil residues was evaluated through the analysis of freezer aged samples. The method extracted greater than 80% of the oxamyl from aged soil samples after a period of seven months.

Confirmatory method

Confirmatory methods are not required for data collection methods.

Overall suitability for regulatory purposes

This method is suitable for the collection of residue data in soil and earthworms. The instrumentation required to perform the analysis methods is available in most well equipped analytical laboratories and all of the sample preparation equipment is commercially available. The method does not require the use of untreated commodity to correct for recoveries.

Studies submitted to the EU for the first time in this submission.

B.5.1.2.15/06

Reference: CA 4.1.2/29	**Report**	Scherer, F. (2015); Oxamyl (DPX-D1410) 10GR: Investigating the deposition of dust from in-furrow application of granules containing oxamyl and determination of residues of oxamyl in guttation fluid of potato in United Kingdom during 2014 **DuPont Report No.:** DuPont-38691 **GLP:** Yes

Description of the method

Oxamyl residues were determined in potato guttation fluid by diluting 100 µL of guttation fluid with 400 µL of an acidified mixture of water and methanol (87.5/12.5 v/v). The autosampler was shaken to homogenize and submitted for LC/MS/MS analysis. Oxamyl residues were determined in dust catching solution by diluting the whole volume of the solution (125 mL) with 25 mL of an acidified water/methanol mixture (40/60 v/v) and shaking to homogenize. The homogenized solution was centrifuged and 1 mL submitted for LC/MS/MS analysis.

Recovery findings

The fortification data reported in the method proposed for monitoring oxamyl residues in soil are summarised in Table 13.

Table 13 Validation data for analytical methods for the determination of oxamyl in dust catching solution and potato guttation fluid (fortification data)

Reference	Matrix	Number of tests	Fortification level (mg/kg)[a,b]	Average recovery (%)	Standard deviation	% Relative standard deviation
Oxamyl (237 → 72)						
DuPont-38691	Dust Catching Solution	3 3 3 5 5 Total = 19	0.000008 0.00008 0.0008 0.001 0.010	108 97 98 106 100 Mean = 102	2.9 5.3 1.3 1.4 1.8 Mean = 2.3	2.7 5.5 1.3 1.3 1.8 Mean = 2.3
	Guttation Fluid	5 5 3 Total = 13	0.0010 0.010 100	103 95 101 Mean = 99	3.9 5.1 2.2 Mean = 4.0	3.8 5.4 2.2 Mean = 4.0

[a] Fortifications were performed with analyte reference standard solutions
[b] Limit of quantification, defined by a signal-to-noise ratio of approximately 10 and the lowest validated fortification level

Linearity

Good linearity was observed for oxamyl in the range of 0.002 to 0.20 ng/mL and 0.25 to 25 ng/mL for dust catching solution and from 0.05 to 4 ng/mL for guttation fluid. All calculations were performed using standards prepared in solvent.

Specificity

The limit of determination of the method for determining oxamyl residues in guttation fluid and dust catching solution was not determined. Analysis of control samples resulted in no detectable apparent residues of oxamyl. It can therefore be concluded that few, if any, apparent residues or blank values would arise.

Limit of quantification

The limit of quantification of the method for oxamyl 1.0 µg/kg.

Repeatability

Repeatability of the method is addressed by the data in Table 13. Relative standard deviations of less than 20% were obtained for fortifications made at the quantification limit for each matrix, as well as at higher levels. Therefore, the repeatability of this method would seem to be adequate for the purposes of oxamyl.

Reproducibility

An independent laboratory validation of DuPont-38691 was not conducted. DuPont-38691 is proposed as a data collection method and not as a monitoring method.

Extraction efficiency

Extraction efficiency was not determined during the course of this study. The collected samples were liquids that were diluted and analysed directly.

Confirmatory method

Confirmation of results were obtained using secondary LC/MS/MS ion transitions collected at the same time as the quantitative transitions. The recovery data obtained using the confirmatory procedure are summarised in Table.

Table 14 Validation data for analytical methods for the determination of oxamyl in dust catching solution and potato guttation fluid (confirmatory data)

Reference	Matrix	Number of tests	Fortification level (mg/kg)[a,b]	Average recovery (%)	Standard deviation	% Relative standard deviation
Oxamyl (237 → 90)						
DuPont-38691	Dust Catching Solution	3 3 3 5 5 Total = 19	0.000008 0.00008 0.0008 0.001 0.010	97 97 100 104 99 Mean = 100	5.0 5.9 1.5 1.5 1.7 Mean = 2.8	5.2 6.1 1.5 1.4 1.7 Mean = 2.8
	Guttation Fluid	5 5 3 Total = 13	0.0010 0.010 100	97 94 99 Mean = 96	2.9 4.6 3.5 Mean = 3.7	3.0 4.9 3.5 Mean = 3.8

[a] Fortifications were performed with analyte reference standard solutions
[b] Limit of quantification, defined by a signal-to-noise ratio of approximately 10 and the lowest validated fortification level

Overall suitability for regulatory purposes

This method is suitable for the collection of data from pollen samples for the purposes of ecotox studies. The instrumentation required to perform both the analysis and confirmatory methods is available in most well equipped analytical laboratories. No toxic or hazardous reagents are required to prepare the samples, and all of the sample preparation equipment is commercially available.

Studies submitted to the EU for the first time in this submission.

B.5.1.2.15/07

Reference: CA 4.1.2/01	**Report**	Berg, C. (2015a); Oxamyl (DPX-D1410) 10GR [100 g/kg]: Oxamyl (DPX-D1410) 10GR [100 g/kg (w/w)]: A semi-field study to evaluate effects on the bumble bee (*Bombus terrestris* L; Hymenoptera, Apidae) in *Solanum tuberosum* in Germany in 2014 **DuPont Report No.:** DuPont-39666 **GLP:** Yes

B.5.1.2.15/08

Reference: CA 4.1.2/02	**Report**	Berg, C. (2015b); Oxamyl (DPX-D1410) 10GR [100 g/kg]: A semi-field study to evaluate effects on the brood of honey bees (*Apis mellifera*; Hymenoptera, Apidae) in *Phacelia tanacetifolia* in Germany 2014 **DuPont Report No.:** DuPont-39667 **GLP:** Yes

B.5.1.2.15/09

Reference: CA 4.1.2/23	**Report**	Mack, P. (2015); Oxamyl (DPX-D1410) 10GR: Determination of residues of oxamyl in pollen, nectar, flowers, and guttation fluid of tobacco in southern Europe 2014 **DuPont Report No.:** DuPont-41322, Revision No. 1 **GLP:** Yes

Description of the method

Oxamyl residues were determined in guttation water, nectar, flowers and pollen matrices during DuPont-39666, DuPont-39667 and DuPont-41322, Revision No. 1. The residues were extracted from pollen specimen with acetonitrile/water (50:50, v/v) solution. After phase separation the lower acetonitrile phase was cleaned up by solid-phase extraction on activated carbon and primary amino phase. The eluate was evaporated to dryness and reconstituted in acetonitrile/water (50:50, v/v).

The residues were extracted from guttation water specimens with acetonitrile/water (50:50, v/v) solution. After phase separation the lower acetonitrile phase was removed, evaporated to dryness and reconstituted in acetonitrile/water (50:50, v/v).

The residues were extracted from nectar specimens with acetonitrile/water (50:50, v/v) solution. After phase separation the lower acetonitrile phase was removed, evaporated to dryness and reconstituted in acetonitrile/water (50:50, v/v).

The residues were extracted from flowers specimen with acetonitrile/water (50:50, v/v) solution. After phase separation the lower acetonitrile phase was cleaned up by solid-phase extraction on activated carbon and primary amino phase. The eluate was evaporated to dryness and reconstituted in acetonitrile/water (50:50, v/v).

The extracts of pollen, guttation water and flowers were analysed by HPLC-MS/MS employing electrospray ionisation in positive mode.

Recovery findings

The fortification data reported in the method proposed for measuring oxamyl residues in guttation water, nectar, flowers and pollen are summarised in Table 15.

Table 15 Validation data for analytical methods for the determination of oxamyl in guttation water, nectar, flowers and pollen (fortification data)

Reference	Matrix	Number of tests	Fortification level (mg/kg)[a,b]	Average recovery (%)	Standard deviation	% Relative standard deviation
Oxamyl (237 → 90)						
DuPont-39666 DuPont-39667	Guttation Water	5 5 5 Total = 15	0.0010 5.0 150	83 79 95 Mean = 86	7 6 3 Mean = 5	9 8 3 Mean = 7
DuPont-41322, Revision No. 1	Pollen	5 5 Total = 10	0.0010 5.0	107 92 Mean = 100	3 1 Mean = 2	3 1 Mean = 2
	Flowers	15 5 15 Total = 35	0.001 0.002 5.0	102 97 98 Mean = 100	4 2 4 Mean = 4	4 2 4 Mean = 4
	Nectar	10 10 Total = 20	0.001 5.0	83 88 Mean = 85	6 4 Mean = 5	7 5 Mean = 6

[a] Fortifications were performed with analyte reference standard solutions
[b] Limit of quantification, defined by a signal-to-noise ratio of approximately 10 and the lowest validated fortification level

Linearity

Good linearity was observed in the range of 0.040 to 100.0 ng/mL for oxamyl. All calculations were performed using standards prepared in blank matrix.

Specificity

The limit of determination of the method proposed for quantifying oxamyl in guttation fluid, flowers, nectar and pollen is 0.003 mg/kg. Analysis of control samples resulted in no detectable apparent residues of oxamyl. It can therefore be concluded that few, if any, apparent residues or blank values would arise.

Limit of quantification

The limit of quantification of the method for oxamyl residues in guttation water, nectar, pollen and flowers is 0.001 mg/kg.

Repeatability

Repeatability of the method is addressed by the data in Table 15. Relative standard deviations of less than 20% were obtained for fortifications made at the quantification limit for each matrix, as well as at higher levels. Therefore, the repeatability of this method would seem to be adequate for the purposes of detecting oxamyl residues in guttation water, nectar, pollen and flowers.

Reproducibility

An independent laboratory validation of the method used in DuPont-39666, DuPont-39667 and DuPont-41322, Revision No. 1 was not conducted. This method is proposed as a data collection method and not as a monitoring method.

Extraction efficiency

Extraction efficiency was not determined as part of this report.

Confirmatory method

Confirmation data were recorded simultaneous to quantitation data during the course of these studies by monitoring the signal arising from the 237→72 ion transition; however recoveries were not calculated.

Overall suitability for regulatory purposes

This method is suitable for the collection of data from guttation water, nectar, flower and pollen samples for the purposes of ecotox studies. The instrumentation required to perform both the analysis and confirmatory methods is available in most well equipped analytical laboratories. No toxic or hazardous reagents are required to prepare the samples, and all of the sample preparation equipment is commercially available.

RMS comments and conclusion: the methods presented are considered acceptable

B.5.1.2.16 Description of methods for determination of residues in water used in support of ecotoxicology studies

Studies submitted to the EU for the first time in this submission.

B.5.1.2.16/01

Reference: CA 4.1.2/07	**Report**	Brougher, D.S., Martin, K.H., Gallagher, S.P., Bodle, E.S. (2013a); Oxamyl (DPX-D1410) technical (98% w/w): A 48-hour static acute toxicity test with *Hyalella azteca* **DuPont Report No.:** DuPont-37397 **GLP:** Yes

B.5.1.2.16/02

Reference: CA 4.1.2/06	**Report**	Brougher, D.S. (2013); Oxamyl (DPX-D1410) technical (98% w/w): A 48-hour static acute toxicity test with the cladoceran (*Ceriodaphnia dubia*) **DuPont Report No.:** DuPont-37399 **GLP:** Yes

B.5.1.2.16/03

Reference: CA 4.1.2/08	**Report**	Brougher, D.S., Martin, K.H., Gallagher, S.P., Bodle, E.S. (2013b); Oxamyl (DPX-D1410) technical (98% w/w): A 48-hour static acute toxicity test with *Chironomus tentans* **DuPont Report No.:** DuPont-37400 **GLP:** Yes

B.5.1.2.16/04

Reference: CA 4.1.2/10	**Report**	Brougher, D.S., Martin, K.H., Gallagher, S.P., Krueger, H.O. (2013a); Oxamyl (DPX-D1410) technical (98% w/w): A 48-hour static acute toxicity test with the mayfly (*Centroptilum triangulifer*) **DuPont Report No.:** DuPont-37401 **GLP:** Yes

B.5.1.2.16/05

Reference: CA 4.1.2/11	**Report**	Brougher, D.S., Martin, K.H., Gallagher, S.P., Krueger, H.O. (2013b); Oxamyl (DPX-D1410) technical (98% w/w): A 48-hour static acute toxicity test with the caddisfly (*Chimarra atterima*) **DuPont Report No.:** DuPont-37402 **GLP:** Yes

Description of the method

These studies utilized standard calibration curves ranging form 0.005 to 0.350 mg/L to quantify the concentration of oxamyl in water. Water samples were collected and diluted with acidified freshwater, as needed, and analysed by high performance liquid chromatography (HPLC) with ultraviolet absorbance detection.

Recovery findings

The fortification data reported in the method proposed for determining oxamyl residues in water are summarised in Table 16.

Table 16 Validation data for analytical methods for the determination of oxamyl in water (fortification data)

Reference	Matrix	Number of tests	Fortification level (mg/L)[a,b]	Average recovery (%)	Standard deviation	% Relative standard deviation[c]
DuPont-37397	Fresh Water	2 2 2 Total = 6	0.0500 0.300 5.00	101 99 100 Mean = 100	1.4 1.2 0.57 Mean = 1.1	1.4 1.3 0.57 Mean = 1.1
DuPont-37399	Fresh Water	2 2 2 Total = 6	0.007 0.070 1.50	105 100 101 Mean = 102	2.1 0.57 1.4 Mean = 1.4	2.0 0.57 1.4 Mean = 1.3
DuPont-37400	Fresh Water	2 2 2 Total = 6	0.0500 0.500 1.20	100 99 102 Mean = 100	0.71 1.5 0.71 Mean = 0.97	0.71 1.5 0.70 Mean = 0.97
DuPont-37401	Fresh Water	2 2 2 Total = 6	0.010 0.070 0.700	86 95 100 Mean = 94	11 6.5 0.57 Mean = 6.0	13 6.8 0.57 Mean = 6.8
DuPont-37402	Fresh Water	2 2 2 Total = 6	0.00700 0.200 6.00	102 107 107 Mean = 105	5.9 4.9 4.2 Mean = 5	5.8 4.6 4.0 Mean = 4.8

[a] Fortifications were performed with analyte reference standard solutions

[b] Limit of quantification, defined by a signal-to-noise ratio of approximately 10 and the lowest validated fortification level

[c] Value estimated from duplicate samples using Microsoft Excel 2010

Linearity

Good linearity was observed in the range of 0.0050 to 0.350 mg/L for oxamyl in these studies.

Specificity

The limit of determination of the method proposed the determination of oxamyl residues in water ranged between 0.000158 to 0.00215 mg/L. Analysis of control resulted in no detectable level of oxamyl; the response in the area of the oxamyl peak always corresponded to less than 20% of the limit of determination.

Limit of quantification

The limit of quantification of the method for water ranged from 0.005 to 0.03 mg/L during the studies referenced above.

Repeatability

Repeatability of the method is addressed by the data in Table 16. The estimated relative standard deviations of less than 20% were obtained for fortifications made at the quantification limit for each matrix, as well as at higher levels.

Reproducibility

An independent laboratory validation of this method was not conducted. It is proposed as a data collection method for ecotox studies and not as an environmental monitoring method.

Confirmatory method

Confirmatory methods are not required for data collection methods.

Overall suitability for enforcement purposes

This method is suitable for the collection of data from water samples for the purposes of ecotox studies. The instrumentation required to perform both the analysis and confirmatory methods is available in most well equipped analytical laboratories. No toxic or hazardous reagents are required to prepare the samples, and all of the sample preparation equipment is commercially available.

Studies submitted to the EU for the first time in this submission.

B.5.1.2.16/06

Reference: CA 4.1.2/34	**Report**	[redacted], [redacted], [redacted] (2000b); Oxamyl 10L: Static, acute, 96-hour, (LC_{50}) test to rainbow trout, *Oncorhynchus mykiss* **DuPont Report No.:** DuPont-2910 **GLP:** Yes

B.5.1.2.16/07

Reference: CA 4.1.2/35	**Report**	[redacted], [redacted], [redacted] (2000c); Oxamyl 10L: Static-renewal, acute, 96-hour, (LC_{50}) test to bluegill sunfish, *Lepomis macrochirus* **DuPont Report No.:** DuPont-2911 **GLP:** Yes

Studies submitted to the EU for the first time in this submission

B.5.1.2.16/08

Reference: CA 4.1.2/33	**Report**	Ward, T.J., Magazu, J.P., Boeri, R.L. (2000a); Oxamyl 10L: Acute, static-renewal, 48-hour EC_{50} to *Daphnia magna* **DuPont Report No.:** DuPont-2556 **GLP:** Yes

B.5.1.2.16/09

Reference: CA 4.1.2/05	**Report**	Boeri, R.L., Ward, T.J. (2000); Oxamyl 10L: Influence on growth and growth rate of the alga, *Selenastrum capricornutum* **DuPont Report No.:** DuPont-3913 **GLP:** Yes

Description of the method

These studies utilized standard calibration curves ranging form 0.030 to 1.50 mg/L to quantify the concentration of oxamyl in water. An 8-mL sample of water was collected from the approximate center of each test chamber at mid-depth with a class A glass pipette. The samples were placed in 40-mL amber glass vials that contained 2 mL of acidified acetonitrile. If needed, the samples were diluted with an acidified solution of 80% water and 20% acetonitrile and subjected to HPLC/UV analysis.

Recovery findings

Experimental samples were fortified and analysed. The experimental samples were all in the range of 93–111% of their expected value. During DuPont-3913, a single laboratory control was fortified, with a recovery of 124%.

Linearity

Good linearity was observed in the range of 0.0300 to 1.50 mg/L for oxamyl in these studies.

Specificity

The limit of determination of the method proposed the determination of oxamyl residues in water ranged between 0.000587 to 0.0115 mg/L. Analysis of control resulted in no detectable level of oxamyl; the response in the area of the oxamyl peak always corresponded to less than 20% of the limit of determination.

Limit of quantification

The limit of quantification of the method for water ranged from 0.00196 to 0.0383 mg/L during the studies referenced above.

Repeatability

Repeatability was not addressed during these studies.

Reproducibility

An independent laboratory validation of this method was not conducted. It is proposed as a data collection method for ecotox studies and not as an environmental monitoring method.

Confirmatory method

Confirmatory methods are not required for data collection methods.

Overall suitability for enforcement purposes

This method is suitable for the collection of data from water samples for the purposes of ecotox studies. The instrumentation required to perform both the analysis and confirmatory methods is available in most well equipped analytical laboratories. No toxic or hazardous reagents are required to prepare the samples, and all of the sample preparation equipment is commercially available.

Studies submitted to the EU for the first time in this submission.

B.5.1.2.16/10

Reference: CA 4.1.2/19	**Report**	[redacted] (2012b); Oxamyl (DPX-D1410) technical (98% w/w): Early life-stage toxicity test with the sheepshead minnow, *Cyprinodon variegatus*, under flow-through conditions **DuPont Report No.:** DuPont-34270 **GLP:** Yes

Studies submitted to the EU for the first time in this submission.

B.5.1.2.16/11

Reference: CA 4.1.2/21	**Report**	Hicks, S.L. (2013); Oxamyl (DPX-D1410) technical (98% w/w): Life-cycle toxicity test of the saltwater mysid, *Americamysis bahia*, conducted under flow-through conditions **DuPont Report No.:** DuPont-34269 **GLP:** No

B.5.1.2.16/12

Reference: CA 4.1.2/28	**Report**	Rebstock, M. (2012b); Oxamyl (DPX-D1410) technical (98% w/w): Acute toxicity with the mysid shrimp, *Americamysis bahia*, determined under flow-through test conditions **DuPont Report No.:** DuPont-34271 **GLP:** Yes

Description of the method

A volume of 5 mL was collected from the control and each treatment vessel and transferred to 10-mL culture tubes. The 5-mL samples were diluted to 10 mL with methanol and further diluted, if necessary, with 50:50 methanol:water to a concentration within the range of the standard curve. Dilution factors ranged from 2 to 20 for test solution samples. QC fortification spikes were processed in a similar manner. Samples were vialed and analysed using LC-MS/MS. The standard curve ranged from 0.107 to 10.7 ng/mL.

Recovery findings

The fortification data reported in the method proposed for determining oxamyl residues in water are summarised in Table 17.

Table 17 Validation data for analytical methods for the determination of oxamyl in water

Reference	Matrix	Number of tests	Fortification level (mg/L)[a,b]	Average recovery (%)	Standard deviation	% Relative standard deviation
DuPont-34269	Water	7 7 Total = 14	0.000648 0.0702	87 95 Mean = 91	8.0 6.7 Mean = 7.4	9.2 7.1 Mean = 8.2
DuPont-34270	Water	3 3 Total = 6	0.0532 53.2	101 103 Mean = 102	1.2 3.6 Mean = 2.4	1.2 3.5 Mean = 2.4
DuPont-34271	Water	3 3 Total = 6	0.00200 0.106	94 105 Mean = 100	5.8 5.7 Mean = 5.8	6.2 5.4 Mean = 5.8

[a] Fortifications were performed with analyte reference standard solutions
[b] Limit of quantification, defined by a signal-to-noise ratio of approximately 10 and the lowest validated fortification level

Linearity

Good linearity was observed in the range of 0.107 to 10.7 ng/mL for oxamyl in these studies.

Specificity

The limit of determination of the method proposed the determination of oxamyl residues in water was 0.00000537 mg/L. Analysis of control resulted in no detectable level of oxamyl; the response in the area of the oxamyl peak always corresponded to less than 20% of the limit of determination.

Limit of quantification

The limit of quantification of the method for water was 0.0000179 mg/L during the studies referenced above.

Repeatability

Repeatability of the method is addressed by the data in Table 17. Relative standard deviations of less than 20% were obtained for fortifications made at the quantification limit for each matrix, as well as at higher levels. Therefore, the repeatability of this method would seem to be adequate for the purposes of detecting oxamyl residues in water.

Reproducibility

An independent laboratory validation of this method was not conducted. It is proposed as a data collection method for ecotox studies and not as an environmental monitoring method.

Confirmatory method

Confirmatory methods are not required for data collection methods.

Overall suitability for enforcement purposes

This method is suitable for the collection of data from water samples for the purposes of ecotox studies. The instrumentation required to perform both the analysis and confirmatory methods is available in most well equipped analytical laboratories. No toxic or hazardous reagents are required to prepare the samples, and all of the sample preparation equipment is commercially available.

Studies submitted to the EU for the first time in this submission.

B.5.1.2.16/13

Reference: **CA 4.1.2/18**	**Report**	[redacted] [redacted] (2012a); Oxamyl technical (DPX-D1410): Short term reproduction assay with the fathead minnow, *Pimephales promelas*, determined under flow-through conditions **DuPont Report No.:** DuPont-31031 **GLP:** Yes

B.5.1.2.16/14

Reference: **CA 4.1.2/16**	**Report**	[redacted] [redacted] (2015); Oxamyl (DPX-D1410) technical: 21-D amphibian metamorphosis assay (AMA) with south African clawed frog, *Xenopus laevis* **DuPont Report No.:** DuPont-31032, Revision No. 1 **GLP:** Yes

Studies submitted to the EU for the first time in this submission.

B.5.1.2.16/15

Reference: CA 4.1.2/27	**Report**	Rebstock, M. (2012a); Oxamyl (DPX-D1410) technical (98% w/w): 7-day growth inhibition test with the freshwater aquatic plant, duckweed, *Lemna gibba* **DuPont Report No.:** DuPont-34272 **GLP:** Yes

B.5.1.2.16/16

Reference: CA 4.1.2/20	**Report**	Hicks, S.L. (2012c); Oxamyl (DPX-D1410) technical (98% w/w): Effect on new shell growth of the eastern oyster (*Crassostrea virginica*) **DuPont Report No.:** DuPont-34273 **GLP:** Yes

B.5.1.2.16/17

Reference: CA 4.1.2/03	**Report**	Bergfield, A. (2012a); Oxamyl (DPX-D1410) 24SL: A greenhouse study to investigate the effects on seedling emergence and growth of tomato, *Lycopersicon esculentum*, following soil exposure **DuPont Report No.:** DuPont-34274 **GLP:** Yes

B.5.1.2.16/18

Reference: CA 4.1.2/04	**Report**	Bergfield, A. (2012b); Oxamyl (DPX-D1410) 24SL: A greenhouse study to investigate the effects on vegetative vigor of ten terrestrial plant species following foliar exposure **DuPont Report No.:** DuPont-34275 **GLP:** Yes

Description of the method

A volume of 5 mL was collected from the control and each treatment vessel and transferred to 10-mL culture tubes. The 5-mL samples were diluted to 10 mL with methanol and further diluted, if necessary, with 50:50 methanol:water to a concentration within the range of the standard curve. QC fortification spikes were processed in a similar manner. Samples were vialed and analysed using LC/UV.

Recovery findings

The fortification data reported in the method proposed for determining oxamyl residues in water are summarised in Table 18.

Table 18 Validation data for analytical methods for the determination of oxamyl in water (fortification data)

Reference	Matrix	Number of tests	Fortification level (mg/L)[a,b]	Average recovery (%)	Standard deviation	% Relative standard deviation[c]
DuPont-31031	Water	3 3	0.0792 55.0	111 119 Mean = 115	0 1 Mean = 1	0 1 Mean = 1
DuPont-31032	Water	4 4	0.013 0.040 0.120	82 100 108 Mean = 97	7.1 2.5 1.2 Mean = 3.6	8.7 2.5 1.1 Mean = 4.1
DuPont-34272	20X AAP Medium in Water	3 3 Total = 6	0.0798 53.2	104 106 Mean = 105	1.5 0.58 Mean = 1.0	1.5 0.55 Mean = 1.0
DuPont-34273	Water	3 3 Total = 6	0.0532 143	99 103 Mean = 101	9.9 2.9 Mean = 6.4	9.9 2.8 Mean = 6.4
DuPont-34274	Water	3 3 Total = 6	0.00291 0.00969	87 92 Mean = 90	0.58 1.5 Mean = 1.0	0.67 1.7 Mean = 1.2
DuPont-34275	Spray Mixture	2 2	2.91 9.69	111 113 Mean = 112	9.9 11 Mean = 11	8.9 9.4 Mean = 9.2

[a] Fortifications were performed with analyte reference standard solutions
[b] Limit of quantification, defined by a signal-to-noise ratio of approximately 10 and the lowest validated fortification level
[c] Estimated from two samples using Microsoft Excel 2010

Linearity

Good linearity was observed in the range of 0.0214 to 1.07 mg/L for oxamyl in these studies.

Specificity

The limit of determination of the method proposed the determination of oxamyl residues in water ranged between 0.0062 to 0.0111 mg/L. Analysis of control resulted in no detectable level of oxamyl; the response in the area of the oxamyl peak always corresponded to less than 20% of the limit of determination.

Limit of quantification

The limit of quantification of the method for water ranged from 0.021 to 0.0370 mg/L during the studies referenced above.

Repeatability

Repeatability of the method is addressed by the data in Table 18. Relative standard deviations of less than 20% were obtained for fortifications made at the quantification limit for each matrix, as well as at higher levels. Therefore, the repeatability of this method would seem to be adequate for the purposes of detecting oxamyl residues in water.

Reproducibility

An independent laboratory validation of this method was not conducted. It is proposed as a data collection method for ecotox studies and not as an environmental monitoring method.

Confirmatory method

Confirmatory methods are not required for data collection methods.

Overall suitability for enforcement purposes

This method is suitable for the collection of data from water samples for the purposes of ecotox studies. The instrumentation required to perform both the analysis and confirmatory methods is available in most well equipped analytical laboratories. No toxic or hazardous reagents are required to prepare the samples, and all of the sample preparation equipment is commercially available.

Study submitted to the EU for the first time in this submission.

B.5.1.2.16/19

Reference: CA 4.1.2/09	**Report**	Brougher, D.S., Martin, K.H., Gallagher, S.P., Bodle, E.S. (2013c); Oxamyl (DPX-D1410) technical (98% w/w): A 48-hour static acute toxicity test with the cladoceran (*Daphnia pulex*) **DuPont Report No.:** DuPont-37398 **GLP:** Yes

Description of the method

Calibration curves ranging from 0.00100 to 0.0100 mg/L were prepared. Freshwater samples were taken and diluted in 10/90/0.1 (v/v/v) methanol/freshwater/formic acid, as needed, and analysed by high performance liquid chromatography with tandem mass spectrometric detection (LC-MS/MS).

Recovery findings

The fortification data reported in the method proposed for determining oxamyl residues in water are summarised in Table 19.

Table 19 Validation data for analytical methods for the determination of oxamyl in water

Reference	Matrix	Number of tests	Fortification level (mg/L)[a,b]	Average recovery (%)	Standard deviation	% Relative standard deviation[c]
DuPont-37398	Fresh Water	2 2 2 Total = 6	0.00400 0.0500 1.50	100 99 101 Mean = 100	1.6 3.2 0.71 Mean = 1.8	1.6 3.2 0.70 Mean = 1.8

[a] Fortifications were performed with analyte reference standard solutions
[b] Limit of quantification, defined by a signal-to-noise ratio of approximately 10 and the lowest validated fortification level
[c] Value estimated from duplicate samples using Microsoft Excel 2010

Linearity

Good linearity was observed in the range of 0.00100 to 0.0100 mg/L for oxamyl in these studies.

Specificity

The limit of determination of the method proposed the determination of oxamyl residues in water was 0.0000880 mg/L. Analysis of control resulted in no detectable level of oxamyl; the response in the area of the oxamyl peak always corresponded to less than 20% of the limit of determination.

Limit of quantification

The limit of quantification of the method for water ranged was defined as 0.00111 mg/L during this study.

Repeatability

Repeatability is addressed in Table 19. The percent RSD for the recovery of oxamyl was less than 20% regardless of the level chosen.

Reproducibility

An independent laboratory validation of this method was not conducted. It is proposed as a data collection method for ecotox studies and not as an environmental monitoring method.

Confirmatory method

Confirmatory methods are not required for data collection methods.

Overall suitability for enforcement purposes

This method is suitable for the collection of data from water samples for the purposes of ecotox studies. The instrumentation required to perform both the analysis and confirmatory methods is available in most well equipped analytical laboratories. No toxic or hazardous reagents are required to prepare the samples, and all of the sample preparation equipment is commercially available.

Studies submitted to the EU for the first time in this submission.

B.5.1.2.16/20

Reference: CA 4.1.2/30	**Report**	Schmitt, H. (2014); Oxamyl (DPX-D1410) technical (98% w/w): Assessment of effects to the honeybee, *Apis mellifera* L., in a 10 days chronic feeding test under laboratory conditions **DuPont Report No.:** DuPont-39665 **GLP:** Yes

B.5.1.2.16/21

Reference: CA 4.1.2/22	**Report**	Klank, C. (2014); Oxamyl (DPX-D1410) technical (98% w/w): Honey bee (*Apis mellifera* L.) larval toxicity test (single feeding exposure) **DuPont Report No.:** DuPont-39678 **GLP:** Yes

Description of the method

After sampling, the samples were stored deep-frozen (below -18°C) until analysis. At the analytical laboratory, the samples were thawed and shaken well using a Vortex mixer. Recovery samples were prepared by fortification of untreated control sample with the test item. The samples were diluted with methanol/water (1:1, v/v) prior to analysis by HPLC-MS/MS.

Recovery findings

The fortification data reported in the method proposed for determining oxamyl residues in water are summarised in Table 20.

Table 20 Validation data for analytical methods for the determination of oxamyl in water

Reference	Matrix	Number of tests	Fortification level (mg/L)[a,b]	Average recovery (%)	Standard deviation	% Relative standard deviation
DuPont-39665	Water	5 5 Total = 10	0.100 30	82 105 Mean = 94	5 2 Mean = 4	6 2 Mean = 4
DuPont-39678	Water	5 5 Total = 10	0.100 400	86 102 Mean = 94	5 1 Mean = 3	6 1 Mean = 4

[a] Fortifications were performed with analyte reference standard solutions
[b] Limit of quantification, defined by a signal-to-noise ratio of approximately 10 and the lowest validated fortification level

Linearity

Standards were analysed in the range of 1 to 100 ng/mL of oxamyl in these studies. The resulting calibration curves were quadratic with r^2 values greater than 0.999.

Specificity

The limit of determination of the method proposed the determination of oxamyl residues in water was estimated to be 0.03 mg/L. Analysis of control resulted in no detectable level of oxamyl; the response in the area of the oxamyl peak always corresponded to less than 20% of the limit of determination.

Limit of quantification

The limit of quantification of the method for water was 0.100 mg/L for the studies referenced above.

Repeatability

Repeatability is addressed in Table 20. The percent RSD for the recovery of oxamyl was always less than 20%, indicating the method is appropriate for the analysis of oxamyl in water.

Reproducibility

An independent laboratory validation of this method was not conducted. It is proposed as a data collection method for ecotox studies and not as an environmental monitoring method.

Confirmatory method

Confirmatory methods are not required for data collection methods.

Overall suitability for enforcement purposes

This method is suitable for the collection of data from water samples for the purposes of ecotox studies. The instrumentation required to perform both the analysis and confirmatory methods is available in most well equipped analytical laboratories. No toxic or hazardous reagents are required to prepare the samples, and all of the sample preparation equipment is commercially available.

RMS comments and conclusion: the study presented is considered acceptable.

B.5.1.2.17 Description of methods for determination of residues in water from the physical and chemical properties tests

B.5.1.2.17/01

Reference: **CA 4.1.2/17**	**Report**	Harsha, N.V. (2009); Oxamyl (DPX-D1410-196): Laboratory study of vapour pressure **DuPont Report No.:** DuPont-26259 **GLP:** Yes

Description of the method

Recovery tests were performed on both the trapping agent (silica gel) and solid support (sea sand) as part of this study. For the trapping agent, 3.08 mg aliquot of test item (purity 98.0%) was accurately weighed into a 10-mL volumetric flask, dissolved, and brought to volume with acetone. A secondary stock solution of 30 mg/L was prepared by diluting 1 mL of stock solution to 10 mL in a volumetric flask. A 60 g sample of silica gel (60 to 120-mesh size) was weighed in duplicate and transferred to 500-mL beakers. The beakers were labeled as S1 and S2. A 1-mL aliquot of the primary and secondary stock solutions were added to the beaker S2 and S1, respectively. The contents of the beakers were mixed manually for approximately 5 minutes, providing a uniform distribution. The beakers were then placed in a fume hood to evaporate acetone. The sorbent traps were then prepared by adding 10 g of coated silica gel into the glass tubes, plugged with cotton swabs. The glass

tubes were connected to the empty stainless steel columns, and nitrogen gas was passed through the tubes at a uniform rate of 7 mL/min. for a period of 7 days. After 7 days, the sorbent traps were removed, and the silica gel was removed and eluted with 100 mL of methanol. The eluate was concentrated, and the recovery of test item was determined. An uncoated silica gel sample was maintained as a control for comparison.

Recovery from the solid support (sea sand) was tested at two concentration levels. The lower concentration level was prepared by adding 1 mL of 301.84 mg/L oxamyl solution to 20 g of sea sand. The contents were mixed manually for about 5 minutes to ensure uniform distribution, and the beaker was placed in a fume hood to evaporate the acetone. For the higher concentration level, oxamyl coated solid support materials (sea sand) from a preliminary study was used. From each treated level, six replicate samples were weighed into extraction bottles, 25 mL of methanol was added, and the samples were placed on a shaker for 30 minutes. The samples were filtered to remove the solid materials. Each diluted extract was assayed, and the relative standard deviation for each treated sand sample was calculated.

Recovery findings

The fortification data reported in the method for monitoring oxamyl in this study are summarised in Table 21. The average recoveries and relative standard deviation are well within the criteria specified in the EU Guidance Document on Residue Analytical Methods (SANCO/825/00 rev.8.1).

Table 21 Validation data for analytical methods for the determination of oxamyl in sea sand and silica gel for the determination of vapour pressure

Reference	Matrix	Number of tests	Fortification level (mg/kg)[a,b]	Average recovery (%)	Standard deviation	% Relative standard deviation
DuPont-26259	Trapping Agent (Silica Gel)	6 6 Total = 12	0.50 5.0	96 97 Mean = 97	2.5 1.6 Mean = 2.1	2.6 1.6 Mean = 2.1
	Solid Support (Sea Sand)	6 6 Total = 12	15 13880	100 100 Mean = 100	5.1 3.9 Mean = 4.5	5.1 3.9 Mean = 4.5

[a] Fortifications were performed with analyte reference standard solutions

[b] Limit of quantification, defined by a signal-to-noise ratio of approximately 10 and the lowest validated fortification level

Linearity

Good linearity was observed over the range of 0.01 to 25 mg/L. All calculations were performed using standards prepared in solvent.

Specificity

Aliquots of the samples from each test system, methanol, milliQ water, mobile phase, sea sand control, and silica gel control were assayed to check the method specificity. No significant interference was observed from methanol, milliQ water, or mobile phase. Analysis of uncoated sand and silica gel samples also did not show any interference.

Limit of quantification

The limit of quantification of the method was determined to be 0.01 mg/L.

Repeatability

The relative standard deviation for the six measurements of peak area from the repeated analysis of a 0.5 mg/L standard was 0.90%, which indicated an acceptable system precision. Experimental samples were analysed in triplicate.

Reproducibility

All samples were analysed in triplicate; no additional reproducibility analysis was conducted.

Confirmatory method

Confirmation is not needed for physical chemical property studies.

Overall suitability for enforcement purposes

This method is suitable for determining the vapour pressure of oxamyl. The instrumentation required to perform the analysis method is available in most well equipped analytical laboratories.

RMS comments and conclusion: the study presented is considered acceptable

B.5.2 Methods for post-approval control and monitoring purposes

Proposals for enforcement and monitoring methods

Justification for analytes chosen

The proposed residue definition in plants based metabolism studies summarised in the Oxamyl RAR Vol.3 B7 AS for oxamyl is parent only. The proposed residue definition in animal tissues, milk, and eggs based metabolism studies summarised in the Oxamyl RAR Vol.3 B7 AS for oxamyl is parent only.

The proposed environmental residue definition in soil and sediment based metabolism studies summarised in the Oxamyl RAR Vol.3 B7 ASfor oxamyl is parent only. The proposed residue definition in water based metabolism studies summarized in the Oxamyl RAR Vol.3 B7 AS for oxamyl is parent only. The proposed environmental residue definition in air for oxamyl is parent only.

Table 22 Proposed analytical methods for monitoring oxamyl residues

Matrix	Reference and report	Separation/ Quantitation	Limit of determination (mg/kg)	Comments
Dry, Watery, Oily, Acidic and Difficult to Analyse Crops	DuPont-41730	QuECHERS MRM HPLC/MS/MS	0.01	QuECHERS MRM
Meat (Bovine) Fat (Bovine) Liver (Bovine) Milk Eggs	DuPont-41763	QuECHERS MRM HPLC/MS/MS	0.01	QuECHERS MRM
Soil	DuPont-38689	HPLC-MS/MS	0.001	DuPont single analyte method
Water	DuPont-5677	HPLC-MS/MS	0.0001 mg/L	DuPont single analyte method.
Air	DuPont-4564	HPLC-MS/MS	0.25 $\mu g/m^3$	DuPont single analyte method
Body Fluids and Tissues	DuPont-38598, Revision No. 1	HPLC-MS/MS	0.05 mg/L	DuPont single analyte method

B.5.2.1 Description of monitoring methods for determination of residues in plants, plant products and processed food commodities.

Study submitted in the EU Dossier in 2001 and included in the first EU approval review.

B.5.2.1/01

Reference: --	**Report**	Françon, B., Jernberg, K.M., Jetzer, M., Steiner, C. (2001); Method validation of the Netherlands multi-residue method 2 (MRM 2, submethod 1: N-Methylcarbamate pesticides) for the determination of oxamyl in vegetable and fruit crops **DuPont Report No.:** DuPont-4722 **GLP:** Yes

Analytical method:

Improved cleanup method for the multiresidue analysis of N-methylcarbamates in grains, fruits and vegetables by means of HPLC with post column reaction and fluorescence detection.

Purpose of the study:

To independently validate the Dutch Multi residue for the analysis of Oxamyl in the matrices, melon, lettuce, sugar beet, potatoes and in citrus fruit.

Principle of the method:

As indicated above in section B.5.2.1.2. Oxamyl is extracted with acetone from the food matrix and then partitioned into dichloromethane/petroleum spirit. A portion of the extract is evaporated to dryness and may be cleaned up by SPE [aminopropyl-bonded silica cartridges]. The sample is then analysed using reverse phase HPLC coupled to a post column hydrolyser after which the methyl amine released is derivatised with o-phthaldehyde. A Fluorescence detector is used to detect the derivative formed.

Validation data: Using the recommended HPLC equipment with a fluorescence detector.

Table 23 Recovery values and %RSD for the analysis of Oxamyl in a range of sample matrices

Sample matrix.	Fortification level (mg/kg)	Number of replicates	Mean recovery %	RSD % (n=5)	Overall RSD% (n= 10)
Melon pulp	0.01	5	93 ±4	4	14
	0.1	5	73 ±7	10	
Melon peel	0.01	5	99 ±13	13	15
	0.1	5	88 ±15	17	
Lettuce leaf.	0.01	5	92 ±17	19	20
	0.1	5	80 ±16	20	
Sugar beet, root.	0.01	5	103 ±6	6	9
	0.1	5	94 ±11	12	
Sugar beet,	0.01	5	95 ±9	9	17
	0.1	5	81 ±15	19	
Potato tuber	0.01	5	108 ±3	3	11
	0.1	5	91 ±9	10	
Citrus fruit, peel.	0.01	5	87 ±9	10	9
	0.1	5	82 ±6	8	
Citrus fruit, pulp.	0.01	5	86 ±17	20	18
	0.1	5	74 ±10	13	

Confirmatory Method: Confirmatory analyses were carried out using the notifier's own developed method [Analytical method for the determination of Oxamyl and its Oxime metabolite in soil using LC/MS analysis, DuPont Study no. DuPont-2392"] which extracts, cleans up and analyses the sample directly using LC/MS without the hydrolysis and derivatisation process. This method is outlined in section B.5.3.1.1.. In the present situation the method was validated only for the analysis of Oxamyl residues. No validation data was generated for the Oxime metabolite.

Validation data : Additional validation data was generated for the LC/MS method as follows.

Table 24 Validation data for the confirmatory LC/MS method for the analysis of Oxamyl

Matrix analysed.	Fortification levels (mg/kg)	Number of replicates analysed.	Mean Recovery (%).
Melon pulp	0.01	2	74
	0.1	2	70
Lettuce leaf.	0.01	2	72
	0.1	2	72
Potato Tuber.	0.01	2	82
	0.1	2	77
Citrus fruit pulp.	0.01	2	89
	0.1	2	74

Conclusion: The validation data presented by the notifier confirms that the Analytical method, Netherlands Multi-Residue Method 2, developed by the Dutch Food control Inspectorate is suitable for the analysis of Oxamyl residues in food of plant origin. In addition the further validation data generated using LC/MS confirms the suitability of the LC method which uses fluorescence detection and indicates that oxamyl residues can be readily analysed using LC/MS without having to use the hydrolysis and derivatisation steps.

The monitoring method for determination of residues in plants, plant products and processed food commodities, DuPont-4722, originally submitted under EU Rev8 Point IIA 4.2.1.2, was conducted under guidelines Directive 91/414/EEC and SANCO 825/00-rev 6 (2000). A review of this study indicates that it partially meets the current guideline SANCO/825/00 rev 8.1; deviations include too few confirmatory replicates and the use of category 2 solvents during extraction. However, reconduct is unlikely to yield a significantly different result based on the method's precision and recovery statistics and this method can be relied upon.

Study submitted in the EU Dossier in 2001 and included in the first EU approval review.

B.5.2.1/02

Reference: --	**Report**	Bacher, R. (2001); Analytical method for the determination of oxamyl and methomyl in animal food stuffs **DuPont Report No.:** DuPont-5132 **GLP:** Yes

Principle of the method:

The sample [50g of milk, 25g of bovine meat, 25g of egg] is extracted by homogenising with ethyl acetate (100 ml). The extract is centrifuged and the supernatant is filtered. The extraction process is repeated twice more. The sample extract is cleaned up using GPC. The GPC fraction is evaporated and the residues of Oxamyl and methomyl are hydrolysed to their respective oximes. The oxime metabolites are then analysed using gas chromatography coupled to a mass spectrometer.

In the case of the Oxamyl oxime quantitation is carried out using the 72 m/z while the 115, 145 and 162/163 m/z are used for confirmation of identity. In the case of the Methomyl oxime quantitation is carried out at the 88 m/z while the 58 and 105 m/z are used for confirmation. GC with an FPD detector used in the Sulfur sensitive mode is used to confirm the identity of the oximes.

Validation data:

Linearity: The linearity of the method was determined for the analysis of Oxamyl Oxime at 9 different concentration levels between 0.01 and 5.0 mg/l. The correlation co-efficient for the calibration range was determined to be = 0.9894 when using GC/MS.

Recovery studies: Recovery studies were carried out using whole milk, egg and bovine muscle. The results of these studies are presented in Table 25 below.

Table 25 Recovery data for the determination of Oxamyl residues in food of animal origin

Fortification level(mg/kg)	Recovery results.	Oxamyl			Methomyl.		
		Whole milk	Bovine muscle	Whole egg	Whole milk	Bovine muscle	Whole egg.
0.01	Average	80%	92%	100%	103%	94%	98%
	RSD	8%	7%	5%	3%	18%	8%
	n	6	5	5	6	5	5
0.1	Average	84%	91%	102%	100%	99%	96%
	RSD	8%	11%	7%	7%	4%	10%
	n	6	6	5	6	5	5
Overall	Average	82%	91%	101%	102%	97%	97%
	RSD	8%	9%	6%	5%	12%	9%
	n	12	11	10	12	10	10

Sample chromatographic charts were presented to show the conditions encountered when analysing for residues of Oxamyl and Methomyl in milk, meat and eggs. The chromatograms were generated for both GC/MS and GC/FPD analytical systems and both systems are satisfactory for the different matrices.

Conclusion:

The method of analysis presented is validated for the analysis of Oxamyl and Methomyl residues present in milk, meat and in eggs as their oxime metabolites. The method is considered to be acceptable.

The monitoring method for determination of residues in plants, plant products and processed food commodities, DuPont-5132, originally submitted under EU Rev8 Point IIA 4.2.1.2, was conducted under guidelines Directive 91/414/EEC, SANCO/825/00 rev. 6 (2000), and OPPTS 860.1340. A review of this study indicates that it

partially meets the current guideline SANCO/825/00 rev 8.1; deviations include too few confirmatory replicates. However, based on the method's precision and recovery statistics, reconduct is unlikely to yield a significantly different result. Therefore, this method is relied upon

Enforcement methods suitable for the European Union region

Studies submitted to the EU for the first time in this submission.

B.5.2.1/03

Reference: CA 4.2/08	**Report**	Lissemore, L., Harris, A., Patterson, C. (2014b); QuEChERS multiresidue method trials for DPX-D1410, DPX-X1179 and DPX-Q8U80 in crop matrices **DuPont Report No.:** DuPont-41730 **Test facility**: University of Guelph, Ontario (Canada) **GLP:** Yes

B.5.2.1/04

Reference: CA 4.2/09	**Report**	Schernikau, N., Colorado, C.S. (2015); Independent laboratory validation of DuPont-41730 and DuPont-41763, "Analytical method for the determination of oxamyl (DPX-D1410) and methomyl (DPX-X1179) in crop and animal matrices by LC/ESI-MS/MS" **DuPont Report No.:** DuPont-41873 **Test facility**: Eurofins Agroscience Service, Hamburg, Germany **GLP:** Yes

B.5.2.1/05

Reference: CA 4.2/10	**Report**	Cochrane, J., (2015) Determination of the Extraction Efficiency of [^{14}C] DPX-D1410 Residue Methods: QuEChERS (DuPont-41730) and Crop Method (DuPont-17601 RV.1) **DuPont Report No.:** DuPont-44316 **GLP:** Yes

Description of the method

The QuECHERS MRM was validated for the analysis of oxamyl residues in dry, watery, acidic, oily, and difficult to analyse crops. Two grams of a given crop matrix are weighed into a 50 mL polypropylene centrifuge tube. To each sample, 100 µL of the internal standard Carbofuran-d3 was added, and the samples were fortified with oxamyl. Ten millilitres of water and 10 mL of 1% acetic acid in acetonitrile were added to the samples along with 1g of anhydrous sodium acetate. The samples were then vortexed for 20 seconds. After the samples had been vortexed, 4 g of anhydrous magnesium sulphate were added to the sample, and the samples were placed on a genogrinder at 1700 rpm for 1 minute. The samples were then centrifuged at 3600 rpm for 5 minutes. Following centrifugation, 5.0 mL of the supernatant was pipetted into a 15 mL polypropylene centrifuge tube and blow down to between 0.2 and 0.3 mL under a stream of nitrogen. This sample was then sequentially diluted with methanol, ammonium acetate and a 1:1 solution of 0.1 M ammonium acetate/methanol. The samples were then syringe filtered into amber vial for LC-MS/MS analysis. Quantitation and confirmation signals for oxamyl were generated from its $[M+NH_4]^+$ ion, 237 m/z. Quantitation of oxamyl was based on the signal arising from the 237 → 72 ion transition. Confirmation was based on the signal arising from the 237 → 90 ion transition.

Recovery findings

The fortification data reported in the method proposed for monitoring oxamyl residues in crop samples are summarised in Table 26. The average recovery specified in the decision-making criteria is 70 to 120%, with a standard deviation of ≤20%. Therefore, the recovery of this method is adequate for the purposes of residue data collection and enforcement of MRLs.

Table 26 Validation data for analytical methods for the determination of oxamyl in food of plant origin

Matrix	Fortification level (mg/kg)[a,b]	Number of tests	Average recovery (%)	Standard deviation	% Relative standard deviation	Reference
Oxamyl (237 → 72)						
Grapes	0.01 0.10	5 5 Total = 10	94 102 Mean = 98	1.6 1.5 Mean = 1.6	1.7 1.5 Mean = 1.6	DuPont-41730
Tomatoes	0.01 0.10	5 5 Total = 10	101 107 Mean = 104	1.9 2.7 Mean = 2.3	1.9 2.5 Mean = 2.2	
Avocado	0.01 0.10	5 5 Total = 10	80 103 Mean = 92	12 2.5 Mean = 7.3	15 2.4 Mean = 8.7	
Wheat	0.01 0.10	5 5 Total = 10	82 106 Mean = 94	4.5 1.3 Mean = 2.9	5.5 1.2 Mean = 3.4	
Tobacco	0.01 0.10	5 5 Total = 10	92 97 Mean = 95	3.8 2.6 Mean = 3.2	4.1 2.7 Mean = 3.4	

[a] Fortifications were performed with analyte reference standard solutions
[b] Limit of quantification, defined by a signal-to-noise ratio of approximately 10 and the lowest validated fortification level

Linearity

Good linearity was observed in the range of 0.05 to 4 ng/mL for oxamyl. Six solutions were injected.

Specificity

The limit of quantification of the method proposed for monitoring oxamyl residues is 0.010 mg/kg for all crops tested. Analysis of control samples resulted in no detectable apparent residues of oxamyl; the response in the area of the oxamyl peak always corresponded to less than 20% of the limit of determination. It can therefore be concluded that few, if any, apparent residues or false positive values would arise.

Limit of quantification

The limit of quantification of the method for the five EU crop groups (dry, acetic, watery, oily, and difficult to analyse) is 0.010 mg/kg.

Repeatability

Repeatability of the method is addressed by the data in Table 26. Relative standard deviations of less than 20% were obtained for fortifications made at the quantification limit for each matrix, as well as at higher levels. Therefore, the repeatability of this method is adequate for the purposes of residue data collection and enforcement of MRLs.

Reproducibility

An independent laboratory validation of DuPont-41730 was conducted, and the results are presented in Table 27. The primary method is identical for all matrix groups, so it is sufficient to perform the ILV for commodities of two of these groups, one of them with high water content (tomatoes). A validated primary method was also required for tobacco, since it is a difficult to analyse commodity.

Table 27 Validation data for analytical methods for the determination of oxamyl in food of plant origin

Matrix	Fortification level (mg/kg)[a,b]	Number of tests	Average recovery (%)	Standard deviation	% Relative standard deviation	Reference
Oxamyl (237 → 72)						
Grapes	0.01 0.10	5 5 Total = 10	80 94 Mean =87	4.8 4.2 Mean = 4.5	6.1 4.5 Mean = 5.3	DuPont-41873
Tomatoes	0.01 0.10	5 5 Total = 10	88 104 Mean = 96	7.5 11 Mean = 9.3	8.5 11 Mean = 9.8	
Tobacco	0.01 0.10	5 5 Total = 10	84 83 Mean = 83	4.5 4.9 Mean = 4.7	5.4 5.9 Mean = 5.7	

[a] Fortifications were performed with analyte reference standard solutions
[b] Limit of quantification, defined by a signal-to-noise ratio of approximately 10 and the lowest validated fortification level

Extraction efficiency

The extraction efficiency of this method was investigated in DuPont-44316 by extracting tomato samples with incurred, radiolabelled residues. Tomato foliage and fruit were extracted using the method found in DuPont-41730 and compared to the metabolism method. On average, DuPont-41730 extracted 84% of the oxamyl residue found in tomato foliage and 126% of the oxamyl residue found in tomato fruit. These data indicated that the extraction method provides acceptable extraction efficiency for residue analysis.

Confirmatory method

Confirmation of results was obtained using secondary LC-MS/MS ion transitions collected at the same time as the quantitative transitions. The recovery data obtained using the confirmatory procedure are summarised in Table 28.

Table 28 Validation data for analytical methods for the determination of oxamyl in food of plant origin

Matrix	Fortification level (mg/kg)[a,b]	Number of tests	Average recovery (%)	Standard deviation	% Relative standard deviation	Reference
Oxamyl (237 → 90)						
Grapes	0.01 0.10	5 5 Total = 10	96 99 Mean = 98	1.0 1.1 Mean = 1.1	1.0 1.1 Mean = 1.1	DuPont-41730
Tomatoes	0.01 0.10	5 5 Total = 10	101 102 Mean = 102	1.0 1.9 Mean = 1.5	1.0 1.9 Mean = 1.5	
Avocado	0.01 0.10	5 5 Total = 10	85 99 Mean = 92	11 2.3 Mean = 6.7	13 2.3 Mean = 7.7	
Wheat	0.01 0.10	5 5 Total = 10	99 102 Mean = 101	1.8 0.90 Mean = 1.4	1.8 0.9 Mean = 1.4	
Tobacco	0.01 0.10	5 5 Total = 10	92 95 Mean = 94	3.5 2.2 Mean = 2.9	3.8 2.3 Mean = 3.1	

[a] Fortifications were performed with analyte reference standard solutions
[b] Limit of quantification, defined by a signal-to-noise ratio of approximately 10 and the lowest validated fortification level

Data from two ion transitions were also collected during the method independent laboratory validation, DuPont-41873.

Overall suitability for enforcement purposes

This method is suitable for enforcement of the MRL for oxamyl in the watery, acidic, oily, dry, and difficult to analyse EU crop groups. The instrumentation required to perform both the analysis and confirmatory methods is available in most well equipped analytical laboratories. No toxic or hazardous reagents are required to prepare the samples, and all of the sample preparation equipment is commercially available. The method does not require the use of untreated commodity to correct for recoveries.

Conclusions

The residue method for the determination of oxamyl residues in the watery, acidic, oily, dry, and difficult to analyse EU crop groups involves simple extraction, clean-up, and analytical determination by HPLC/MS/MS detection. A limit of quantification of 0.01 mg/kg can be achieved consistently all of the stipulated EU crop groups.

RMS comments and conclusion: the methods presented are considered acceptable.

B.5.2.2 Description of monitoring methods for determination of residues in commodities of animal origin

Studies submitted to the EU for the first time in this submission.

B.5.2.2/01

Reference: CA 4.2/03	**Report**	Henze, R.M., Klems, J.P. (2014b); Analytical method for the determination of oxamyl in liver, milk, eggs, fat and muscle using LC/MS/MS **DuPont Report No.:** DuPont-38597 **GLP:** No

B.5.2.2/02

Reference: CA 4.2/01	**Report**	Fiorito, B. (2014); Independent Laboratory Validation of DuPont-38597 "Analytical method for the determination of oxamyl in liver, milk, eggs, fat and muscle using HPLC/MS/MS" **DuPont Report No.:** DuPont-39679 **GLP:** Yes

Description of the method

Oxamyl is extracted from animal matrices in this method using a procedure based on the radio-validated extraction performed in AMR 1004-87, cited in Oxamyl RAR Vol.3 B7 . Following extraction, the sample is centrifuged at 3000 rpm for 10 minutes and the extract is decanted. Oily co-extracts are removed by vortexing the samples in the presence of 10 mL of hexane and centrifuging at 3000 rpm for 5 minutes. The hexane layer is then discarded. The extracts are cleaned up further through the addition of 0.25 g of SAX sorbent, and submitted for LC-MS/MS analysis.

Recovery findings

The fortification data reported in the method for oxamyl residues in animal tissues, milk, and eggs are summarised in Table 29. The average recovery specified in the decision-making criteria is 70–120%, with a standard deviation of ≤20%. Therefore, the recovery of this method is adequate for the purposes of residue data collection and enforcement of MRLs.

Table 29 Validation data for analytical methods for the determination oxamyl in food of animal origin (fortification data)

Reference	Matrix	Number of tests	Fortification level (mg/kg)[a,b]	Average recovery (%)	Standard deviation	% Relative standard deviation
Oxamyl (237 → 72)						
DuPont-38597	Meat (Muscle)	5 5 Total = 10	0.010 0.10	103 89 Mean = 96	6.1 1.3 Mean = 3.7	6.3 1.2 Mean = 3.8
	Fat	5 5 Total = 10	0.01 0.10	88 82 Mean = 85	14 5.0 Mean = 9.5	12 4.1 Mean = 8.1
	Liver	5 5 Total = 10	0.010 0.10	102 86 Mean = 94	4.1 3.3 Mean = 3.7	4.2 2.8 Mean = 3.5
	Cream	5 5 Total = 10	0.010 0.10	104 92 Mean = 98	4.2 6.8 Mean = 5.5	4.4 6.3 Mean = 5.4
	Whole Milk	5 5 Total = 10	0.010 0.10	103 95 Mean = 99	6.6 9.5 Mean = 8.1	6.8 9.0 Mean = 7.9
	Low Fat Milk	5 5 Total = 10	0.010 0.10	102 96 Mean = 99	5.2 7.9 Mean = 6.6	5.3 7.6 Mean = 6.5
	Eggs	5 5 Total = 10	0.010 0.10	102 90 Mean = 96	7.3 4.9 Mean = 6.1	7.4 4.4 Mean =5.9

[a] Fortifications were performed with analyte reference standard solutions
[b] Limit of quantification, defined by a signal-to-noise ratio of approximately 10 and the lowest validated fortification level

Linearity

Good linearity was observed in the range of 0.05 to 10 ng/mL for oxamyl. All calibration standards were prepared in solvent.

Specificity

Analysis of control samples resulted in no detectable apparent residues oxamyl. The response in the areas of the oxamyl peaks always corresponded to less than 20% of the limit of determination. It can therefore be concluded that few, if any, apparent residues or false positive values would arise.

Limit of quantification

The limit of quantification of the method for milk, meat (muscle), liver, fat, and eggs is 0.010 mg/kg, which is sufficient to detect oxamyl at the proposed MRLs.

Repeatability

The same analyst obtained these recovery data over the course of multiple days. Relative standard deviations of less than 20% were obtained for fortifications made at the quantification limit for each matrix, as well as at higher levels. Therefore, the repeatability of this method is adequate for the purposes of residue data collection and enforcement of MRLs.

Reproducibility

An independent laboratory validation of the method described in DuPont-38597 was conducted, and the results are presented in Table 30. The primary method is identical for all matrices listed under Section 4.3 of SANCO/825/00 rev 8.1; therefore, it is sufficient to perform the ILV with two of these matrices.

Table 30 Validation data for analytical methods for the determination of oxamyl in food of animal origin (independent laboratory method validation data)

Reference	Matrix	Number of tests	Fortification level (mg/kg)[a,b]	Average recovery (%)	Standard deviation	% Relative standard deviation
Oxamyl (237 → 72)						
DuPont-39679	Meat (Muscle)	5 5 Total = 10	0.010 0.10	81 84 Mean = 83	4.4 4.8 Mean = 4.6	5 6 Mean = 6
	Eggs	5 5 Total = 10	0.010 0.10	70 80 Mean = 75	4.0 7.7 Mean = 5.9	6 10 Mean = 8

[a] Fortifications were performed with analyte reference standard solutions
[b] Limit of quantification, defined by a signal-to-noise ratio of approximately 10 and the lowest validated fortification level

Extraction efficiency

The residue extraction method described in this report utilizes the same solvents as the metabolism method outlined in AMR 1004-87 (cited in Oxamyl RAR Vol. 3 B7 AS), at a similar solvent to sample ratio of 6 to 1, with two minor exceptions. First, the methanol was acidified with formic acid to increase the stability of oxamyl and denature any co-extracted proteins. Second, methylene chloride was eliminated from the solvent system. Methylene chloride is a Category 2 carcinogen, and its use is forbidden under section 2.4 of SANCO/825/00 rev. 8.1. A separate study of ^{14}C metabolism in lactating goats, AMR 2578-92 (cited in Oxamyl RAR Vol. 3 B7 AS), showed that less than 2% of the radioactivity was extracted by methylene chloride. Therefore, this difference is not expected to negatively affect extraction efficiency.

Confirmatory method

Confirmation of results was obtained using secondary LC-MS/MS ion transitions collected at the same time as the quantitative transitions. The recovery data obtained using the confirmatory procedure are summarised in Table 31.

Table 31 Validation data for analytical methods for the determination oxamyl in food of animal origin (confirmatory data)

Reference	Matrix	Number of tests	Fortification level (mg/kg)[a,b]	Average recovery (%)	Standard deviation	% Relative standard deviation
Oxamyl (237 → 90)						
DuPont-38597	Meat (Muscle)	5 5 Total = 10	0.010 0.10	99 87 Mean = 93	3.1 2.2 Mean = 2.7	3.1 2.5 Mean =2.8
	Fat	5 5 Total = 10	0.01 0.10	86 80 Mean = 83	6.3 8.8 Mean = 7.6	7.3 11 Mean = 9.2
	Liver	5 5 Total = 10	0.010 0.10	105 90 Mean = 98	6.9 6.5 Mean = 6.7	6.6 7.2 Mean = 6.9
	Cream	5 5 Total = 10	0.010 0.10	98 89 Mean = 94	6.5 6.3 Mean = 6.4	6.6 7.1 Mean = 6.9
	Whole Milk	5 5 Total = 10	0.010 0.10	104 99 Mean = 102	6.2 3.4 Mean = 4.8	6.0 3.4 Mean = 4.7
	Low Fat Milk	5 5 Total = 10	0.010 0.10	104 99 Mean = 102	7.7 3.2 Mean = 5.5	7.4 3.2 Mean = 5.3
	Eggs	5 5 Total = 10	0.010 0.10	97 96 Mean = 97	8.4 7.8 Mean = 8.1	8.7 8.1 Mean = 8.4

[a] Fortifications were performed with analyte reference standard solutions
[b] Limit of quantification, defined by a signal-to-noise ratio of approximately 10 and the lowest validated fortification level

Confirmatory data were also generated during the method independent laboratory validation, DuPont-39679.

Overall suitability for enforcement purposes

This method is suitable for enforcement of oxamyl MRLs in milk, meat (muscle), liver, fat, and egg. This procedure can be applied to other tissues not tested during method validation. The instrumentation required to perform the analysis is available in most well equipped analytical laboratories. No toxic or hazardous reagents are required to prepare the samples, and all of the sample preparation equipment is commercially available.

Conclusions

The residue method for the determination of oxamyl in milk, meat (muscle), liver, fat, and egg involves simple extraction, clean-up, and analytical determination by HPLC/MS/MS detection. A limit of quantification of 0.010 mg/kg can be achieved consistently for all tissues and is sufficient to determine oxamyl residues at the proposed MRLs provided in the Oxamyl RAR Vol. 3 B7 AS .

Studies submitted to the EU for the first time in this submission.

B.5.2.2/03

Reference: CA 4.2/07	**Report**	Lissemore, L., Harris, A., Patterson, C. (2014a); QuEChERS multiresidue method trials for DPX-D1410, DPX-X1179 and DPX-Q8U80 in animal tissues **DuPont Report No.:** DuPont-41763 **GLP:** Yes

B.5.2.2/04

Reference: CA 4.2/09	**Report**	Schernikau, N., Colorado, C.S. (2015); Independent laboratory validation of DuPont-41730 and DuPont-41763, "Analytical method for the determination of oxamyl (DPX-D1410) and methomyl (DPX-X1179) in crop and animal matrices by LC/ESI-MS/MS" **DuPont Report No.:** DuPont-41873 **GLP:** Yes

Description of the method

The QuECHERS MRM was validated for the analysis of oxamyl residues in milk, egg, muscle, liver, and fat. Two grams of a given animal tissue matrix were weighed into a 50 mL polypropylene centrifuge tube. To each sample, 100 µL of the internal standard Carbofuran-d3 was added, and the samples were fortified with oxamyl. Ten millilitres of water and 10 mL of 1% acetic acid in acetonitrile were added to the samples along with 1 g of anhydrous sodium acetate. The samples were then vortexed for 20 seconds. After the samples had been vortexed, 4 g of anhydrous magnesium sulphate were added to the sample, and the samples were placed on a genogrinder at 1700 rpm for 1 minute. The samples were then centrifuged at 3600 rpm for 5 minutes. Following centrifugation, 5.0 mL of the supernatant was pipetted into a 15 mL polypropylene centrifuge tube and blown down to between 0.2 and 0.3 mL under a stream of nitrogen. This sample was then sequentially diluted with methanol, ammonium acetate, and a 1:1 solution of 0.1M ammonium acetate/methanol. The samples were then syringe filtered into amber vial for LC-MS/MS analysis. Quantitation and confirmation signals for oxamyl were generated from its $[M+NH_4]^+$ ion, 237 m/z. Quantitation of oxamyl was based on the signal arising from the 237 → 72 ion transition. Confirmation was based on the signal arising from the 237 → 90 ion transition.

Recovery findings

The fortification data reported in the method proposed for monitoring oxamyl residues in food of animal origin are summarised in Table 32. The average recovery specified in the decision-making criteria is 70 to 120%, with a standard deviation of ≤20%. Therefore, the recovery of this method is adequate for the purposes of residue data collection and enforcement of MRLs.

Table 32 Validation data for analytical methods for the determination of oxamyl in food of animal origin

Matrix	Fortification level (mg/kg)[a,b]	Number of tests	Average recovery (%)	Standard deviation	% Relative standard deviation	Reference
Oxamyl (237 → 72)						
Milk	0.01 0.10	5 5 Total = 10	100 106 Mean = 103	2.3 1.3 Mean = 1.8	2.3 1.2 Mean = 1.8	DuPont-41763
Eggs	0.01 0.10	5 5 Total = 10	97 100 Mean = 99	1.3 2.3 Mean = 1.8	1.3 2.3 Mean = 1.8	
Muscle	0.01 0.10	5 5 Total = 10	95 105 Mean = 100	1.6 2.5 Mean = 2.1	1.7 2.4 Mean = 2.1	
Liver	0.01 0.10	5 5 Total = 10	83 97 Mean = 90	6.6 2.5 Mean = 4.6	8.0 2.6 Mean = 5.3	
Fat	0.01 0.10	5 5 Total = 10	84 90 Mean = 87	6.2 3.2 Mean = 4.7	7.4 3.6 Mean = 5.5	

[a] Fortifications were performed with analyte reference standard solutions
[b] Limit of quantification, defined by a signal-to-noise ratio of approximately 10 and the lowest validated fortification level

Linearity

Good linearity was observed in the range of 0.05 to 4.0 ng/mL for oxamyl.

Specificity

The limit of quantification of the method proposed for monitoring oxamyl residues is 0.010 mg/kg for all matrices tested. Analysis of control samples resulted in no detectable apparent residues of oxamyl; the response in the area of the oxamyl peak always corresponded to less than 20% of the limit of determination. It can therefore be concluded that few, if any, apparent residues or false positive values would arise.

Limit of quantification

The limit of quantification of the method for all of the tissues studied is 0.010 mg/kg.

Repeatability

Repeatability of the method is addressed by the data in Table 32. Relative standard deviations of less than 20% were obtained for fortifications made at the quantification limit for each matrix, as well as at higher levels. Therefore, the repeatability of this method is adequate for the purposes of residue data collection and enforcement of MRLs.

Reproducibility

An independent laboratory validation of DuPont-41730 was conducted, and the results are presented in Table 33. The primary method is identical for all matrices, so it is sufficient to perform the ILV for two of these commodities. This method was validated on three matrices: muscle, milk, and egg.

Table 33 Validation data for analytical methods for the determination of oxamyl in food of animal origin

Matrix	Fortification level (mg/kg)[a,b]	Number of tests	Average recovery (%)	Standard deviation	% Relative standard deviation	Reference
Oxamyl (237 → 72)						
Muscle	0.01 0.10	5 5 Total = 10	86 100 Mean = 93	3.6 3.3 Mean = 3.5	4.2 3.3 Mean = 3.8	DuPont-41873
Milk	0.01 0.10	5 5 Total = 10	95 95 Mean = 95	1.9 1.1 Mean = 1.5	2.0 1.2 Mean = 1.6	
Eggs	0.01 0.10	5 5 Total = 10	86 94 Mean = 95	1.9 1.1 Mean = 1.5	1.2 1.8 Mean = 1.5	

[a] Fortifications were performed with analyte reference standard solutions
[b] Limit of quantification, defined by a signal-to-noise ratio of approximately 10 and the lowest validated fortification level

Extraction efficiency

While no incurred, radiolabelled material was available to test the extraction efficiency of DuPont-41873 using animal tissues, the extraction efficiency of this method was investigated in DuPont-44316 by extracting tomato samples with incurred, radiolabelled residues. Tomato foliage and fruit were extracted using the method found in DuPont-41730 and compared to the metabolism method. On average, DuPont-41730 extracted 84% of the oxamyl residue found in tomato foliage and 126% of the oxamyl residue found in tomato fruit indicating that this method provides acceptable extraction efficiency for residue analysis.

Confirmatory method

Confirmation of results was obtained using secondary LC-MS/MS ion transitions collected at the same time as the quantitative transitions. The recovery data obtained using the confirmatory procedure are summarised in Table 34.

Table 34 Validation data for analytical methods for the determination of oxamyl in food of animal origin

Matrix	Fortification level (mg/kg)[a,b]	Number of tests	Average recovery (%)	Standard deviation	% Relative standard deviation	Reference
Oxamyl (237 → 90)						
Milk	0.01 0.10	5 5 Total = 10	100 101 Mean = 101	0.73 2.2 Mean = 1.5	0.73 2.2 Mean = 1.5	DuPont-41763
Eggs	0.01 0.10	5 5 Total = 10	93 99 Mean = 96	2.2 1.3 Mean = 1.8	2.4 1.3 Mean = 1.9	
Muscle	0.01 0.10	5 5 Total = 10	95 101 Mean = 98	2.3 2.0 Mean = 2.2	2.4 2.0 Mean = 2.2	
Liver	0.01 0.10	5 5 Total = 10	86 93 Mean = 90	5.0 1.5 Mean = 3.3	5.8 1.6 Mean = 3.7	
Fat	0.01 0.10	5 5 Total = 10	88 87 Mean = 88	7.2 2.3 Mean = 4.3	8.2 2.6 Mean = 5.4	

[a] Fortifications were performed with analyte reference standard solutions
[b] Limit of quantification, defined by a signal-to-noise ratio of approximately 10 and the lowest validated fortification level

Data from two ion transitions was also collected during the method independent laboratory validation, DuPont-41873.

Overall suitability for enforcement purposes

This method is suitable for enforcement of oxamyl MRLs in milk, meat (muscle), liver, fat, and egg. This procedure can be applied to other tissues not tested during method validation. The instrumentation required to perform the analysis is available in most well equipped analytical laboratories. No toxic or hazardous reagents are required to prepare the samples, and all of the sample preparation equipment is commercially available.

Conclusions

The residue method for the determination of oxamyl in milk, meat (muscle), liver, fat, and egg involves simple extraction, clean-up, and analytical determination by HPLC/MS/MS detection. A limit of quantification of 0.010 mg/kg can be achieved consistently for all tissues and is sufficient to oxamyl at the proposed MRLs provided in the Oxamyl RAR Vol. 3 B7 AS .

RMS comments and conclusion: the methods presented are considered acceptable

B.5.2.3 Description of monitoring methods for determination of residues soil

Enforcement methods suitable for the European Union region

Studies submitted to the EU for the first time in this submission.

B.5.2.3/01

Reference: CA 4.2/02	**Report**	Henze, R.M., Klems, J.P. (2014a); Analytical method for the determination of oxamyl in soil using LC/MS/MS **DuPont Report No.:** DuPont-38689 **GLP:** No

Description of the method

Oxamyl residues were extracted from soil samples using a solution of aqueous formic acid and methanol. The sample extracts were filtered and diluted by a factor of ten before being submitted for LC-MS/MS analysis. The analysis was performed in positive mode (ESI +) and the transition were 237>72 amu (quantifier) and 237>90 amu

Recovery findings

The fortification data reported in the method proposed for monitoring oxamyl residues in soil are summarised in Table 35.

Table 35 Validation data for analytical methods for the determination of oxamyl in soil

Reference	Matrix	Number of tests	Fortification level (mg/kg)[a,b]	Average recovery (%)	Standard deviation	% Relative standard deviation
Oxamyl (237 → 72)						
DuPont-38689	Speyer Soil	5 5 Total = 10	0.0010 0.010	96 96 Mean = 96	7.3 2.8 Mean = 5.1	7.6 2.9 Mean = 5.3
	Nambsheim Soil	5 5 Total = 10	0.0010 0.010	89 95 Mean = 92	6.0 7.1 Mean = 6.6	6.7 7.5 Mean = 7.1
	Drummer Soil	5 5 Total = 10	0.0010 0.010	82 86 Mean = 84	2.9 2.8 Mean = 2.9	3.5 3.3 Mean = 3.4

[a] Fortifications were performed with analyte reference standard solutions
[b] Limit of quantification, defined by a signal-to-noise ratio of approximately 10 and the lowest validated fortification level

Linearity

Good linearity was observed in the range of 0.025 to 10.0 ng/mL for oxamyl. All calculations were performed using standards prepared in solvent.

Specificity

The limit of determination of the method proposed for monitoring oxamyl residues in soil is 0.24 µg/kg. Analysis of control samples of different soil types resulted in no detectable apparent residues of oxamyl; the response in the area of the oxamyl peak always corresponded to less than 20% of the limit of determination. It can therefore be concluded that few, if any, apparent residues or blank values would arise.

Limit of quantification

The limit of quantification of the method for oxamyl residues in soil is 1.0 µg/kg.

Repeatability

Repeatability of the method is addressed by the data in Table 35. The same analyst obtained these recovery data over the course of two days per matrix. Relative standard deviations of less than 20% were obtained for fortifications made at the quantification limit for each matrix, as well as at higher levels. Therefore, the repeatability of this method would seem to be adequate for the purposes of detecting oxamyl residues in soil.

Reproducibility

An independent laboratory validation of DuPont-38689 was not conducted. An independent laboratory validation of soil methods is not required.

Extraction efficiency

The extraction solvents used in DuPont-38689 have been validated using incurred ^{14}C labelled residues in multiple studies, including AMR 1851-90, (cited in Oxamyl RAR Vol. 3 B7 AS) . During AMR 1851-90, residues were extracted using three 1:1 methanol/water extractions followed by two extractions methanol extractions. The final extract is composed of 70% methanol and 30% water. The extraction method used in DuPont-38689 uses two extractions with a 9:1 methanol to water solution. The higher level of methanol in the residue method extraction is not expected to affect the extraction efficiency of oxamyl as it has very high solubility in methanol.

Confirmatory method

Confirmation of results was obtained using secondary LC-MS/MS ion transitions collected at the same time as the quantitative transitions. The recovery data obtained using the confirmatory procedure are summarised in Table 36.

Table 36 Validation data for analytical methods for the determination of oxamyl in soil

Reference	Matrix	Number of tests	Fortification level (mg/kg)[a,b]	Average recovery (%)	Standard deviation	% Relative standard deviation
Oxamyl (237 → 72)						
DuPont-38689	Speyer Soil	5 5 Total = 10	0.0010 0.010	96 97 Mean = 97	7.4 2.2 Mean = 4.8	7.7 2.3 Mean = 5.0
	Nambsheim Soil	5 5 Total = 10	0.0010 0.010	89 93 Mean = 91	2.9 2.5 Mean = 2.7	3.3 2.7 Mean = 3.0
	Drummer Soil	5 5 Total = 10	0.0010 0.010	78 86 Mean = 82	4.4 2.0 Mean = 3.2	5.7 2.3 Mean = 4.0

[a] Fortifications were performed with analyte reference standard solutions
[b] Limit of quantification, defined by a signal-to-noise ratio of approximately 10 and the lowest validated fortification level

Overall suitability for regulatory purposes

This method is suitable for use by regulatory agencies to detect oxamyl residues in soil. The instrumentation required to perform both the analysis and confirmatory methods is available in most well equipped analytical laboratories. No toxic or hazardous reagents are required to prepare the samples, and all of the sample preparation equipment is commercially available. The method does not require the use of untreated commodity to correct for recoveries.

RMS comments and conclusion: the method presented is considered acceptable

B.5.2.4 Description of monitoring methods for determination of residues in water

Enforcement methods suitable for the European Union region

Study submitted in the EU Dossier in 2001 and included in the first EU approval review.

B.5.2.4/01

Reference: --	**Report**	Hill, S.J., Maliszewski, P.P., Stry, J.J. (2001); Analytical method for the determination of oxamyl and its oxime metabolite in water using LC/MS/MS analysis - 0.1 ppb LOQ **DuPont Report No.:** DuPont-5677 **GLP:** No

Principle of the method:

Water (100ml) is cleaned up by eluting through an SPE SAX cartridge(1g) in sequence with an SPE Oasis HLB cartridge (1g). The oxamyl is retained on the Oasis HLB cartridge. Prior to using the SPE clean-up cartridges they are both conditioned with methanol and water respectively making sure that they never go to dryness. The Oasis cartridge is washed with a solution of water:methanol (70:30) prior to the oxamy residue being eluted with a solution of methanol and water (50:50)[12ml]. Acetic acid (10 □l) is added to the methanol/water eluate, the eluate is mixed, filtered and analysed using HPLC using MS/MS. An analytical column (Phenomenex Luna) with a Phenyl-Hexyl stationary phase was used to analyse the samples while using a gradient mobile phase of methanol/water. Identification and confirmation was carried out by performing MS/MS on the parent Oxamyl mass ion and by monitoring for the presence and ratios of the two daughter ions at 72 and 90 m/z.

Validation data:

The following recovery data was carried out to validate the method for the analysis of Oxamyl residues in water and is presented in Table 37 below.

Table 37 Recovery of Oxamyl residues from fortified water samples (%)

Water source	Water type	Fortification level (μg/l)	Mean Recovery (%)	Range (%)	% RSD (n)
Kembelsville, PA	Ground water	0.1	95.8	92 - 100	3.5 (5)
		1.0	90.8	85- 98	5.6 (5)
S.H.R.C. Municipal, DE.	Drinking water.	0.1	89	86- 92	2.5 (5)
		1.0	83.2	80- 86	3.1 (5)
Lums Pond, DE	Surface water	0.1	95.25	91- 100	4.2 (5)
		1.0	90.6	85- 94	4.2 (5)
St. Jones River, DE.	Surface water	0.1	101.6	92- 107	6.1 (5)
		1.0	93.2	83- 103	8.7 (5)
Brandywine River, DE.	Surface water	0.1	100.8	92- 109	8.2 (5)
		1.0	97.2	86- 105	7.9 (5)

Relevant chromatograms were provided for the analysis of Oxamyl residues in different types of water.

Findings:

Recovery studies, as outlined above in Table 37. are acceptable. Limit of Quantitation: A limit of quantitation of 0.1 μg/litre has been validated.

Conclusions:

The analytical method as presented has been validated for the analysis of Oxamyl residues in ground, drinking and surface water. The method is acceptable.

RMS comments and conclusion

The monitoring method for determination of residues in water, DuPont-5677, originally submitted under EU Rev8 Point IIA 4.2.3.1, was conducted under guideline Directive 91/414/EEC. A review of this study indicates it partially meets the current guideline SANOC/825/00 REV. 8.1; deviations include lack of sufficient confirmatory data. However, when submitted in conjunction with DuPont-5677, Supplement No. 1 and DuPont-5677, Supplement No. 2, summarized below, it adequately completes the understanding of methods for risk assessment.

Study submitted in the EU Dossier in 2001 and included in the first EU approval review.

B.5.2.4/02

Reference: --	**Report**	Connolly, P. (2001); Independent laboratory validation of method number DuPont-5677, analytical method for the determination of oxamyl in water using LC/MS/MS **DuPont Report No.:** DuPont-6157 **GLP:** Yes

Principle of the method:

The analytical method is that reported in section B.5.3.2.1. above which was independently validated by "Centre Analytical Laboratories, Inc., 3048 Research Drive, State College, PA 16801, USA".

Validation data:

The following validation data was generated for the analysis of Oxamyl residues in water and is presented in Table 38. The data was generated for ground, surface and drinking water. Analysis was by LC/MS/MS where quantitation and identification was based on the parent ion transition to the product ions 72 and 90 m/z and confirmation of the ion ratios of the 72 and 90 m/z product ions.

Table 38 Recovery of Oxamyl residues from fortified water samples (%)

Water source	Water type	Fortification level (µg/l)	Mean Recovery (%)	Range (%)	% RSD (n)
Connolly Well, Centre Hall,	Ground water	0.1	73.6	70- 76	3.1 (5)
		0.2	84.2	77- 101	11.4 (5)
Deer Park Spring water (purchased	Drinking water.	0.1	84.8	79- 93	7 (5)
		0.2	93	81- 103	8.7 (5)
Whipple Dam Resevoir, Centre County,	Surface water	0.1	86.2	77- 93	7.3 (5)
		0.2	90	89- 92	1.6 (5)

Sample chromatograms were submitted as part of the study report.

Conclusion:

The above study independently validates that the analytical method, as reported in DuPont report 5677, is capable of determining Oxamyl residues in water at 0.1 µg/L.

The independent laboratory validation of DuPont-5677, DuPont-6157, originally submitted under EU Rev8 Point IIA 4.2.3.1, was conducted under guidelines SANCO/825/00 rev. 6 (2000) and OPPTS 850.7100 (Draft 1996). A review of this study indicates that it fully meets the current guideline: SANCO/825/00 rev 8.1 (2011).

For completeness the original method report (DuPont-5677), the independent laboratory validation (DuPont-6157) and the updated supplement reports are summarised below.

Studies submitted to the EU for the first time in this submission.

B.5.2.4/03

Reference: CA 4.2/04	**Report**	Klems, J.P. (2014a); Analytical method for the determination of oxamyl in water using LC/MS/MS **DuPont Report No.:** DuPont-5677, Supplement No. 1 **GLP:** No

B.5.2.4/04

Reference: CA 4.2/05	**Report**	Klems, J.P. (2014b); Analytical method for the determination of oxamyl in blood using LC/MS/MS **DuPont Report No.:** DuPont-5677, Supplement No. 2 **GLP:** No

Description of the method

Oxamyl was isolated from water samples by filtering 100 mL of water through a SAX solid phase extraction (SPE) cartridge and concentrating the oxamyl on an Oasis HLB SPE cartridge. Following a wash step, oxamyl was eluted in 12 mL of 50:50 methanol: water (v/v). The eluent was evaporated to approximately 7 mL and the volume adjusted to 10 mL using water. Oxamyl was separated from co-extracts by reversed-phase liquid chromatography and was detected by MS/MS. DuPont-5677, Supplement No. 1 updates the method to include validation on characterized surface water. DuPont-5677, Supplement No. 2 updates the method to include confirmatory ion data generated from the original data. This confirmatory data can be found in Table 41.

Recovery findings

The fortification data reported in the method proposed for monitoring oxamyl residues in water are summarised in Table 39.

Table 39 Validation data for analytical methods for the determination of oxamyl in water

Reference	Matrix	Number of tests	Fortification level (mg/L)[a,b]	Average recovery (%)	Standard deviation	% Relative standard deviation
Oxamyl (220 → 72)						
DuPont-5677, DuPont-5677 Supplement No. 1	Ground Water	5 5 Total = 10	0.00010 0.0010	89 83 Mean = 86	3 2 Mean = 3	3 3 Mean = 3
	Drinking Water	5 5 Total = 10	0.00010 0.0010	96 91 Mean = 94	4 6 Mean = 5	4 6 Mean = 7
	Surface Water A	5 5 Total = 10	0.00010 0.0010	95 91 Mean = 93	4 4 Mean = 4	4 4 Mean = 4
	Surface Water B	5 5 Total = 10	0.00010 0.0010	102 93 Mean = 98	6 8 Mean = 7	6 9 Mean = 8
	Surface Water C	5 5 Total = 10	0.00010 0.0010	101 97 Mean = 99	8 8 Mean = 8	8 8 Mean = 8
	Surface Water D	5 5 Total = 10	0.00010 0.0010	89 94 Mean = 92	5 4 Mean = 5	6 4 Mean = 5

[a] Fortifications were performed with analyte reference standard solutions

[b] Limit of quantification, defined by a signal-to-noise ratio of approximately 10 and the lowest validated fortification level

Linearity

Good linearity was observed in the range of 0.50 to 12.0 ng/mL for oxamyl. All calculations were performed using standards prepared in solvent.

Specificity

The limit of determination of the method proposed for monitoring oxamyl residues in water is 0.03 µg/L. Analysis of control samples of six different water sources, including four surface water sites, resulted in no detectable apparent residues of oxamyl; the response in the area of the oxamyl peak always corresponded to less than 20% of the limit of determination. It can therefore be concluded that few, if any, apparent residues or false positive values would arise.

Limit of quantification

The limit of quantification of the method for water is 0.0001 mg/L (0.1 µg/L). The EU guidelines for drinking water methods specify that methods must be capable of measuring levels at or above 0.1 µg/L, which is the maximum allowable level of any crop protection chemical in drinking water.

Repeatability

Repeatability of the method is addressed by the data in Table 39. Multiple analysts obtained these recovery data over the course of two days per matrix. Relative standard deviations of less than 20% were obtained for fortifications made at the quantification limit for each matrix, as well as at higher levels. Therefore, the repeatability of this method would seem to be adequate for the purposes of detecting oxamyl residues in water.

Reproducibility

An independent laboratory validation of DuPont-5677 was conducted and the results are summarized in Table 40.

Table 40 Validation data for analytical methods for the determination of oxamyl in water

Reference	Matrix	Number of tests	Fortification level (mg/kg)[a,b]	Average recovery (%)	Standard deviation	% Relative standard deviation
Oxamyl (220 → 72)						
DuPont-6157	Ground Water	5 5 Total = 10	0.00010 0.00020	74 84 Mean = 79	2.3 9.6 Mean = 6	3 11 Mean = 7
	Surface Water	5 4 Total = 9	0.00010 0.00020	86 90 Mean = 88	6.3 1.4 Mean = 3.9	7 2 Mean = 5
	Drinking Water	5 5 Total = 10	0.00010 0.00020	85 93 Mean = 89	5.9 8.1 Mean = 7.0	7 9 Mean = 8

[a] Fortifications were performed with analyte reference standard solutions
[b] Limit of quantification, defined by a signal-to-noise ratio of approximately 10 and the lowest validated fortification level

Confirmatory method

Confirmation of results was obtained using secondary LC-MS/MS ion transitions collected at the same time as the quantitative transitions. The recovery data obtained using the confirmatory procedure are summarised in Table 41.

Table 41 Validation data for analytical methods for the determination of oxamyl in water

Reference	Matrix	Number of tests	Fortification level (mg/kg)[a,b]	Average recovery (%)	Standard deviation	% Relative standard deviation
Oxamyl (220 → 90)						
DuPont-5677 Supplement No. 2	Ground Water	5 5 Total = 10	0.00010 0.0010	92 85 Mean = 89	5.7 3.5 Mean = 4.6	6.2 4.1 Mean = 5.2
	Drinking Water	5 5 Total = 10	0.00010 0.0010	92 90 Mean = 91	6.6 5.4 Mean = 6	7.2 6.0 Mean = 6.6
	Surface Water A	5 5 Total = 10	0.00010 0.0010	102 98 Mean = 100	8.1 6.8 Mean = 7.5	7.9 6.9 Mean = 7.4
	Surface Water B	5 5 Total = 10	0.00010 0.0010	99 92 Mean = 96	7.3 4.4 Mean = 5.9	7.4 4.8 Mean = 6.1
	Surface Water C	5 5 Total = 10	0.00010 0.0010	102 95 Mean = 99	13 10 Mean = 12	13 10 Mean = 12

[a] Fortifications were performed with analyte reference standard solutions
[b] Limit of quantification, defined by a signal-to-noise ratio of approximately 10 and the lowest validated fortification level

Overall suitability for enforcement purposes

This method is suitable for use by regulatory agencies to detect oxamyl residues in surface water, ground water, and drinking water. The instrumentation required to perform both the analysis and confirmatory methods is available in most well equipped analytical laboratories. No toxic or hazardous reagents are required to prepare the samples, and all of the sample preparation equipment is commercially available. The method does not require the use of untreated commodity to correct for recoveries.

RMS comments and conclusion
IT: the method presented is considered acceptable

B.5.2.5 Description of monitoring methods for determination of residues air

Study submitted in the EU Dossier in 2001 and included in the first EU approval review.

B.5.2.5/01

Reference: CA 4.2	**Report**	Bacher, R. (2000); Development and validation of an analytical method for the determination of oxamyl and methomyl in air **DuPont Report No.:** DuPont-4564 **GLP:** Yes

Principle of the Method:

The method of analysis uses air sampling cartridges which are filled with two portions of an adsorbent (Supelpak 20E XAD-2 porous polymer) which are held in place by 3 glass wool plugs. Air is sucked through these sampling tubes at a flow rate of 1 – 2 litres/minute for a period of up to 6 hours. The air flow is determined using a flowmeter.

Following the sampling process the adsorbent material is extracted with acetonitrile x3 and the combined extracts are concentrated to low volume (circa 0.5 ml). Water is added to adjust the volume to 1ml and the extract is analysed using a Phenomenex Luna RP-C18 HPLC column and with a HPLC/MS fitted with an atmospheric pressure chemical ionisation (APCI) source. The characteristic ions used are for Oxamyl 237 m/z (M+ H2O)+ for Oxamyl oxime 163 m/z (M+ H)+ for Methomyl 163 m/z (M+ H)+ for Methomyl oxime 106 m/z (M+H)+ Recovery studies were carried out by fortifying the adsorbent material with Oxamyl and with Methomyl at levels corresponding to (A) 1.25 µg of Oxamyl and 3 µg of Methomyl and at (B) 0.125 µg of Oxamyl and 0.3 µg of Methomyl. For sampling volumes of 0.5 m3 of air these values correspond to target air concentrations of 0.25 and 2.5 µg/m3 for Oxamyl and 0.6 and 6.0 µg/m3 of Methomyl. Air at ambient temperatures and warm humid air were used during the course of the validation process.

Validation data: The results of the validation studies are presented below in

Table 42 Recovery data for Oxamyl and Methomyl residues from fortified adsorbent tubes

.	Oxamyl				Methomyl				
Sample type	**Fortificaion level (μ)**	**Mean air conc (μg/m³)**	**Mean recovery (%)**	**% RSD.**	**Fortificaion level (μ)**	**Mean air conc (μg/m³)**	**Mean recovery (%)**	**% RSD.**	**n**
Ambient air.	0.125	0.24	106%	2%	0.3	0.58	98	8%	5
	1.25	2.4	89%	14%	3.0	5.8	79	14%	5
	Overall recovery.		98%	13%	Overall recovery.		89%	16%	10
Warm Humid air.	0.125	0.24	106%	2%	0.3	0.58	98	8%	5
	1.25	2.4	89%	14%	3.0	5.8	79	14%	5
	Overall recovery.		98%	13%	Overall recovery.		89%	16%	10

At the fortification levels studied there was no breakthrough of the residue into the second sample trap. Copies of the sample chromatograms were presented as part of the study. Residues of Oxamyl and of Methomyl adsorbed onto XAD-2 resin were found to be stable for 5+ days when stored in a freezer in the dark.

Confirmation of the method can be carried out by hydrolysing Oxamyl and Methomyl residues to their respective oximes and the analysis of these oxime metabolites using LC/MS.

Conclusions:

The method provided is acceptable for the analysis of Oxamyl residues in air.

The monitoring method for determination of residues air, DuPont-4564, originally submitted under EU Rev8 Point IIA 4.2.4.1, was conducted under guidelines Directive 91/414/EEC and SANCO/825/00 rev. 6 (2000). A review of this study indicates that it fully meets the current guideline SANCO/825/00 rev 8.1.

B.5.2.6 Description of monitoring methods for determination of residues in body fluids and tissues

Study submitted to the EU for the first time in this submission.

B.5.2.6/01

Reference: CA 4.2/06	**Report**	Klems, J.P. (2014b); Analytical method for the determination of oxamyl in blood using LC/MS/MS **DuPont Report No.:** DuPont-38598, Revision No. 1 **GLP:** No

Description of the method

Oxamyl was extracted from blood samples using a 1:1 mixture of methanol and water. Following extraction, the samples are diluted and cleaned up using a dispersive SPE step. Aliquots of this sample are then transferred to LC vials for MS/MS analysis. The determination was performed in positive mode (ESI+) with the following transitions: 237>72 m/z(quantifier), 237>90 m/z.

Recovery findings

The residue results were within guideline requirements (70-120%, RSD ≤20%). A summary of the results is provided in Table 43.

Table 43 Validation data for analytical methods for the determination of oxamyl in body fluids

Reference	Matrix	Number of tests	Fortification level (mg/kg)[a,b]	Average recovery (%)	Standard deviation	% Relative standard deviation
Oxamyl (237 → 72)						
DuPont-38598, Revision No. 1	Blood	5 5 Total = 10	0.050 0.50	102 99 Mean = 101	14 15 Mean = 15	14 15 Mean = 15

[a] Fortifications were performed with analyte reference standard solutions
[b] Limit of quantification, defined by a signal-to-noise ratio of approximately 10 and the lowest validated fortification level

Linearity

Good linearity was observed in the range of 0.050 to 10.0 ng/mL for oxamyl. All calculations were performed using standards prepared in solvent. The linearity curve was constructed with six solutions.

Specificity

Analysis of control samples resulted in no detectable apparent residues of oxamyl. The response in the areas of the oxamyl peak always corresponded to less than 20% of the limit of determination. It can therefore be concluded that few, if any, apparent residues or false positive values would arise.

Limit of quantification

The limit of quantification of the method blood is 0.05 mg/L This method is capable of determining oxamyl residues in blood samples.

Repeatability

Repeatability of this method was demonstrated by the standard deviation of the recovery values given in Table 43. The relative standard deviation of recovery data obtained is within the guideline of ≤20%. This method is adequate for determining oxamyl residues in blood.

Reproducibility

An independent laboratory validation of DuPont-38598, Revision No. 1 was not conducted. Independent validations are not required for body fluid methods.

Extraction efficiency

The same solvents are used for this residue method as were used for the metabolism study AMR 1004-87 (cited in Oxamyl RAR Vol. 3 B7 AS and in this document). Since the extractions are similar, an extraction efficiency study is not needed.

Confirmatory method

Confirmation of results was obtained using secondary LC-MS/MS ion transitions collected at the same time as the quantitative transitions. The recovery data obtained using the confirmatory procedure are summarised in Table 44.

Table 44 Validation data for analytical methods for the determination of oxamyl in body fluids (confirmatory data)

Reference	Matrix	Number of tests	Fortification level (mg/kg)[a,b]	Average recovery (%)	Standard deviation	% Relative standard deviation
Oxamyl (237 → 72)						
DuPont-38598, Revision No. 1	Blood	5 5 Total = 10	0.050 0.50	102 99 Mean = 101	14 15 Mean = 15	14 15 Mean = 15

[a] Fortifications were performed with analyte reference standard solutions
[b] Limit of quantification, defined by a signal-to-noise ratio of approximately 10 and the lowest validated fortification level

Overall suitability for enforcement purposes

This method is suitable for enforcement of the MRL for oxamyl in body fluids. The instrumentation required to perform both the analysis and confirmatory methods is available in most well equipped analytical laboratories. No toxic or hazardous reagents are required to prepare the samples, and all of the sample preparation equipment is commercially available. The method does not require the use of untreated commodity to correct for recoveries.

Conclusion

The residue method for the determination of oxamyl and metabolites in body fluids involves simple extraction, clean-up, and analytical determination by HPLC/MS/MS detection. A limit of quantification of 0.050 mg/kg can be achieved consistently and is sufficient to detect oxamyl at the levels stipulated by SANCO/825/00 RV1.

RMS comments and conclusion: the method presented is considered acceptable

B.5.3 References relied on

List of information, tests and studies which are considered as relied upon by the RMS for the evaluation with a view to the approval of the active substance.

Studies marked in yellow are submitted for the first time.

Data protection will be claimed at MSs level.

Sorted by Annex Point

Data Requirement No., Reference No.	Author(s)	Year	Title Source Company Report No. GLP or GEP Status (where relevant) Published or not	Vertebrate study Y/N	Data Protection Y/N	Justification if data protection is claimed	Owner
B.5.1.1.1/01	Pandey, S., McNally, M.E.P.	2015f	Determination of oxamyl (DPX-D1410) in technical grade oxamyl and end-use products DuPont-42001 GLP: No Published: No	N	N		DuPont
B.5.1.1.1/02	Pandey, S.	2015b	Validation of the analytical method for determination of oxamyl (DPX- D1410) in technical grade oxamyl and oxamyl end-use products by reverse phase high performance liquid chromatography (RPLC) and ultra performance liquid chromatography (UPLC) Syngene DuPont-36605 and DuPont-36605 Confidential attachment GLP: Yes Published: No	N	Y	The study is necessary for the regulatory decision, conducted according to GLP and has not previously been protected or submitted	DuPont
B.5.1.2.1/01	Nanita, S.C.	2009	Analytical method for the determination of oxamyl and its oxime metabolite in soil using LC/MS analysis DuPont Experimental Station DuPont-2392, Revision No. 1 GLP: No Published: No	N	N		DuPont

Data Requirement No., Reference No.	Author(s)	Year	Title Source Company Report No. GLP or GEP Status (where relevant) Published or not	Vertebrate study Y/N	Data Protection Y/N	Justification if data protection is claimed	Owner
B.5.1.2.1/02	Stry, J.J.	2000	Independent laboratory validation of method number DuPont-2392, "Analytical method for the determination of oxamyl and its oxime metabolite in soil using LC/MS analysis" DuPont Experimental Station DuPont-3738 GLP: Yes Published: No	N	Y	The study is necessary for the regulatory decision, conducted according to GLP	DuPont
B.5.1.2.1/03	Zietz, E.	2002	Field soil dissipation of oxamyl nematicide and insecticide applied as Vydate 10 L by means of drip irrigation to cucurbits in a greenhouse in Italy - Season 2000 Institut Fresenius Chemische und Biologische/Gmbh DuPont-4800 GLP: Yes Published: No	N	Y	The study is necessary for the regulatory decision, conducted according to GLP	DuPont
B.5.2.1/01	Françon, B., Jernberg, K.M., Jetzer, M., Steiner, C.	2001	Method validation of the Netherlands multi-residue method 2 (MRM 2, submethod 1: N-Methylcarbamate pesticides) for the determination of oxamyl in vegetable and fruit crops Battelle Europe-Centre de Recherche de Geneve DuPont-4722 Previously submitted at the EU level for Annex I inclusion Published: No	N	N		DuPont

Data Requirement No., Reference No.	Author(s)	Year	Title Source Company Report No. GLP or GEP Status (where relevant) Published or not	Vertebrate study Y/N	Data Protection Y/N	Justification if data protection is claimed	Owner
B.5.2.1/02	Bacher, R.	2001	Analytical method for the determination of oxamyl and methomyl in animal food stuffs PTRL Europe DuPont-5132 Previously submitted at the EU level for Annex I inclusion Published: No	N	N		DuPont
B.5.2.1/03	Lissemore, L., Harris, A., Patterson, C.	2014b	QuEChERS multiresidue method trials for DPX-D1410, DPX-X1179 and DPX-Q8U80 in crop matrices University of Guelph-Ontario DuPont-41730 GLP: Yes Published: No	N	Y	The study is necessary for the regulatory decision, conducted according to GLP and has not previously been protected or submitted	DuPont
B.5.2.1/04	Schernikau, N., Colorado, C.S.	2015	Independent laboratory validation of DuPont-41730 and DuPont-41763, "Analytical method for the determination of oxamyl (DPX-D1410) and methomyl (DPX-X1179) in crop and animal matrices by LC/ESI-MS/MS" Eurofins Agroscience Services Chem GmbH DuPont-41873 GLP: Yes Published: No	N	Y	The study is necessary for the regulatory decision, conducted according to GLP and has not previously been protected or submitted	DuPont
B.5.2.1/05	Cochrane, J.	2015	Determination of the Extraction Efficiency of [14C] DPX-D1410 Residue Methods: QuEChERS (DuPont-41730) and Crop Method (DuPont-17601 RV.1) DuPont-44316 GLP: Yes Published: No	N	N		

Data Requirement No., Reference No.	Author(s)	Year	Title Source Company Report No. GLP or GEP Status (where relevant) Published or not	Vertebrate study Y/N	Data Protection Y/N	Justification if data protection is claimed	Owner
B.5.2.2/01	Henze, R.M., Klems, J.P.	2014b	Analytical method for the determination of oxamyl in liver, milk, eggs, fat and muscle using LC/MS/MS DuPont Stine-Haskell Research Center DuPont-38597 GLP: No Published: No	N	N		DuPont
B.5.2.2/02	Fiorito, B.	2014	Independent Laboratory Validation of DuPont-38597 "Analytical method for the determination of oxamyl in liver, milk, eggs, fat and muscle using HPLC/MS/MS" Alliance Pharma DuPont-39679 GLP: Yes Published: No	N	Y	The study is necessary for the regulatory decision, conducted according to GLP and has not previously been protected or submitted	DuPont
B.5.2.2/03	Lissemore, L., Harris, A., Patterson, C.	2014a	QuEChERS multiresidue method trials for DPX-D1410, DPX-X1179 and DPX-Q8U80 in animal tissues University of Guelph-Ontario DuPont-41763 GLP: Yes Published: No	N	Y	The study is necessary for the regulatory decision, conducted according to GLP and has not previously been protected or submitted	DuPont

Data Requirement No., Reference No.	Author(s)	Year	Title Source Company Report No. GLP or GEP Status (where relevant) Published or not	Vertebrate study Y/N	Data Protection Y/N	Justification if data protection is claimed	Owner
B.5.2.2/04	Schernikau, N., Colorado, C.S.	2015	Independent laboratory validation of DuPont-41730 and DuPont-41763, "Analytical method for the determination of oxamyl (DPX-D1410) and methomyl (DPX-X1179) in crop and animal matrices by LC/ESI-MS/MS" Eurofins Agroscience Services Chem GmbH DuPont-41873 GLP: Yes Published: No	N	Y	The study is necessary for the regulatory decision, conducted according to GLP and has not previously been protected or submitted	DuPont
B.5.2.3/01	Henze, R.M., Klems, J.P.	2014a	Analytical method for the determination of oxamyl in soil using LC/MS/MS DuPont Stine-Haskell Research Center DuPont-38689 GLP: No Published: No	N	N		DuPont
B.5.2.4/01	Hill, S.J., Maliszewski, P.P., Stry, J.J.	2001	Analytical method for the determination of oxamyl in water using LC/MS/MS DuPont Stine-Haskell Research Center DuPont-5677 Previously submitted at the EU level for Annex I inclusion Published: No	N	N		DuPont

Data Requirement No., Reference No.	Author(s)	Year	Title Source Company Report No. GLP or GEP Status (where relevant) Published or not	Vertebrate study Y/N	Data Protection Y/N	Justification if data protection is claimed	Owner
B.5.2.4/02	Connolly, P.	2001	Independent laboratory validation of method number DuPont-5677, analytical method for the determination of oxamyl in water using LC/MS/MS Centre Analytical Laboratories, Inc. DuPont-6157 Previously submitted at the EU level for Annex I inclusion Published: No	N	N		DuPont
B.5.2.4/03	Klems, J.P.	2014a	Analytical method for the determination of oxamyl in water using LC/MS/MS DuPont Stine-Haskell Research Center DuPont-5677, Supplement No. 2 GLP: No Published: No	N	N		DuPont
B.5.2.4/04	Klems, J.P.	2014b	Analytical method for the determination of oxamyl in blood using LC/MS/MS DuPont Stine-Haskell Research Center DuPont-38598, Revision No. 1 GLP: No Published: No	N	N		DuPont
B.5.2.5/01	Bacher, R.	2000	Development and validation of an analytical method for the determination of oxamyl and methomyl in air PTRL Europe DuPont-4564 Previously submitted at the EU level for Annex I inclusion Published: No	N	N		DuPont

Data Requirement No., Reference No.	Author(s)	Year	Title Source Company Report No. GLP or GEP Status (where relevant) Published or not	Vertebrate study Y/N	Data Protection Y/N	Justification if data protection is claimed	Owner
B.5.2.6/01	Klems, J.P.	2014b	Analytical method for the determination of oxamyl in blood using LC/MS/MS DuPont Stine-Haskell Research Center DuPont-38598, Revision No. 1 GLP: No Published: No	N	N		DuPont
B.5.1.2.13/01	Françon, B., Jetzer, M., Matthey, C.	2000	Method validation for the determination of oxamyl and its oxime metabolite in different crops Battelle Europe-Centre de Recherche de Geneve DuPont-3702 Previously submitted at the EU level for Annex I inclusion Published: No	N	N	DuPont	
B.5.1.2.13/02	Doran, A.M., Cairns, S.D., McGuire, G.M., Vance, C.J.	2003	Validation of an analytical method for the determination of oxamyl in potatoes using LC-MS Inveresk Research International (IRI) Limited (Scotland) DuPont-11125 GLP: Yes Published: No	N	Y	The study is necessary for the regulatory decision, conducted according to GLP	DuPont
B.5.1.2.13/03	Chapleo, S., Johnson, J.	2014	The metabolism of ^{14}C-oxamyl in tomato plants Charles River Laboratories (UK) DuPont-32188 GLP: Yes Published: No	N	Y	The study is necessary for the regulatory decision, conducted according to GLP and has not previously been protected or submitted	DuPont

Data Requirement No., Reference No.	Author(s)	Year	Title Source Company Report No. GLP or GEP Status (where relevant) Published or not	Vertebrate study Y/N	Data Protection Y/N	Justification if data protection is claimed	Owner
B.5.1.2.13/04	Cairns, S.D., Davidson, J.	2006	Validation of an analytical method for the determination of oxamyl in green, dried and fermented tobacco leaves Inveresk Research DuPont-17601 GLP: Yes Published: No	N	Y	The study is necessary for the regulatory decision, conducted according to GLP and has not previously been protected or submitted	DuPont
B.5.1.2.13/05	Cairns, S.	2012	Method validation for the analysis of oxamyl (DPX-D1410) in representative crop commodities using LC-MS/MS Charles River Laboratories (UK) DuPont-33191 GLP: Yes Published: No	N	Y	The study is necessary for the regulatory decision, conducted according to GLP and has not previously been protected or submitted	DuPont
B.5.1.2.13/06	Chapleo, S., Johnson, J.	2014	The metabolism of ^{14}C-oxamyl in tomato plants Charles River Laboratories (UK) DuPont-32188 GLP: Yes Published: No	N	Y	The study is necessary for the regulatory decision, conducted according to GLP and has not previously been protected or submitted	DuPont
B.5.1.2.15/01	Schwarz, A., Eichler, M.	2014	Field study on residues in arthropods, earthworms and seedlings (wildlife food items) IBACON DuPont-40221 GLP: Yes Published: No	N	Y	The study is necessary for the regulatory decision, conducted according to GLP and has not previously been protected or submitted	DuPont

Data Requirement No., Reference No.	Author(s)	Year	Title Source Company Report No. GLP or GEP Status (where relevant) Published or not	Vertebrate study Y/N	Data Protection Y/N	Justification if data protection is claimed	Owner
B.5.1.2.15/02	Cairns, S.	2012	Method validation for the analysis of oxamyl (DPX-D1410) in representative crop commodities using LC-MS/MS Charles River Laboratories (UK) DuPont-33191 GLP: Yes Published: No	N	Y	The study is necessary for the regulatory decision, conducted according to GLP and has not previously been protected or submitted	DuPont
B.5.1.2.15/03	Chapleo, S., Johnson, J.	2014	The metabolism of ^{14}C-oxamyl in tomato plants Charles River Laboratories (UK) DuPont-32188 GLP: Yes Published: No	N	Y	The study is necessary for the regulatory decision, conducted according to GLP and has not previously been protected or submitted	DuPont
B.5.1.2.15/04	Meinerling, M.	2014	Oxamyl (DPX-D1410) technical (98% w/w): Accumulation and elimination in earthworms (*Eisenia fetida*) in artificial soil Institut für Biologische Analytik und Consulting IBACON GmbH DuPont-38477 GLP: Yes Published: No	N	Y	Data protection on a MS by MS basis. The study provides additional data for the regulatory decision, conducted according to GLP and has not previously been protected or submitted in all MS.	DuPont
B.5.1.2.15/05	McClory, J.P.	2004	Analytical method for the determination of oxamyl and IN-A2213 metabolite in soil using LC/MS/MS TNO Nutrition and Food Research DuPont-7191, Revision No. 1 GLP: No Published:	N	N		DuPont

Data Requirement No., Reference No.	Author(s)	Year	Title Source Company Report No. GLP or GEP Status (where relevant) Published or not	Vertebrate study Y/N	Data Protection Y/N	Justification if data protection is claimed	Owner
B.5.1.2.15/06	Scherer, F.	2015	Oxamyl (DPX-D1410) 10GR: Investigating the deposition of dust from in-furrow application of granules containing oxamyl and determination of residues of oxamyl in guttation fluid of potato in United Kingdom during 2014 Eurofins Agroscience Services EcoChem GmbH DuPont-38691 GLP: Yes Published: No	N	Y	The study is necessary for the regulatory decision, conducted according to GLP and has not previously been protected or submitted	DuPont
B.5.1.2.15/07	Berg, C.	2015a	Oxamyl (DPX-D1410) 10GR [100 g/kg (w/w)]: A semi-field study to evaluate effects on the bumble bee (*Bombus terrestris* L; Hymenoptera, Apidae) in *Solanum tuberosum* in Germany in 2014 DuPont-39666 GLP: Yes Published: No	N	Y	The study is necessary for the regulatory decision, conducted according to GLP and has not previously been protected or submitted	DuPont
B.5.1.2.15/08	Berg, C.	2015b	Oxamyl (DPX-D1410) 10GR [100 g/kg]: A semi-field study to evaluate effects on the brood of honey bees (*Apis mellifera*; Hymenoptera, Apidae) in *Phacelia tanacetifolia* in Germany 2014 Eurofins Agroscience Services EcoChem GmbH DuPont-39667 GLP: Yes Published: No	N	Y	The study is necessary for the regulatory decision, conducted according to GLP and has not previously been protected or submitted	DuPont

Data Requirement No., Reference No.	Author(s)	Year	Title Source Company Report No. GLP or GEP Status (where relevant) Published or not	Vertebrate study Y/N	Data Protection Y/N	Justification if data protection is claimed	Owner
B.5.1.2.15/09	Mack, P., Knabe, S.	2015	Guttation residues after in-furrow application to sugar beet Eurofins Agroscience Services EcoChem GmbH DuPont-41322, Revision No. 1 GLP: Yes Published: No	N	Y	The study is necessary for the regulatory decision, conducted according to GLP and has not previously been protected or submitted	DuPont
B.5.1.2.16/02	Brougher, D.S.	2013	Oxamyl (DPX-D1410) technical (98% w/w): A 48-hour static acute toxicity test with the cladoceran (*Ceriodaphnia dubia*) Wildlife International Ltd. (USA) DuPont-37399 GLP: Yes Published: No	N	Y	The study is necessary for the regulatory decision, conducted according to GLP and has not previously been protected or submitted	DuPont
B.5.1.2.16/01	Brougher, D.S., Martin, K.H., Gallagher, S.P., Bodle, E.S.	2013a	Oxamyl (DPX-D1410) technical (98% w/w): A 48-hour static acute toxicity test with *Hyalella azteca* Wildlife International Ltd. (USA) DuPont-37397 GLP: Yes Published: No	N	Y	The study is necessary for the regulatory decision, conducted according to GLP and has not previously been protected or submitted	DuPont
B.5.1.2.16/03	Brougher, D.S., Martin, K.H., Gallagher, S.P., Bodle, E.S.	2013b	Oxamyl (DPX-D1410) technical (98% w/w): A 48-hour static acute toxicity test with *Chironomus tentans* Wildlife International Ltd. (USA) DuPont-37400 GLP: Yes Published: No	N	Y	The study is necessary for the regulatory decision, conducted according to GLP and has not previously been protected or submitted	DuPont

Data Requirement No., Reference No.	Author(s)	Year	Title Source Company Report No. GLP or GEP Status (where relevant) Published or not	Vertebrate study Y/N	Data Protection Y/N	Justification if data protection is claimed	Owner
B.5.1.2.16/04	Brougher, D.S., Martin, K.H., Gallagher, S.P., Krueger, H.O.	2013a	Oxamyl (DPX-D1410) technical (98% w/w): A 48-hour static acute toxicity test with the mayfly (*Centroptilum triangulifer*) Wildlife International Ltd. (USA) DuPont-37401 GLP: Yes Published: No	N	Y	The study is necessary for the regulatory decision, conducted according to GLP and has not previously been protected or submitted	DuPont
B.5.1.2.16/05	Brougher, D.S., Martin, K.H., Gallagher, S.P., Krueger, H.O.	2013b	Oxamyl (DPX-D1410) technical (98% w/w): A 48-hour static acute toxicity test with the caddisfly (*Chimarra atterima*) Wildlife International Ltd. (USA) DuPont-37402 GLP: Yes Published: No	N	Y	The study is necessary for the regulatory decision, conducted according to GLP and has not previously been protected or submitted	DuPont
B.5.1.2.16/06	[redacted]	2000b	Oxamyl 10L: Static, acute, 96-hour, (LC_{50}) test to rainbow trout, *Oncorhynchus mykiss* [redacted] DuPont-2910 GLP: Yes Published: No	Y	Y	The study is necessary for the regulatory decision, conducted according to GLP	DuPont
B.5.1.2.16/07	[redacted]	2000c	Oxamyl 10L: Static-renewal, acute, 96-hour, (LC_{50}) test to bluegill sunfish, *Lepomis macrochirus* [redacted] DuPont-2911 GLP: Yes Published: No	Y	Y	The study is necessary for the regulatory decision, conducted according to GLP	DuPont

Data Requirement No., Reference No.	Author(s)	Year	Title Source Company Report No. GLP or GEP Status (where relevant) Published or not	Vertebrate study Y/N	Data Protection Y/N	Justification if data protection is claimed	Owner
B.5.1.2.16/08	Ward, T.J., Magazu, J.P., Boeri, R.L.	2000a	Oxamyl 10L: Acute, static-renewal, 48-hour EC_{50} to *Daphnia magna* T.R. Wilbury Laboratories, Inc. DuPont-2556 GLP: Yes Published: No	N	Y	The study is necessary for the regulatory decision, conducted according to GLP	DuPont
B.5.1.2.16/09	Boeri, R.L., Ward, T.J.	2000	Oxamyl 10L: Influence on growth and growth rate of the alga, *Selenastrum capricornutum* T.R. Wilbury Laboratories, Inc. DuPont-3913 GLP: Yes Published: No	N	Y	The study is necessary for the regulatory decision, conducted according to GLP	DuPont
B.5.1.2.16/10	[redacted]	2012b	Oxamyl (DPX-D1410) technical (98% w/w): Early life-stage toxicity test with the sheepshead minnow, *Cyprinodon variegatus*, under flow-through conditions [redacted] DuPont-34270 GLP: Yes Published: No	Y	Y	The study is necessary for the regulatory decision, conducted according to GLP and has not previously been protected or submitted	DuPont
B.5.1.2.16/11	Hicks, S.L.	2013	Oxamyl (DPX-D1410) technical (98% w/w): Life-cycle toxicity test of the saltwater mysid, *Americamysis bahia*, conducted under flow-through conditions ABC Laboratories, Inc. (Missouri) DuPont-34269 GLP: Yes Published: No	N	Y	The study is necessary for the regulatory decision, conducted according to GLP and has not previously been protected or submitted	DuPont

Data Requirement No., Reference No.	Author(s)	Year	Title Source Company Report No. GLP or GEP Status (where relevant) Published or not	Vertebrate study Y/N	Data Protection Y/N	Justification if data protection is claimed	Owner
B.5.1.2.16/12	Rebstock, M.	2012b	Oxamyl (DPX-D1410) technical (98% w/w): Acute toxicity with the mysid shrimp, *Americamysis bahia*, determined under flow-through test conditions ABC Laboratories, Inc. (Missouri) DuPont-34271 GLP: Yes Published: No	N	Y	The study is necessary for the regulatory decision, conducted according to GLP and has not previously been protected or submitted	DuPont
B.5.1.2.16/13	[redacted]	2012a	Oxamyl technical (DPX-D1410): Short term reproduction assay with the fathead minnow, *Pimephales promelas*, determined under flow-through conditions [redacted] DuPont-31031 GLP: Yes Published: No	Y	Y	The study is necessary for the regulatory decision, conducted according to GLP and has not previously been protected or submitted	DuPont
B.5.1.2.16/14	[redacted]	2015	Oxamyl (DPX-D1410) technical: 21-D amphibian metamorphosis assay (AMA) with south African clawed frog, *Xenopus laevis* [redacted] DuPont-31032, Revision No. 1 GLP: Yes Published: No	Y	Y	The study is necessary for the regulatory decision, conducted according to GLP and has not previously been protected or submitted	DuPont
B.5.1.2.16/15	Rebstock, M.	2012a	Oxamyl (DPX-D1410) technical (98% w/w): 7-day growth inhibition test with the freshwater aquatic plant, duckweed, *Lemna gibba* ABC Laboratories, Inc. (Missouri) DuPont-34272 GLP: Yes Published: No	N	Y	The study is necessary for the regulatory decision, conducted according to GLP and has not previously been protected or submitted	DuPont

Data Requirement No., Reference No.	Author(s)	Year	Title Source Company Report No. GLP or GEP Status (where relevant) Published or not	Vertebrate study Y/N	Data Protection Y/N	Justification if data protection is claimed	Owner
B.5.1.2.16/16	Hicks, S.L.	2012c	Oxamyl (DPX-D1410) technical (98% w/w): Effect on new shell growth of the eastern oyster (*Crassostrea virginica*) ABC Laboratories, Inc. (Missouri) DuPont-34273 GLP: Yes Published: No	N	Y	The study is necessary for the regulatory decision, conducted according to GLP and has not previously been protected or submitted	DuPont
B.5.1.2.16/17	Bergfield, A.	2012a	Oxamyl (DPX-D1410) 24SL: A greenhouse study to investigate the effects on seedling emergence and growth of tomato, *Lycopersicon esculentum*, following soil exposure ABC Laboratories, Inc. (Missouri) DuPont-34274 GLP: Yes Published: No	N	Y	The study is necessary for the regulatory decision, conducted according to GLP and has not previously been protected or submitted	DuPont
B.5.1.2.16/18	Bergfield, A.	2012b	Oxamyl (DPX-D1410) 24SL: A greenhouse study to investigate the effects on vegetative vigor of ten terrestrial plant species following foliar exposure ABC Laboratories, Inc. (Missouri) DuPont-34275 GLP: Yes Published: No	N	Y	The study is necessary for the regulatory decision, conducted according to GLP and has not previously been protected or submitted	DuPont
B.5.1.2.16/19	Brougher, D.S., Martin, K.H., Gallagher, S.P., Bodle, E.S.	2013c	Oxamyl (DPX-D1410) technical (98% w/w): A 48-hour static acute toxicity test with the cladoceran (*Daphnia pulex*) Wildlife International Ltd. (USA) DuPont-37398 GLP: Yes Published: No	N	Y	The study is necessary for the regulatory decision, conducted according to GLP and has not previously been protected or submitted	DuPont

Data Requirement No., Reference No.	Author(s)	Year	Title Source Company Report No. GLP or GEP Status (where relevant) Published or not	Vertebrate study Y/N	Data Protection Y/N	Justification if data protection is claimed	Owner
B.5.1.2.16/20	Schmitt, H.	2014	Oxamyl (DPX-D1410) technical (98% w/w): Assessment of effects to the honeybee, *Apis mellifera* L., in a 10 days chronic feeding test under laboratory conditions Eurofins Agroscience Services, Gmbh DuPont-39665 GLP: Yes Published: No	N	Y	The study is necessary for the regulatory decision, conducted according to GLP and has not previously been protected or submitted	DuPont
B.5.1.2.16/21	Klank, C.	2014	Oxamyl (DPX-D1410) technical (98% w/w): Honey bee (*Apis mellifera* L.) larval toxicity test (single feeding exposure) Eurofins Agroscience Services EcoChem GmbH DuPont-39678 GLP: Yes Published: No	N	Y	The study is necessary for the regulatory decision, conducted according to GLP and has not previously been protected or submitted	DuPont
B.5.1.2.17/01	Harsha, N.V.	2009	Oxamyl (DPX-D1410-196): Laboratory study of vapour pressure International Institute of Biotechnology and Toxicology (IIBAT) DuPont-26259 GLP: Yes Published: No	N	Y	The study is necessary for the regulatory decision, conducted according to GLP and has not previously been protected or submitted	DuPont

Sorted by Author

Data Requirement No., Reference No.	Author(s)	Year	Title Source Company Report No. GLP or GEP Status (where relevant) Published or not	Vertebrate study Y/N	Data Protection Y/N	Justification if data protection is claimed	Owner
B.5.2.1/02	Bacher, R.	2001	Analytical method for the determination of oxamyl and methomyl in animal food stuffs PTRL Europe DuPont-5132 Previously submitted at the EU level for Annex I inclusion Published: No	N	N		DuPont
B.5.2.5/01	Bacher, R.	2000	Development and validation of an analytical method for the determination of oxamyl and methomyl in air PTRL Europe DuPont-4564 Previously submitted at the EU level for Annex I inclusion Published: No	N	N		DuPont
B.5.1.2.15/07	Berg, C.	2015a	Oxamyl (DPX-D1410) 10GR [100 g/kg (w/w)]: A semi-field study to evaluate effects on the bumble bee (*Bombus terrestris* L; Hymenoptera, Apidae) in *Solanum tuberosum* in Germany in 2014 DuPont-39666 GLP: Yes Published: No	N	Y	The study is necessary for the regulatory decision, conducted according to GLP and has not previously been protected or submitted	DuPont

Data Requirement No., Reference No.	Author(s)	Year	Title Source Company Report No. GLP or GEP Status (where relevant) Published or not	Vertebrate study Y/N	Data Protection Y/N	Justification if data protection is claimed	Owner
B.5.1.2.15/08	Berg, C.	2015b	Oxamyl (DPX-D1410) 10GR [100 g/kg]: A semi-field study to evaluate effects on the brood of honey bees (*Apis mellifera*; Hymenoptera, Apidae) in *Phacelia tanacetifolia* in Germany 2014 Eurofins Agroscience Services EcoChem GmbH DuPont-39667 GLP: Yes Published: No	N	Y	The study is necessary for the regulatory decision, conducted according to GLP and has not previously been protected or submitted	DuPont
B.5.1.2.16/17	Bergfield, A.	2012a	Oxamyl (DPX-D1410) 24SL: A greenhouse study to investigate the effects on seedling emergence and growth of tomato, *Lycopersicon esculentum*, following soil exposure ABC Laboratories, Inc. (Missouri) DuPont-34274 GLP: Yes Published: No	N	Y	The study is necessary for the regulatory decision, conducted according to GLP and has not previously been protected or submitted	DuPont
B.5.1.2.16/18	Bergfield, A.	2012b	Oxamyl (DPX-D1410) 24SL: A greenhouse study to investigate the effects on vegetative vigor of ten terrestrial plant species following foliar exposure ABC Laboratories, Inc. (Missouri) DuPont-34275 GLP: Yes Published: No	N	Y	The study is necessary for the regulatory decision, conducted according to GLP and has not previously been protected or submitted	DuPont

Data Requirement No., Reference No.	Author(s)	Year	Title Source Company Report No. GLP or GEP Status (where relevant) Published or not	Vertebrate study Y/N	Data Protection Y/N	Justification if data protection is claimed	Owner
B.5.1.2.16/09	Boeri, R.L., Ward, T.J.	2000	Oxamyl 10L: Influence on growth and growth rate of the alga, *Selenastrum capricornutum* T.R. Wilbury Laboratories, Inc. DuPont-3913 GLP: Yes Published: No	N	Y	The study is necessary for the regulatory decision, conducted according to GLP	DuPont
B.5.1.2.16/02	Brougher, D.S.	2013	Oxamyl (DPX-D1410) technical (98% w/w): A 48-hour static acute toxicity test with the cladoceran (*Ceriodaphnia dubia*) Wildlife International Ltd. (USA) DuPont-37399 GLP: Yes Published: No	N	Y	The study is necessary for the regulatory decision, conducted according to GLP and has not previously been protected or submitted	DuPont
B.5.1.2.16/01	Brougher, D.S., Martin, K.H., Gallagher, S.P., Bodle, E.S.	2013a	Oxamyl (DPX-D1410) technical (98% w/w): A 48-hour static acute toxicity test with *Hyalella azteca* Wildlife International Ltd. (USA) DuPont-37397 GLP: Yes Published: No	N	Y	The study is necessary for the regulatory decision, conducted according to GLP and has not previously been protected or submitted	DuPont
B.5.1.2.16/03	Brougher, D.S., Martin, K.H., Gallagher, S.P., Bodle, E.S.	2013b	Oxamyl (DPX-D1410) technical (98% w/w): A 48-hour static acute toxicity test with *Chironomus tentans* Wildlife International Ltd. (USA) DuPont-37400 GLP: Yes Published: No	N	Y	The study is necessary for the regulatory decision, conducted according to GLP and has not previously been protected or submitted	DuPont

Data Requirement No., Reference No.	Author(s)	Year	Title Source Company Report No. GLP or GEP Status (where relevant) Published or not	Vertebrate study Y/N	Data Protection Y/N	Justification if data protection is claimed	Owner
B.5.1.2.16/19	Brougher, D.S., Martin, K.H., Gallagher, S.P., Bodle, E.S.	2013c	Oxamyl (DPX-D1410) technical (98% w/w): A 48-hour static acute toxicity test with the cladoceran (*Daphnia pulex*) Wildlife International Ltd. (USA) DuPont-37398 GLP: Yes Published: No	N	Y	The study is necessary for the regulatory decision, conducted according to GLP and has not previously been protected or submitted	DuPont
B.5.1.2.16/04	Brougher, D.S., Martin, K.H., Gallagher, S.P., Krueger, H.O.	2013a	Oxamyl (DPX-D1410) technical (98% w/w): A 48-hour static acute toxicity test with the mayfly (*Centroptilum triangulifer*) Wildlife International Ltd. (USA) DuPont-37401 GLP: Yes Published: No	N	Y	The study is necessary for the regulatory decision, conducted according to GLP and has not previously been protected or submitted	DuPont
B.5.1.2.16/05	Brougher, D.S., Martin, K.H., Gallagher, S.P., Krueger, H.O.	2013b	Oxamyl (DPX-D1410) technical (98% w/w): A 48-hour static acute toxicity test with the caddisfly (*Chimarra atterima*) Wildlife International Ltd. (USA) DuPont-37402 GLP: Yes Published: No	N	Y	The study is necessary for the regulatory decision, conducted according to GLP and has not previously been protected or submitted	DuPont
B.5.1.2.13/05	Cairns, S.	2012	Method validation for the analysis of oxamyl (DPX-D1410) in representative crop commodities using LC-MS/MS Charles River Laboratories (UK) DuPont-33191 GLP: Yes Published: No	N	Y	The study is necessary for the regulatory decision, conducted according to GLP and has not previously been protected or submitted	DuPont

Data Requirement No., Reference No.	Author(s)	Year	Title Source Company Report No. GLP or GEP Status (where relevant) Published or not	Vertebrate study Y/N	Data Protection Y/N	Justification if data protection is claimed	Owner
B.5.1.2.15/02	Cairns, S.	2012	Method validation for the analysis of oxamyl (DPX-D1410) in representative crop commodities using LC-MS/MS Charles River Laboratories (UK) DuPont-33191 GLP: Yes Published: No	N	Y	The study is necessary for the regulatory decision, conducted according to GLP and has not previously been protected or submitted	DuPont
B.5.1.2.13/04	Cairns, S.D., Davidson, J.	2006	Validation of an analytical method for the determination of oxamyl in green, dried and fermented tobacco leaves Inveresk Research DuPont-17601 GLP: Yes Published: No	N	Y	The study is necessary for the regulatory decision, conducted according to GLP and has not previously been protected or submitted	DuPont
B.5.1.2.13/03	Chapleo, S., Johnson, J.	2014	The metabolism of ^{14}C-oxamyl in tomato plants Charles River Laboratories (UK) DuPont-32188 GLP: Yes Published: No	N	Y	The study is necessary for the regulatory decision, conducted according to GLP and has not previously been protected or submitted	DuPont
B.5.1.2.13/06	Chapleo, S., Johnson, J.	2014	The metabolism of ^{14}C-oxamyl in tomato plants Charles River Laboratories (UK) DuPont-32188 GLP: Yes Published: No	N	Y	The study is necessary for the regulatory decision, conducted according to GLP and has not previously been protected or submitted	DuPont

Data Requirement No., Reference No.	Author(s)	Year	Title Source Company Report No. GLP or GEP Status (where relevant) Published or not	Vertebrate study Y/N	Data Protection Y/N	Justification if data protection is claimed	Owner
B.5.1.2.15/03	Chapleo, S., Johnson, J.	2014	The metabolism of ^{14}C-oxamyl in tomato plants Charles River Laboratories (UK) DuPont-32188 GLP: Yes Published: No	N	Y	The study is necessary for the regulatory decision, conducted according to GLP and has not previously been protected or submitted	DuPont
B.5.2.1/05	Cochrane, J.	2015	Determination of the Extraction Efficiency of [14C] DPX-D1410 Residue Methods: QuEChERS (DuPont-41730) and Crop Method (DuPont-17601 RV.1) DuPont-44316 GLP: Yes Published: No	N	N		
B.5.2.4/02	Connolly, P.	2001	Independent laboratory validation of method number DuPont-5677, analytical method for the determination of oxamyl in water using LC/MS/MS Centre Analytical Laboratories, Inc. DuPont-6157 Previously submitted at the EU level for Annex I inclusion Published: No	N	N		DuPont
B.5.1.2.13/02	Doran, A.M., Cairns, S.D., McGuire, G.M., Vance, C.J.	2003	Validation of an analytical method for the determination of oxamyl in potatoes using LC-MS Inveresk Research International (IRI) Limited (Scotland) DuPont-11125 GLP: Yes Published: No	N	Y	The study is necessary for the regulatory decision, conducted according to GLP	DuPont

Data Requirement No., Reference No.	Author(s)	Year	Title Source Company Report No. GLP or GEP Status (where relevant) Published or not	Vertebrate study Y/N	Data Protection Y/N	Justification if data protection is claimed	Owner
B.5.2.2/02	Fiorito, B.	2014	Independent Laboratory Validation of DuPont-38597 "Analytical method for the determination of oxamyl in liver, milk, eggs, fat and muscle using HPLC/MS/MS" Alliance Pharma DuPont-39679 GLP: Yes Published: No	N	Y	The study is necessary for the regulatory decision, conducted according to GLP and has not previously been protected or submitted	DuPont
B.5.1.2.16/14	[redacted]	2015	Oxamyl (DPX-D1410) technical: 21-D amphibian metamorphosis assay (AMA) with south African clawed frog, *Xenopus laevis* [redacted] DuPont-31032, Revision No. 1 GLP: Yes Published: No	Y	Y	The study is necessary for the regulatory decision, conducted according to GLP and has not previously been protected or submitted	DuPont
B.5.1.2.13/01	Françon, B., Jetzer, M., Matthey, C.	2000	Method validation for the determination of oxamyl and its oxime metabolite in different crops Battelle Europe-Centre de Recherche de Geneve DuPont-3702 Previously submitted at the EU level for Annex I inclusion Published: No	N	N	DuPont	

Data Requirement No., Reference No.	Author(s)	Year	Title Source Company Report No. GLP or GEP Status (where relevant) Published or not	Vertebrate study Y/N	Data Protection Y/N	Justification if data protection is claimed	Owner
B.5.2.1/01	Françon, B., Jernberg, K.M., Jetzer, M., Steiner, C.	2001	Method validation of the Netherlands multi-residue method 2 (MRM 2, submethod 1: N-Methylcarbamate pesticides) for the determination of oxamyl in vegetable and fruit crops Battelle Europe-Centre de Recherche de Geneve DuPont-4722 Previously submitted at the EU level for Annex I inclusion Published: No	N	N		DuPont
B.5.1.2.17/01	Harsha, N.V.	2009	Oxamyl (DPX-D1410-196): Laboratory study of vapour pressure International Institute of Biotechnology and Toxicology (IIBAT) DuPont-26259 GLP: Yes Published: No	N	Y	The study is necessary for the regulatory decision, conducted according to GLP and has not previously been protected or submitted	DuPont
B.5.2.2/01	Henze, R.M., Klems, J.P.	2014b	Analytical method for the determination of oxamyl in liver, milk, eggs, fat and muscle using LC/MS/MS DuPont Stine-Haskell Research Center DuPont-38597 GLP: No Published: No	N	N		DuPont

Data Requirement No., Reference No.	Author(s)	Year	Title Source Company Report No. GLP or GEP Status (where relevant) Published or not	Vertebrate study Y/N	Data Protection Y/N	Justification if data protection is claimed	Owner
B.5.2.3/01	Henze, R.M., Klems, J.P.	2014a	Analytical method for the determination of oxamyl in soil using LC/MS/MS DuPont Stine-Haskell Research Center DuPont-38689 GLP: No Published: No	N	N		DuPont
B.5.1.2.16/10	[redacted]	2012b	Oxamyl (DPX-D1410) technical (98% w/w): Early life-stage toxicity test with the sheepshead minnow, *Cyprinodon variegatus*, under flow-through conditions [redacted] DuPont-34270 GLP: Yes Published: No	Y	Y	The study is necessary for the regulatory decision, conducted according to GLP and has not previously been protected or submitted	DuPont
B.5.1.2.16/11	Hicks, S.L.	2013	Oxamyl (DPX-D1410) technical (98% w/w): Life-cycle toxicity test of the saltwater mysid, *Americamysis bahia*, conducted under flow-through conditions ABC Laboratories, Inc. (Missouri) DuPont-34269 GLP: Yes Published: No	N	Y	The study is necessary for the regulatory decision, conducted according to GLP and has not previously been protected or submitted	DuPont
B.5.1.2.16/13	[redacted]	2012a	Oxamyl technical (DPX-D1410): Short term reproduction assay with the fathead minnow, *Pimephales promelas*, determined under flow-through conditions [redacted] DuPont-31031 GLP: Yes Published: No	Y	Y	The study is necessary for the regulatory decision, conducted according to GLP and has not previously been protected or submitted	DuPont

Data Requirement No., Reference No.	Author(s)	Year	Title Source Company Report No. GLP or GEP Status (where relevant) Published or not	Vertebrate study Y/N	Data Protection Y/N	Justification if data protection is claimed	Owner
B.5.1.2.16/16	Hicks, S.L.	2012c	Oxamyl (DPX-D1410) technical (98% w/w): Effect on new shell growth of the eastern oyster (*Crassostrea virginica*) ABC Laboratories, Inc. (Missouri) DuPont-34273 GLP: Yes Published: No	N	Y	The study is necessary for the regulatory decision, conducted according to GLP and has not previously been protected or submitted	DuPont
B.5.2.4/01	Hill, S.J., Maliszewski, P.P., Stry, J.J.	2001	Analytical method for the determination of oxamyl in water using LC/MS/MS DuPont Stine-Haskell Research Center DuPont-5677 Previously submitted at the EU level for Annex I inclusion Published: No	N	N		DuPont
B.5.1.2.16/21	Klank, C.	2014	Oxamyl (DPX-D1410) technical (98% w/w): Honey bee (*Apis mellifera* L.) larval toxicity test (single feeding exposure) Eurofins Agroscience Services EcoChem GmbH DuPont-39678 GLP: Yes Published: No	N	Y	The study is necessary for the regulatory decision, conducted according to GLP and has not previously been protected or submitted	DuPont
B.5.2.4/03	Klems, J.P.	2014a	Analytical method for the determination of oxamyl in water using LC/MS/MS DuPont Stine-Haskell Research Center DuPont-5677, Supplement No. 2 GLP: No Published: No	N	N		DuPont

Data Requirement No., Reference No.	Author(s)	Year	Title Source Company Report No. GLP or GEP Status (where relevant) Published or not	Vertebrate study Y/N	Data Protection Y/N	Justification if data protection is claimed	Owner
B.5.2.4/04	Klems, J.P.	2014b	Analytical method for the determination of oxamyl in blood using LC/MS/MS DuPont Stine-Haskell Research Center DuPont-38598, Revision No. 1 GLP: No Published: No	N	N		DuPont
B.5.2.6/01	Klems, J.P.	2014b	Analytical method for the determination of oxamyl in blood using LC/MS/MS DuPont Stine-Haskell Research Center DuPont-38598, Revision No. 1 GLP: No Published: No	N	N		DuPont
B.5.2.1/03	Lissemore, L., Harris, A., Patterson, C.	2014b	QuEChERS multiresidue method trials for DPX-D1410, DPX-X1179 and DPX-Q8U80 in crop matrices University of Guelph-Ontario DuPont-41730 GLP: Yes Published: No	N	Y	The study is necessary for the regulatory decision, conducted according to GLP and has not previously been protected or submitted	DuPont
B.5.2.2/03	Lissemore, L., Harris, A., Patterson, C.	2014a	QuEChERS multiresidue method trials for DPX-D1410, DPX-X1179 and DPX-Q8U80 in animal tissues University of Guelph-Ontario DuPont-41763 GLP: Yes Published: No	N	Y	The study is necessary for the regulatory decision, conducted according to GLP and has not previously been protected or submitted	DuPont

Data Requirement No., Reference No.	Author(s)	Year	Title Source Company Report No. GLP or GEP Status (where relevant) Published or not	Vertebrate study Y/N	Data Protection Y/N	Justification if data protection is claimed	Owner
B.5.1.2.15/09	Mack, P., Knabe, S.	2015	Guttation residues after in-furrow application to sugar beet Eurofins Agroscience Services EcoChem GmbH DuPont-41322, Revision No. 1 GLP: Yes Published: No	N	Y	The study is necessary for the regulatory decision, conducted according to GLP and has not previously been protected or submitted	DuPont
B.5.1.2.15/05	McClory, J.P.	2004	Analytical method for the determination of oxamyl and IN-A2213 metabolite in soil using LC/MS/MS TNO Nutrition and Food Research DuPont-7191, Revision No. 1 GLP: No Published:	N	N		DuPont
B.5.1.2.15/04	Meinerling, M.	2014	Oxamyl (DPX-D1410) technical (98% w/w): Accumulation and elimination in earthworms (*Eisenia fetida*) in artificial soil Institut für Biologische Analytik und Consulting IBACON GmbH DuPont-38477 GLP: Yes Published: No	N	Y	Data protection on a MS by MS basis. The study provides additional data for the regulatory decision, conducted according to GLP and has not previously been protected or submitted in all MS.	DuPont
B.5.1.2.1/01	Nanita, S.C.	2009	Analytical method for the determination of oxamyl and its oxime metabolite in soil using LC/MS analysis DuPont Experimental Station DuPont-2392, Revision No. 1 GLP: No Published: No	N	N		DuPont

Data Requirement No., Reference No.	Author(s)	Year	Title Source Company Report No. GLP or GEP Status (where relevant) Published or not	Vertebrate study Y/N	Data Protection Y/N	Justification if data protection is claimed	Owner
B.5.1.1.1/02	Pandey, S.	2015b	Validation of the analytical method for determination of oxamyl (DPX- D1410) in technical grade oxamyl and oxamyl end-use products by reverse phase high performance liquid chromatography (RPLC) and ultra performance liquid chromatography (UPLC) Syngene DuPont-36605 and DuPont-36605 Confidential attachment GLP: Yes Published: No	N	Y	The study is necessary for the regulatory decision, conducted according to GLP and has not previously been protected or submitted	DuPont
B.5.1.1.1/01	Pandey, S., McNally, M.E.P.	2015f	Determination of oxamyl (DPX-D1410) in technical grade oxamyl and end-use products DuPont-42001 GLP: No Published: No	N	N		DuPont
B.5.1.2.16/12	Rebstock, M.	2012b	Oxamyl (DPX-D1410) technical (98% w/w): Acute toxicity with the mysid shrimp, *Americamysis bahia*, determined under flow-through test conditions ABC Laboratories, Inc. (Missouri) DuPont-34271 GLP: Yes Published: No	N	Y	The study is necessary for the regulatory decision, conducted according to GLP and has not previously been protected or submitted	DuPont

Data Requirement No., Reference No.	Author(s)	Year	Title Source Company Report No. GLP or GEP Status (where relevant) Published or not	Vertebrate study Y/N	Data Protection Y/N	Justification if data protection is claimed	Owner
B.5.1.2.16/15	Rebstock, M.	2012a	Oxamyl (DPX-D1410) technical (98% w/w): 7-day growth inhibition test with the freshwater aquatic plant, duckweed, *Lemna gibba* ABC Laboratories, Inc. (Missouri) DuPont-34272 GLP: Yes Published: No	N	Y	The study is necessary for the regulatory decision, conducted according to GLP and has not previously been protected or submitted	DuPont
B.5.1.2.15/06	Scherer, F.	2015	Oxamyl (DPX-D1410) 10GR: Investigating the deposition of dust from in-furrow application of granules containing oxamyl and determination of residues of oxamyl in guttation fluid of potato in United Kingdom during 2014 Eurofins Agroscience Services EcoChem GmbH DuPont-38691 GLP: Yes Published: No	N	Y	The study is necessary for the regulatory decision, conducted according to GLP and has not previously been protected or submitted	DuPont
B.5.2.1/04	Schernikau, N., Colorado, C.S.	2015	Independent laboratory validation of DuPont-41730 and DuPont-41763, "Analytical method for the determination of oxamyl (DPX-D1410) and methomyl (DPX-X1179) in crop and animal matrices by LC/ESI-MS/MS" Eurofins Agroscience Services Chem GmbH DuPont-41873 GLP: Yes Published: No	N	Y	The study is necessary for the regulatory decision, conducted according to GLP and has not previously been protected or submitted	DuPont

Data Requirement No., Reference No.	Author(s)	Year	Title Source Company Report No. GLP or GEP Status (where relevant) Published or not	Vertebrate study Y/N	Data Protection Y/N	Justification if data protection is claimed	Owner
B.5.2.2/04	Schernikau, N., Colorado, C.S.	2015	Independent laboratory validation of DuPont-41730 and DuPont-41763, "Analytical method for the determination of oxamyl (DPX-D1410) and methomyl (DPX-X1179) in crop and animal matrices by LC/ESI-MS/MS" Eurofins Agroscience Services Chem GmbH DuPont-41873 GLP: Yes Published: No	N	Y	The study is necessary for the regulatory decision, conducted according to GLP and has not previously been protected or submitted	DuPont
B.5.1.2.16/20	Schmitt, H.	2014	Oxamyl (DPX-D1410) technical (98% w/w): Assessment of effects to the honeybee, *Apis mellifera* L., in a 10 days chronic feeding test under laboratory conditions Eurofins Agroscience Services, Gmbh DuPont-39665 GLP: Yes Published: No	N	Y	The study is necessary for the regulatory decision, conducted according to GLP and has not previously been protected or submitted	DuPont
B.5.1.2.15/01	Schwarz, A., Eichler, M.	2014	Field study on residues in arthropods, earthworms and seedlings (wildlife food items) IBACON DuPont-40221 GLP: Yes Published: No	N	Y	The study is necessary for the regulatory decision, conducted according to GLP and has not previously been protected or submitted	DuPont

Data Requirement No., Reference No.	Author(s)	Year	Title Source Company Report No. GLP or GEP Status (where relevant) Published or not	Vertebrate study Y/N	Data Protection Y/N	Justification if data protection is claimed	Owner
B.5.1.2.1/02	Stry, J.J.	2000	Independent laboratory validation of method number DuPont-2392, "Analytical method for the determination of oxamyl and its oxime metabolite in soil using LC/MS analysis" DuPont Experimental Station DuPont-3738 GLP: Yes Published: No	N	Y	The study is necessary for the regulatory decision, conducted according to GLP	DuPont
B.5.1.2.16/06	[redacted]	2000b	Oxamyl 10L: Static, acute, 96-hour, (LC_{50}) test to rainbow trout, *Oncorhynchus mykiss* [redacted] DuPont-2910 GLP: Yes Published: No	Y	Y	The study is necessary for the regulatory decision, conducted according to GLP	DuPont
B.5.1.2.16/07	[redacted]	2000c	Oxamyl 10L: Static-renewal, acute, 96-hour, (LC_{50}) test to bluegill sunfish, *Lepomis macrochirus* [redacted] DuPont-2911 GLP: Yes Published: No	Y	Y	The study is necessary for the regulatory decision, conducted according to GLP	DuPont
B.5.1.2.16/08	Ward, T.J., Magazu, J.P., Boeri, R.L.	2000a	Oxamyl 10L: Acute, static-renewal, 48-hour EC_{50} to *Daphnia magna* T.R. Wilbury Laboratories, Inc. DuPont-2556 GLP: Yes Published: No	N	Y	The study is necessary for the regulatory decision, conducted according to GLP	DuPont

Data Requirement No., Reference No.	Author(s)	Year	Title Source Company Report No. GLP or GEP Status (where relevant) Published or not	Vertebrate study Y/N	Data Protection Y/N	Justification if data protection is claimed	Owner
B.5.1.2.1/03	Zietz, E.	2002	Field soil dissipation of oxamyl nematicide and insecticide applied as Vydate 10 L by means of drip irrigation to cucurbits in a greenhouse in Italy - Season 2000 Institut Fresenius Chemische und Biologische/Gmbh DuPont-4800 GLP: Yes Published: No	N	Y	The study is necessary for the regulatory decision, conducted according to GLP	DuPont