European Commission

VOLUME 3 – Annex B (AS)

- CHLOROTHALONIL -

B.7 Residue

Rapporteur Member State: The Netherlands

November 2017

Renewal Assessment Report and Proposed decision of the Netherlands prepared in the context of the possible approval of chlorothalonil under Regulation (EC) 1107/2009

Version history page

Date	Version history
2000 January 2001	DAR Addenda
May 2016	Draft Assessment Renewal Report (containing 'old' DAR, including the Addenda relevant for residues, only when relevant for the representative uses for the renewal)
August 2016	Renewal Assessment Report
August 2017	Updated RAR according to the comments in the Evaluation Table. Revisions are in yellow, except for typo's which are not marked.
September 2017	Updated RAR according to the EFSA Evaluation table and comments during Expert Pesticides Peer Review meeting # 164.

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Residue data, introduction with table of metabolites

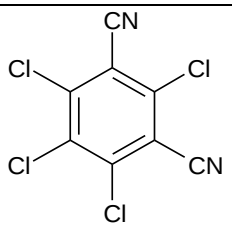
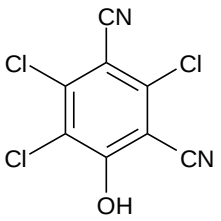
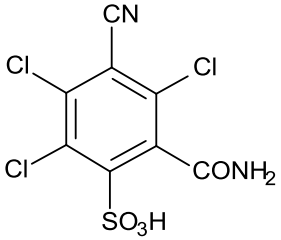
Chlorothalonil has been approved as a fungicide. The representative use ultimately supported in the original peer review was an outdoor foliar application on wheat. For the current renewal of chlorothalonil, the requested intended use patterns are on wheat, barley, tomatoes and potatoes.

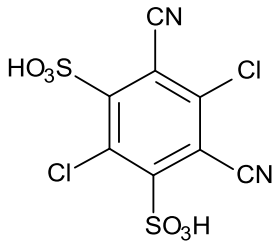
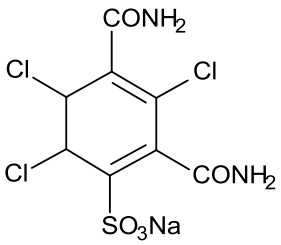
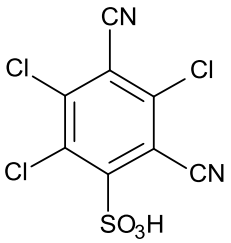
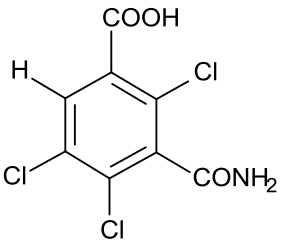
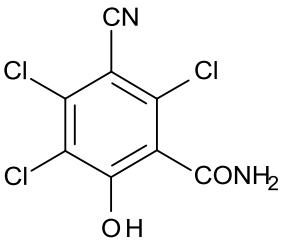
Every study summary in this RAR starts with a box, stating whether the study has already been evaluated elsewhere. When a study has already been evaluated for the original peer review (i.e. there is a box in which is mentioned 'previous evaluation: in DAR'), the study summary/evaluation is copied as such into this RAR.

During the initial peer review, many studies were stated as being GLP-studies. However, many of these studies were conducted at a time when GLP officially did not exist. Since these studies were conducted in line with the future GLP conditions, the GLP-statements are considered acceptable.

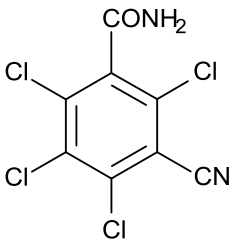
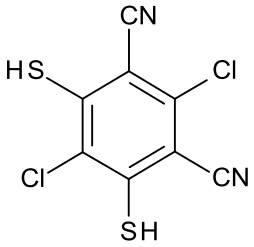
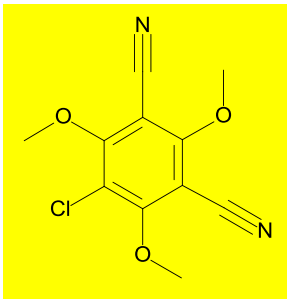
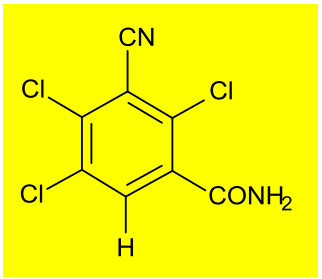
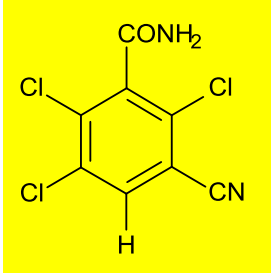
To improve the readability of the evaluation on the residue behaviour of chlorothalonil, the following table can be used, which contains the substances and metabolites with their accompanying codes, numbers or synonyms, relevant for the residues evaluation.

Table B.7-1: Substances and metabolites, relevant for the residues evaluation

Code Number (Synonyms)	**Description**	**Structure**
Chlorothalonil R044686 SDS-2787 1897-45-6	IUPAC name: 2,4,5,6-tetrachloro-isophtalonitrile	
R182281 SDS-3701 R1 Compound 2 C5 28343-61-5 CSCA105253	IUPAC name: 2,5,6-trichloro-4-hydroxyisophthalonitrile	
R417888 M12 VIS01 R6 Compound 10 U6 CSCC890840	IUPAC name: 2-amido-3,5,6-trichloro-4-cyanobenzenesulfonic acid	

Code Number (Synonyms)	**Description**	**Structure**
R418503 M13 R8 Compound 11 CSCA654600 SYN548708 (Na salt)[1]	IUPAC name: 2,5 dichloro-4,6 dicyano-benzene-1,3 disulfonic acid	
R471811 M4 R7 Compound 13 CSCA202566	IUPAC name: sodium 2,4-bis-amido-3,5,6-trichlorobenzenesulfonate	
R611553 R4 Compound 9 CSCC926922	IUPAC: 3,5,6-trichloro-2,4-dicyano- benzenesulfonic acid	
R611965 M5 SDS-46851 R14 Compound 4	IUPAC name: 3-amido-2,4,5-trichlorobenzoic acid	
R611968 M9 SDS-47525 R5	IUPAC name: 2,4,5-trichloro-3-cyano-6-hydroxybenzamide	

[1] used for gentox testing

Code Number (Synonyms)	Description	Structure
R613636 M14 SDS-19221 R2 Compound 3 CSCC548417	IUPAC name: 2,4,5,6-tetrachloro-3-cyanobenzamide	
R613800 C15	2,5-dichloro-4,6-bis(sulfanyl)benzene-1,3-dicarbonitrile	
SDS-3316	5-chloro-2,4,6-trimethoxyisophthalonitrile	
R613801 SDS 005473 MM230 C-1 CSAA509968 AGR359-025 CNIL/14	IUPAC name: 2,4,5-trichlorobenzene-1,3-dicarbonitrile	
R611966 SDS 47523 Compound 5	IUPAC name: 2,4,5-trichloro-3-cyano benzamide	
R611967 SDS 47524 Compound 6	IUPAC name: 2,5,6-trichloro-3-cyano benzamide	

B.7.1 Storage stability of residues

The stability of chlorothalonil residues during storage was investigated in various crops and animal products for the initial peer review. Furthermore, several additional studies have been submitted for the sake of the renewal of chlorothalonil.

B.7.1.1 Storage stability in various crops

Previous evaluation	In DAR Additional rows are added to the results tables in which the percentage recovery is calculated. The percentage recovery at day 0 is set at 100%, and the recoveries at the subsequent storage intervals are corrected for the recovery at day 0.
RMS remark	Not acceptable The study is not conducted conform guidance according to the applicant, and no further additional information has been submitted.

Characteristics

reference	:	see results individual crops	treatment	:	field study
type of study	:	storage stability study	rate	:	see study design
year of execution	:	1988 (all studies)	formulation	:	Bravo 720 (54.2% chlorothalonil)
test substance	:	chlorothalonil	GLP statement	:	yes
crop/commodity	:	Various, see study design	guidelines	:	not applicable

Study design

Crops were treated with Bravo 720 according to the specification listed below. Crops were harvested and samples were stored at -7°C or below until analysis. Analysis for chlorothalonil, SDS-3701 and SDS-46851 levels were performed following storage periods of at least 2 years.

Table 7.6.2.1 Crop treatment data and the references of the accessory reports.

Crop	Location	Application rate (kg a.i./ha)	Number of applications	Reference
Cherries	New York	3.4	10 [2]	Zeneca, King, 1993 (6.6.2 (study 1))
Wheat grain	Missouri	12.5	7	Zeneca, Kenyon, 1993 (6.6.2 (study 2))
Tomatoes	Ohio	2.5 + 7.5 [1]	8 + 1	Zeneca, Kenyon, 1993 (6.6.2 (study 3))
Cucumbers	Ohio	2.5 + 7.5 [1]	3 + 1	Zeneca, Wiedmann, 1993 (6.6.2 (study 4))
Potatoes	Wisconsin	12.5 [1]	15	Zeneca, Rose, 1993 (6.6.2 (study 5))

Crop	Location	Application rate (kg a.i./ha)	Number of applications	Reference
Carrots	Wisconsin	16.7 [1]	11	Zeneca, Rose, 1993 (6.6.2 (study 6))
Soybeans	Missouri	16.7[1]	8	Zeneca, Kenyon, 1993 (6.6.2 (study 7))
Celery	Wisconsin	2.5	16 [2]	Zeneca, King, 1993 (6.6.2 (study 8))
Almonds	California	3.3	13 [2]	Zeneca, Wiedmann, 1993 (6.6.2 (study 9))
Peanuts	Georgia	10.0-11.9 [1]	11	Zeneca, King, 1993 (6.6.2 (study 10))

1 exaggerated rate relative to the recommended use

2 exaggerated number relative to the recommended use

Results

The storage stability by storage period for the different crops is summarized in the tables below (tables 7.6.2.2-7.6.2.11):

Table 7.6.2.2 Storage stability by storage period for cherries

Time (days)	0	1	7	30	86	195	272	363	545	727	910	1091	1269	1457
Commodity	Chlorothalonil (mg/kg)[1]													
cherries	10.8	15.7	14.7	13.2	20.2	10.0	9.7	18.0	16.9	21.5	16.0	24.4	16.8	22.2
	Recovery (%)													
cherries	100	145	136	122	187	93	90	167	156	199	148	226	156	206

1 SDS-3701 levels were around or below 0.06 mg/kg throughout the study; SDS-46851 levels were close to or below the LOD of 0.03 mg/kg throughout the study.

Table 7.6.2.3 Storage stability by storage period for wheat grain

Time (days)	0	1	7	30	91	179	273	362	544	728	924	1095	1274	1468
Commodity	Chlorothalonil (mg/kg)[1]													
wheat grain	44.3	44.4	51.7	54.3	43.3	49.1	47.2	45.2	45.5	39.9	38.1[2]	41.6	47.9	41.5
	Recovery (%)													
wheat grain	100	100	117	123	98	111	107	102	103	90	86	94	108	94

1 SDS-3701 levels were 0.12-0.21 mg/kg throughout the study. SDS-46851 levels were all below the LOD of 0.03 mg/kg.

2 reextraction due to poor recoveries

Table 7.6.2.4 Storage stability by storage period for tomatoes

Time (days)	**0**	**1**	**7**	**30**	**92**	**174**	**274**	**363**	**546**	**723**	**916**	**1107**	**1275**	**1456**[1]
Commodity	**Chlorothalonil (mg/kg)**[1]													
tomatoes	8.8	10.8	15.5	10.3	10.4	11.2	9.5	9.6	10.7	8.0	11.4	10.4[2]	11.0	8.8
	Recovery (%)													
tomatoes	100	123	176	117	118	127	108	109	122	91	130	118	125	100

1 SDS-3701 levels were in general close to or below 0.06 mg/kg throughout the study. SDS-46851 levels were all below the LOD of 0.03 mg/kg.

2 reextraction due to recoveries > 200%

Table 7.6.2.5 Storage stability by storage period for cucumbers

Time (days)	**0**	**1**	**7**	**28**	**91**	**182**	**276**	**360**	**552**	**727**	**805**	**910**	**1098**	**1296**	**1463**
Commodity	**Chlorothalonil (mg/kg)**[1]														
cucumbers	0.91	1.25	2.32	1.42	1.32	1.56	1.31	0.60	2.15	0.39	1.59	1.07	1.76	1.06	1.23
	Recovery (%)														
cucumbers	100	137	255	156	145	171	144	66	236	43	175	118	193	116	135

1 SDS-3701 levels were all close to or below the LOD of 0.01 mg/kg; SDS-46851 were all at or below the LOD of 0.03 mg/kg.

Table 7.6.2.6 Storage stability by storage period for potatoes

Time (days)	**0**	**1**	**7**	**30**	**90**	**180**	**270**	**363**	**549**	**714**	**869**	**1099**	**1135**	**1292**	**1448**
Commodity	**Chlorothalonil (mg/kg)**[1]														
potatoes	1.75	2.18	1.47	1.81	1.32	2.73	1.13	1.83	0.42[2]	1.70	1.40	0.38	1.20	0.74	0.98
	Recovery (%)														
potatoes	100	125	84	103	75	156	65	105	24	97	80	22	69	42	56

1SDS-3701 levels generally ranged from 0.13 to 0.28 mg/kg throughout the study; SDS-46851 ranged from 0.11 to 0.23 mg/kg throughout the study.

2 residue level very low. Additional measurements on days 572, 577, 631 and 633 showed residues of 0.67, 0.66, 1.06 and 0.82 mg a.i/kg

Table 7.6.2.7 Storage stability by storage period for carrots

Time (days)	**0**	**1**	**7**	**33**	**90**	**180**	**271**	**363**	**546**	**729**	**894**	**1135**[1]	**1309**	**1463**
Commodity	**Chlorothalonil (mg/kg)**[1]													
carrots	1.84	1.11	1.55	1.02	1.00	1.41	1.32	1.22	1.41	1.78	0.93	1.63	1.07[2]	1.85
	Recovery (%)													
carrots	100	60	84	55	54	77	72	66	77	97	51	89	58	101

1 SDS-3701 levels ranged from 0.06 to 0.13 mg/kg throughout the study; SDS-46851 levels were generally close to or below the LOD of 0.03 mg/kg.

2 reextractions

Table 7.6.2.8 Storage stability by storage period for soybeans

Time (days)	**0**	**1**	**7**	**34**	**87**	**181**	**272**	**363**	**542**	**723**	**913**	**1092**	**1280**	**1498**
Commodity	**Chlorothalonil (mg/kg)**[1]													
soybeans	11.3	16.2	31.6	13.2	26.1	22.1	19.4	12.9	15.6	10.9	13.7	11.5	21.1	18.9
	Recovery (%)													
soybeans	100	143	280	117	231	196	172	114	138	96	121	102	187	167

1 SDS-3701 levels were close to or below 0.1 mg/kg throughout the study; SDS-46851 levels were all close to or below the LOD of 0.03 mg/kg.

Table 7.6.2.9 Storage stability by storage period for celery

Time (days)	**0**	**1**	**7**	**28**	**91**	**181**	**280**	**364**	**546**	**729**	**911**	**1114**	**1266**	**1459**
Commodity	**Chlorothalonil (mg/kg)**[1]													
celery	4.0	5.8	6.2	8.8	6.8	8.3	4.6	10.9	7.5	7.6	6.6	4.4	5.2	7.8
	Recovery (%)													
celery	100	145	155	220	170	208	115	273	188	190	165	110	130	195

1 SDS-3701 levels were generally close to or below 0.05 mg/kg throughout the study; SDS-46851 levels were generally below the LOD of 0.03 mg/kg.

Table 7.6.2.10 Storage stability by storage period for almonds

Time (days)	**0**	**1**	**7**	**28**	**99**	**182**	**276**	**378**	**553**	**730**
Commodity	**Chlorothalonil (mg/kg)**[1]									
almond nutmeats	0.24	0.34	0.18	0.17	0.13	0.14	0.11	0.11	0.26	0.09
almond hulls	55.8	37.8	40.1	53.6	70.0	60.8	62.1	53.2	54.7	71.6
	Recovery (%)									
almond nutmeats	100	142	75	71	54	58	46	46	108	38
almond hulls	100	68	72	96	125	109	111	95	98	128

1 SDS-3701 levels were all close to or below the LOD of 0.01 mg/kg in nutmeats and ranged from 0.4-0.7 mg/kg in hulls; SDS-46851 levels were generally below the LOD of 0.03 mg/kg in nutmeats and hulls.

Table 7.6.2.11 Storage stability by storage period for peanuts

Time (days)	**0**	**2**	**7**	**28**	**83**	**174**	**267**	**301**	**329/ 330**	**363**
Commodity	**Chlorothalonil (mg/kg)**[1]									
peanuts	13.0	12.7	12.9	15.4	14.8	13.3	8.3	8.1	7.3	6.7
	Recovery (%)									
peanuts	100	98	99	118	114	102	64	62	56	52

Time (days)	427	486/ 489	546	602/ 609	665	728/ 729	903	1133/ 1126	1254	1450
Commodity	**Chlorothalonil (mg/kg)**[1]									
peanuts (continued)	10.0	12.2	10.5	8.8	7.7	8.6	10.7	8.2	8.4	7.9
	Recovery (%)									
peanuts (continued)	77	94	81	68	59	66	82	63	65	61

1 SDS-3701 levels increased from between 0.06 to 0.1 mg/kg after short storage intervals to up to 0.5-0.6 mg/kg on the final analysis days; SDS-46851 levels ranged from 0.11 to 0.20 mg/kg at the beginning of the study and from 0.2 to 0.5 mg/kg at the end of the study.

Conclusions

Chlorothalonil levels appeared stable under frozen storage conditions (at -7°C or below) for at least 2 (almonds nutmeats and hulls) to 4 years (cherries, wheat, grain, tomatoes, cucumbers, carrots, soybeans and celery). In potatoes and peanuts, chlorothalonil levels tended to decrease upon long term storage, yet remained relatively stable during the first year and half year of storage, respectively. Relatively low and stable levels of SDS-3701 and SDS-46851 were detected in most commodities. Only in peanuts, SDS-3701 and SDS-46851 levels tended to increase upon prolonged storage, but the levels remained below 10% of the levels of chlorothalonil.

B.7.1.2 Storage stability in various crops

Previous evaluation	Submitted for the purpose of renewal
RMS remark	~~Acceptable for results with chlorothalonil, but not acceptable for results with SDS-3701.~~ Not acceptable During the expert meeting (Peer Review Meeting #164), the study was discussed and it was concluded that due to identified defeciencies, these data cannot be considered as acceptable for chlorothalonil and metabolite SDS-3701. Limitations paragraph has been updated accordingly.

Report:	K-CA 6.1/01. Anderson L and Chaggar S. (2007), Chlorothalonil (R44686) and R182281 (SDS3701): storage stability of field-incurred residues in homogenised crops stored deep frozen for up to two years. Syngenta, Jealott's Hill International Research Centre, Bracknell, United Kingdom. Syngenta Report Number T000559-06-REG. (Syngenta File No: R182281/0023).

Guidelines

Not stated but evaluated as compliant with testing guideline OECD Test No. 506 ~~meets the requirements of Guideline: Commission of the European Communities: Storage Stability of Residue Samples (SANCO 7032/V1/95 rev. 5 22/7/1997).~~

GLP

The study was carried out according to the principles of Good Laboratory Practice.

EXECUTIVE SUMMARY

Samples of cucumber, tomato, whole melon, carrot root, carrot top (foliage), whole orange, barley grain, barley straw and soya beans containing incurred residues of chlorothalonil and R182281 from supervised residue trials were stored under frozen conditions (≤-18° C) and analysed at intervals up to 24 months (five sampling points). An additional analysis after 27 months storage was conducted for barley straw and soya bean samples only. The limit of quantification (LOQ) for both chlorothalonil and R182281 was 0.01 mg/kg.

Residues of chlorothalonil were stable for at least 24 months in tomato, cucumber, melon (high water crop group), barley grain (high starch group), for at least 27 months in soya bean (high oil group) and for 12 months in barley straw when stored in the freezer at ≤-18°C.

There was no apparent degradation of R182281 in tomatoes, oranges or soya bean. Measured residues of R182281 increased in cucumber, melon, carrots (root and foliage) and barely (grain and straw), probably via transformation of chlorothalonil to R182281. However, some of the incurred residues were below the LOQ of the method (0.01 mg/kg) and therefore difficult to quantify with accuracy and precision.

~~The study demonstrated that field incurred residues of chlorothalonil and R182281 remained stable on storage when prepared without the use of dry ice or acid.~~

A1. Test Materials

The purity of the analytical standards used in this study is listed in Table 7.1.2-1.

Table 7.1.2-1: Purity of analytical standards

Analyte	Standard reference no.	Purity (wt. %)
Chlorothalonil	AMS 237/2	99
R 182281 (SDS-3701)	ASJ10209-02	100

A2. Test Commodity

The test commodities were mature crops obtained from field trials.

A3. Test Facilities

The field phase of this study was performed at various locations in Europe and the USA. The analytical phase was performed at Syngenta, Jealott's Hill International Research Centre, Bracknell, Berkshire, RG42 6EY, UK.

B. STUDY DESIGN AND METHODS

B1. Fortification and Storage of Samples

Samples of tomato (variety "Petula"), cucumber (variety "Tiria"), whole melon, (variety "Ferreol"), whole orange (variety "Navelina"), carrot root and tops (variety "Solo"), barley straw and grain (variety "Landi") were obtained from a residue study conducted in Switzerland. Soya beans (variety "Pioneer 93M80") were obtained from a residue study conducted in the USA. In all trials application of chlorothalonil was

made by foliar spray at a nominal rate of 4500 g a.s./ha (tomato, cucumber, melon, orange, carrot), 3000 g a.s./ha (barley) or 3430 g a.s./ha (soya bean). Mature harvest samples of tomato, cucumber, melon, orange, carrot and barley straw were homogenised by chopping (without dry ice); neither dry ice nor acid was used for the treated samples, while acid was used for the preparation of the control samples. Samples of barley grain and soya bean were not prepared. Sub-samples of each commodity were stored under frozen conditions (≤18°C) and analysed in triplicate at intervals up to 24 months (five sampling points). An additional analysis after 27 months storage was conducted for barley straw and soya bean. The initial (0 month) samples were analysed in triplicate immediately after sample preparation.

B2. Analytical Method

Two methods of analysis were used. Analytical method SOP RAM 365/02 was used for samples taken at 0, 3, 6 and 12 months. A full method description and validation data are presented in B.5.2.1 (K-CA 4.2/01-K-CA 4.2/07). The method involved extraction of the samples with acetone/5M sulphuric acid followed by centrifugation. An aliquot of the extract was taken, partitioned into toluene and then subjected to C8 solid phase extraction (SPE) clean up. Chlorothalonil and R182281 were determined by gas liquid chromatography with mass selective detection (GC-MSD) using three fragment ions (m/z > 100) for quantification. R182281 was derivatised with trimethylsilyl diazomethane to produce methyl R182281 (R619464). The LOQ was 0.01 mg/kg for both analytes.

Analytical method SOP RAM 365/02 was replaced by method GRM005.01A for samples analysed after 24 and 27 months. A full method description and validation data are presented in B.5.2.1 (K-CA 4.2/01-K-CA 4.2/07). The method involved extraction of the samples with acetone/5M sulphuric acid followed by centrifugation. For chlorothalonil aliquots of the extracts were diluted with water and subjected to SPE clean up, before analysis by GC-MSD. For R18221, aliquots of the sample extracts were diluted with acetonitrile: water and analysed by liquid chromatography tandem mass spectrometry (LC-MS/MS). The LOQ was 0.01 mg/kg for both analytes.

Procedural recoveries were determined using freshly fortified samples at each interval. Samples were fortified with chlorothalonil at 0.1 – 25 mg/kg or R182281 at 0.01 – 1.5 mg/kg. Individual recoveries, mean recoveries and %RSD are summarised in Table 7.1.2-2 (chlorothalonil) and Table 7.1.2-3 (R182281).

Table 7.1.2-2: Summary of procedural recoveries for chlorothalonil in crops

Crop	Fortification level (mg/kg)	Recovery (%)	Mean recovery (%)	RSD (%)
Tomato	1	81	89	12
	3	103, 101, 73, 75, 88, 91, 88, 87		
	5	98		
Cucumber	1	99	95	8.8
	2	98, 101, 92, 89, 94, 92, 85, 86		
	5	113		
Melon	0.1	90	98	7.8
	1	99, 97, 98, 113, 111, 93, 92, 96, 95		

Crop	Fortification level (mg/kg)	Recovery (%)	Mean recovery (%)	RSD (%)
Orange	0.1	79	95	6.6
	1	94		
	10	99, 101, 96, 93, 95, 99, 95, 100		
Carrot root	0.1	95	95	4.3
	1	99, 91, 91, 95, 103, 91, 97, 93, 91		
Carrot top	1	92	96	5.0
	5	94		
	25	103, 100, 89, 92, 92, 97, 97, 102		
Barley straw	5	100	96	5.1
	10	103		
	20	96, 104, 96, 99, 96, 94, 92, 99, 89, 89		
Barley grain	0.1	96, 77, 88	90	6.5
	1	86, 87, 94, 92, 95, 95, 89		
Soya bean	0.1	88	80	8.5
	1	81, 77, 93		
	1.5	79, 67, 76, 79		
	2.0	74, 76, 84, 82		

Recovery data were obtained by using method SOP RAM 365/02 and method GRM005.01A.

Table 7.1.2-3: Summary of procedural recoveries for R18221 in crops

Crop	Fortification level (mg/kg)	Recovery (%)	Mean recovery (%)	RSD (%)
Tomato	0.01	83, 101, 92, 87, 69, 72, 122, 94, 99, 101	92	16
	0.1	80		
	0.05	102		
Cucumber	0.01	107, 113, 105, 101, 97, 105, 101, 82, 100, 96	101	7.6
	0.1	97		
	0.05	105		
Melon	0.01	87, 107, 100, 106, 103, 109, 96, 110, 103	101	8.7
	0.1	85		
Orange	0.01	90	98	7.3
	0.02	97, 92, 114, 102, 102, 99, 97, 91		
	0.1	94		
Carrot root	0.01	83	96	10
	0.03	100, 108		
	0.04	99, 91, 101, 103, 101, 96		
	0.1	77		
Carrot top	0.01	108	107	9.4
	0.1	94		
	0.25	124, 128, 100, 104		

Crop	Fortification level (mg/kg)	Recovery (%)	Mean recovery (%)	RSD (%)
	0.3	100, 99, 108, 111, 108, 102		
Barley straw	0.1	108	105	6.8
	1	102, 99, 97, 106, 100, 109, 100, 98, 105		
	1.5	120, 115		
Barley grain	0.01	93, 86, 101, 96, 101	96	7.7
	0.05	99, 88, 106, 106		
	0.1	88		
Soya bean	0.01	76, 79	98	12
	0.02	112, 108, 83, 98, 94, 92, 101, 117		
	0.03	103, 106		
	0.1	99, 101		

Recovery data were obtained by using method SOP RAM 365/02 and method GRM005.01A

Storage Stability of Residues

The recoveries of chlorothalonil and R182281 in crops stored at ≤-18°C are summarised in Table 7.1.2-4 and Table 7.1.2-5, respectively. The results are shown for both corrected as well as not corrected for freshly fortified recoveries.

Table 7.1.2-4: Storage Intervals and % Recoveries of Chlorothalonil in Various Crops

Interval		Uncorrected Residue (mg kg^{-1})	Mean Uncorrected Residue (A) (mg kg^{-1})	Corrected Residue (mg kg^{-1})	Mean Corrected Residue (B) (mg kg^{-1})	Mean Procedural Recovery (%)	Mean Recovered Uncorrected Residue (C=A/0 time A x100%)	Mean Recovered Corrected Residue (D=B/0 time B x100%)
Months (Nominal)	Days (Actual)							
Tomato								
Zero time	0	2.8, 2.7, 3.0	2.8	3.1, 3.0, 3.3	3.1	89	100	100
3	98	3.0, 2.7, 3.1	3.0	3.0, 2.7, 3.1	2.9	102	106	92
6	211	1.8, 1.9, 2.1	1.9	2.4, 2.6, 2.9	2.6	73	**69**	84
12	385	2.6, 2.7, 2.5	2.6	2.9, 3.0, 2.8	2.9	90	93	92
24	786	2.5, 2.5, 2.3	2.5	2.8, 2.9, 2.7	2.8	87	88	89
Cucumber								
Zero time	7	1.8, 2.3, 1.5	1.9	1.7, 2.2, 1.5	1.8	106	100	100
3	104	1.6, 1.6, 1.6	1.6	1.6, 1.7, 1.6	1.6	99	86	92
6	209	1.4, 1.5, 1.5	1.5	1.5, 1.7, 1.6	1.6	90	78	91
12	383	1.4, 1.5, 1.4	1.4	1.6, 1.6, 1.5	1.5	93	76	87
24	784	1.6, 1.3, 1.3	1.4	1.9, 1.6, 1.5	1.7	85	76	94
Melon								
Zero time	0	0.57, 0.65, 0.65, 0.79, 0.62	0.66	0.60, 0.68, 0.69, 0.83, 0.66	0.69	95	100	100
3	99	0.55, 0.52, 1.02	0.70	0.57, 0.54, 1.04	0.72	97	106	104
6	216	0.70, 0.66, 0.71	0.69	0.62, 0.59, 0.63	0.61	113	104	89
12	378	0.71, 0.41, 0.51	0.54	0.76, 0.44, 0.55	0.58	93	83	84
24	779	0.69, 0.53, 0.80	0.68	0.72, 0.56, 0.84	0.70	96	103	102

Interval		Uncorrected Residue	Mean Uncorrected Residue (A)	Corrected Residue	Mean Corrected Residue (B)	Mean Procedural Recovery	Mean Recovered Uncorrected Residue	Mean Recovered Corrected Residue
Months (Nominal)	**Days (Actual)**	**(mg kg^{-1})**	**(mg kg^{-1})**	**(mg kg^{-1})**	**(mg kg^{-1})**	**(%)**	**(C=A/0 time A x100%)**	**(D=B/0 time B x100%)**
Orange								
Zero time	0	11, 8.2, 8.9, 11, 11	10	13, 9.5, 10, 13, 13	12	87	100	100
3	102	8.0, 8.7, 8.6	8.4	8.0, 8.7, 8.6	8.4	100	84	73
6	223	7.6, 8.1, 8.3	8.0	8.1, 8.6, 8.8	8.5	95	80	73
12	404	8.5, 8.6, 8.2	8.4	8.8, 8.9, 8.4	8.7	97	84	75
24	788	8.0, 8.5, 7.8	8.1	8.2, 8.7, 8.0	8.3	97	81	72
Carrot Roots								
Zero time	0	0.73, 0.69, 0.70, 0.71, 0.64	0.69	0.75, 0.71, 0.72, 0.73, 0.66	0.71	97	100	100
3	97	0.67, 0.62, 0.74	0.68	0.73, 0.69, 0.81	0.74	91	98	104
6	216	0.60, 0.62, 0.57	0.60	0.61, 0.63, 0.57	0.60	99	86	84
12	405	0.60, 0.61, 0.60	0.60	0.64, 0.65, 0.64	0.64	94	87	90
24	781	0.50, 0.52, 0.53	0.51	0.54, 0.56, 0.57	0.56	92	74	78
Carrot Tops								
Zero time	0	101, 85, 94, 92, 87	92	110, 92, 101, 99, 94	99	93	100	100
3	92	92, 89, 87	89	91, 88, 86	88	101	97	89
6	211	79, 80, 73	77	87, 88, 81	85	91	84	86
12	400	90, 101, 94	95	95, 107, 99	100	95	103	101
24	784	77, 77, 73	75	77, 77, 73	76	100	82	76
Barley Straw								
Zero time	0	25, 25, 28, 24, 26	26	25, 25, 28, 24, 26	25	101	100	100
3	104	21, 21, 20	20	21, 21, 20	21	100	80	81
6	209	18, 18, 20	18	18, 18, 21	19	97	72	75
12	406	19, 18, 17	18	20, 19, 18	19	95	70	74
24	790	15, 15, 16	15	15, 16, 17	16	95	**59**	**63**
27	840	13, 14, 15	14	14, 15, 16	15	89	**53**	**60**
Barley Grain								
Zero time	0	0.71, 0.80, 0.73, 0.74, 0.83	0.76	0.78, 0.87, 0.81, 0.82, 0.91	0.84	91	100	100
3	92	0.82, 0.82, 0.88	0.84	0.99, 0.99, 1.1	1.0	83	110	**121**
6	203	0.67, 0.80, 0.65	0.71	0.74, 0.89, 0.72	0.79	90	93	94
12	391	0.79, 0.76, 0.77	0.77	0.84, 0.81, 0.82	0.82	94	101	98
24	770	0.81, 0.58, 0.85	0.74	0.88, 0.63, 0.92	0.81	92	98	97
Soya Bean								
Zero time	0	1.4, 1.4, 1.3, 1.3, 1.4	1.4	1.7, 1.7, 1.6, 1.6, 1.6	1.6	84	100	100
3	91	1.4, 1.3, 1.4	1.4	1.9, 1.8, 1.9	1.9	73	100	114
6	202	1.5, 1.6, 1.6	1.6	1.8, 1.9, 1.8	1.9	85	115	114
12	390	1.5, 1.4, 1.4	1.5	2.0, 1.9, 1.9	1.9	75	106	119
24	770	1.2, 0.69, 0.84	0.91	1.5, 0.82, 1.0	1.1	83	**68**	**68**
27	810	0.97, 1.2, 1.5	1.2	1.3, 1.5, 1.9	1.6	77	88	96

Values in bold are outside the acceptable range of 70-120% recovery.

Table 7.1.2-5: Storage Intervals and % Recoveries of R182281 in Various Crops

Interval		Uncorrected Residue	Mean Uncorrected Residue (A)	Corrected Residue	Mean Corrected Residue (B)	Mean Procedural Recovery	Mean Recovered Uncorrected Residue	Mean Recovered Corrected Residue
Months (Nominal)	Days (Actual)	(mg kg^{-1})	(mg kg^{-1})	(mg kg^{-1})	(mg kg^{-1})	(%)	(C=A/0 time A x100%)	(D=B/0 time B x100%)
Tomato								
Zero time	0	0.008, 0.008, 0.007, 0.009, 0.007	0.008	0.01, 0.009, 0.008, 0.01, 0.009	0.009	82	100	100
0 day repeat	9	0.008, 0.009, 0.010		0.008, 0.009, 0.01		101		
3	98	0.008, 0.009 (0.019**)	0.008	0.008, 0.01 **	0.009	90	102	101
6	211	0.006, 0.007, 0.007	0.007	0.009, 0.01, 0.01	0.01	71	83	105
12	385	0.008, 0.01, 0.009	0.009	0.007, 0.01, 0.008	0.009	108	113	93
24	786	0.008, 0.009, 0.01	0.009	0.008, 0.009, 0.01	0.009	100	110	98
Cucumber								
Zero time	0	0.004, 0.002, 0.002, 0.003, 0.004	0.004	0.004, 0.002, 0.002, 0.003, 0.004	0.003	102	100	100
0 day repeat	7	0.003, 0.004, 0.005		0.003, 0.004, 0.005		109		
3	104	0.008, 0.010, 0.010	0.009	0.008, 0.010, 0.009	0.009	103	**264**	**270**
6	209	0.014, 0.017, 0.020	0.017	0.014, 0.016, 0.019	0.017	101	**482**	**500**
12	383	0.021, 0.016, 0.021	0.019	0.023, 0.018, 0.023	0.021	91	**556**	**638**
24	784	0.026, 0.024, 0.025	0.025	0.027, 0.024, 0.026	0.026	98	**714**	**766**
Melon								
Zero time	0	0.003 0.005, 0.005, 0.005, 0.003	0.004	0.003, 0.005, 0.005, 0.005, 0.004	0.005	86	100	100
3	99	0.004, 0.003, 0.005	0.004	0.004, 0.003, 0.005	0.004	103	97	80
6	216	0.005, 0.005, 0.006	0.006	0.005, 0.005, 0.006	0.005	105	**140**	115
12	378	0.005, 0.003, 0.006	0.004	0.004, 0.003, 0.006	0.004	103	111	93
24	779	0.009, 0.008, 0.008	0.009	0.009, 0.008, 0.008	0.008	106	**220**	**178**
Orange								
Zero time	0	0.024, 0.014, 0.015, 0.028, 0.029	0.022	0.026, 0.015, 0.016, 0.03, 0.03	0.024	92	100	100
3	116	0.022, 0.021, 0.028	0.024	0.023, 0.022, 0.030	0.025	94	109	106
6	223	0.020, 0.019, 0.019	0.019	0.018, 0.017, 0.017	0.018	108	86	73
12	404	0.016, 0.020, 0.018	0.018	0.016, 0.020, 0.018	0.018	100	82	76
24	788	0.016, 0.017, 0.018	0.017	0.017, 0.018, 0.019	0.018	94	77	75

Interval		Uncorrected Residue	Mean Uncorrected Residue (A)	Corrected Residue	Mean Corrected Residue (B)	Mean Procedural Recovery	Mean Recovered Uncorrected Residue	Mean Recovered Corrected Residue
Months (Nominal)	**Days (Actual)**	**(mg kg^{-1})**	**(mg kg^{-1})**	**(mg kg^{-1})**	**(mg kg^{-1})**	**(%)**	**(C=A/0 time A x100%)**	**(D=B/0 time B x100%)**
Carrot Roots								
Zero time	0	0.033, 0.030, 0.030, 0.033, 0.030	0.031	0.041, 0.037, 0.038, 0.041, 0.037	0.039	80	100	100
3	97	0.048, 0.042, 0.043	0.044	0.046, 0.041, 0.041	0.043	104	**143**	110
6	216	0.050, 0.047, 0.047	0.048	0.052, 0.050, 0.050	0.051	95	**154**	**131**
12	405	0.059, 0.063, 0.061	0.061	0.058, 0.062, 0.060	0.060	102	**196**	**154**
24	781	0.084, 0.076, 0.081	0.080	0.085, 0.077, 0.082	0.081	99	**259**	**209**
Carrot Tops								
Zero time	0	0.28, 0.24, 0.25, 0.26, 0.26	0.26	0.28, 0.23, 0.25, 0.25, 0.26	0.25	101	100	100
3	92	0.45, 0.41, 0.42	0.40	0.36, 0.23, 0.33	0.34	126	**157**	**134**
3*	104	0.37, 0.42, 0.36		0.36, 0.41, 0.35		102		
6	211	0.42, 0.38, 0.38	0.39	0.42, 0.39, 0.39	0.40	99	**153**	**156**
12	400	0.50, 0.49, 0.51	0.50	0.46, 0.45, 0.46	0.46	108	**194**	**180**
24	784	0.60, 0.70, 0.58	0.62	0.57, 0.66, 0.55	0.60	105	**243**	**234**
Barley Straw								
Zero time	0	1.2, 1.2, 1.2, 1.1, 1.2	1.2	1.1, 1.1, 1.2, 1.1, 1.2	1.1	105	100	100
3	104	1.3, 1.4, 1.3	1.3	1.3, 1.4, 1.3	1.3	98	111	118
6	209	1.4, 1.5, 1.4	1.4	1.3, 1.5, 1.4	1.4	103	**121**	**123**
12	406	1.6, 1.6, 1.7	1.6	1.5, 1.6, 1.6	1.6	104	**138**	**138**
24	790	1.9, 2.0, 2.0	2.0	1.8, 2.0, 2.0	1.9	102	**166**	**172**
27	840	1.3, 1.0, 2.0	1.4	1.1, 0.8, 1.7	1.2	117	119	107
Barley Grain								
Zero time	0	0.052, 0.053, 0.053, 0.056, 0.057	0.054	0.058, 0.058, 0.058, 0.062, 0.063	0.060	90	100	100
3	92	0.066, 0.075, 0.072	0.071	0.070, 0.080, 0.077	0.076	94	**131**	**126**
6	203	0.114, 0.124, 0.112	0.117	0.107, 0.117, 0.106	0.110	106	**215**	**184**
12	391	0.067, 0.069, 0.068	0.068	0.071, 0.073, 0.073	0.072	94	**125**	**121**
24	770	0.089, 0.093, 0.097	0.093	0.091, 0.095, 0.099	0.095	98	**172**	**159**
Soya Bean								
Zero time	0	0.024, 0.021, 0.022,	0.028	0.028, 0.024, 0.026,	0.031	87	100	100
0 day repeat*	0	0.036, 0.032, 0.035, 0.031, 0.032		0.040, 0.036, 0.039, 0.034, 0.036		90		
3	91	0.022, 0.020, 0.015	0.019	0.021, 0.019, 0.014	0.018	105	**68**	**58**
6	202	0.026, 0.029, 0.028	0.027	0.023, 0.026, 0.025	0.025	110	98	79
12	390	0.018, 0.024, 0.020	0.021	0.020, 0.026, 0.022	0.023	91	74	72

Interval		Uncorrected Residue (mg kg^{-1})	Mean Uncorrected Residue (A) (mg kg^{-1})	Corrected Residue (mg kg^{-1})	Mean Corrected Residue (B) (mg kg^{-1})	Mean Procedural Recovery (%)	Mean Recovered Uncorrected Residue (C=A/0 time A x100%)	Mean Recovered Corrected Residue (D=B/0 time B x100%)
Months (Nominal)	Days (Actual)							
24	770	0.016, 0.014, 0.015	0.015	0.017, 0.015, 0.016	0.016	93	**53**	**51**
27	810	0.022, 0.022, 0.029	0.024	0.020, 0.020, 0.027	0.022	109	88	72

*Reanalysis of the extracts was carried out to confirm the low level residue in the soya bean control. Further analysis of the controls (RDP 05-0606) concluded this could have been due to laboratory contamination. A low level, persistent residue was determined therefore matrix standards and procedural recoveries were corrected for this apparent control residue The mean result from all the 0 day analyses is used for the subsequent calculations.
** an outlier, identified by Grubbs Test, has been omitted from the table.
* reanalysis to confirm previous results as batch recovery was high.
Values in bold are outside the acceptable range of 70-120% recovery.

III. CONCLUSIONS

Residues of chlorothalonil were stable for at least 24 months in tomato, cucumber, melon (high water crop group) and barley grain (high starch group), for at least 27 months in soya bean (high oil group) and for 12 months in barley straw when stored in the freezer at ≤-18°C.

There was no apparent degradation of R182281 in tomatoes, oranges or soya bean. Measured residues of R182281 increased significantly in cucumber, melon, carrots (root and foliage) and barley (grain and straw), probably via transformation of chlorothalonil to R182281. The study design uses field incurred residues of parent and metabolite. Residues of R182281 are very low in comparison to parent residues. Therefore, a small amount of parent degradation would have a large impact on the recovery of R182281. ~~However~~ However, since this process has not been observed in all samples, but only in part of the data, it was concluded by the expert meeting, that the data is not conclusive for the metabolite. Furthermore, some of the incurred residues (in tomatoes, cucumbers, melons) were below the LOQ of the method (0.01 mg/kg) and therefore difficult to quantify with accuracy and precision.

The applicant considers the study to be no longer acceptable, however, the RMS considers that the study demonstrated that field incurred residues of chlorothalonil ~~and R182281~~ remained stable during freezer storage when prepared without the use of dry ice or acid.

Limitations

During the expert meeting it was concluded that the study cannot be considered as standalone study and the data is considered not acceptable. It was noted that only the mean recoveries have been reported for all commodities and it is not clear how the means were calculated. It was also not clear if the analysis were made simultaneously on the same sample or for each compound separately, which is relevant in the view of high rates of metabolite SDS-3701 is several samples. Moreover, for the metabolite SDS-3701 recoveries of the residues were not consistent and highy exaggerated, which makes the data not reliable for both compounds.

B.7.1.3 Storage stability in various crops

Previous evaluation	Submitted for the purpose of renewal

RMS remark	Acceptable, however, some deficiencies of the study have been discussed during the peer review meeting

Report:	K-CA 6.1/02. Anderson L. (2007), Chlorothalonil (R44686) and R182281 (SDS3701): storage stability in various crops prepared in acid and stored deep frozen for up to two years. Syngenta, Jealott's Hill International Research Centre, Bracknell, United Kingdom. Syngenta Report Number T005407-04-REG. (Syngenta File No: R44686/4298).

Guidelines

Not stated but evaluated as compliant with testing guideline OECD Test No. 506 ~~meets the requirements of Guideline: Commission of the European Communities: Storage Stability of Residue Samples (SANCO 7032/V1/95 rev. 5 22/7/1997).~~

GLP

The study was carried out according to the principles of Good Laboratory Practice.

EXECUTIVE SUMMARY

Samples of strawberry, onion, broccoli, cauliflower, cucumber, tomato, melon, grape, Brussels sprout, cabbage, French bean, pea, apple, potato, carrot root, leek, plum, sugar beet root, olive and banana that had been homogenised in the presence of acid (10% v/w sulphuric acid) were fortified at 1 mg/kg with chlorothalonil and R182281. Triplicate samples were stored under frozen conditions (≤-18°C) and analysed at intervals up to 24 months (five sampling points). The LOQ for both chlorothalonil and R182281 was 0.01 mg/kg.

There was no significant decrease (>30% compared to the zero time value) in the levels of chlorothalonil in any crops over 24 months when stored at ≤-18°C.

Residues of chlorothalonil were found to be stable in crops representing the high water (onion, broccoli, cauliflower, cucumber, melon, tomato, Brussels sprout, cabbage, French bean, pea, apple, leek, plum, sugar beet root and banana), high oil (olives), high starch (potato, carrot root) and high acid (strawberry, grapes) crop groups for at least 24 months when stored in the freezer at ≤-18°C.

Residues of R182281 were stable for at least 24 months in broccoli, cauliflower, cucumber, melon, tomato, Brussels sprout, cabbage, French bean, pea, apple, leek, plum, sugar beet root and banana (high water crop group), olive (high oil crop group), potato, carrot root (high starch crop group) and strawberry (high acid crop group) when stored in the freezer at ≤-18°C. Residues of R182281 were found to be stable in grapes (high acid) for up to 3 months and were stable for less than 3 months in onions (high water) when stored frozen at ≤-18°C.

The study demonstrated that residues of chlorothalonil and R182281 remained stable during freezer storage when prepared in the presence of acid.

A1. Test Materials

The purity of the analytical standards used in this study is listed in Table 7.1.3-1.

Table 7.1.3-1: Purity of analytical standards

Analyte	Standard reference no.	Purity (wt. %)
Chlorothalonil	AMS 237/2	99
R 182281 (SDS-3701)	ASJ10209-02	100

A2. Test Commodity

The test commodities were strawberry, onion, broccoli, cauliflower, cucumber, tomato, melon (whole fruit), grape, Brussels sprout, cabbage, French bean, pea, apple, potato, carrot root, leek, plum (fruit minus stone), sugar beet root, olive (fruit minus stone) and banana (whole fruit). Samples were purchased from a local supermarket with the exception of the sugar beet root samples which were from the Syngenta control sample collection.

A3. Test Facilities

This study was performed at Syngenta, Jealott's Hill International Research Centre, Bracknell, Berkshire, RG42 6EY, UK.

B. STUDY DESIGN AND METHODS

B1. Fortification and Storage of Samples

Samples were partially homogenised, then 1M sulphuric acid was added at a ratio of 10% v/w before homogenisation was continued. The homogenised samples were fortified at 1.0 mg/kg with chlorothalonil and R182281 in acetone. Triplicate samples were stored under frozen conditions (≤-18°C) and analysed at intervals up to 24 months (five sampling points). Control samples were analysed at zero time and at each time point to ensure that no residues of chlorothalonil or R182281 were present at levels above 30% of the LOQ.

B2. Analytical Method

Analysis of the samples was performed according to analytical methods SOP RAM 365/01 and SOP 365/02 at intervals of 0, 3, 6, 12 and 24 months. RAM 365/02 was issued to correct a typographical error in method RAM 365/01 therefore the methods can be considered identical. A full method description and validation data are presented in B.5.2.1 (K-CA 4.2/01-K-CA 4.2/07). The method involved extraction of the samples with acetone/5M sulphuric acid followed by centrifugation. An aliquot of the extract was taken, partitioned into toluene and then subjected to SPE clean up. Chlorothalonil and R182281 were determined by GC-MSD using three fragment ions (m/z > 100) for quantification. R182281 was derivatised with trimethylsilyl diazomethane to produce methyl R182281 (R619464). The LOQ was 0.01 mg/kg for both analytes.

II. RESULTS AND DISCUSSION

Method Validation

Procedural recoveries were determined using freshly fortified samples at each interval. Samples were fortified with chlorothalonil and R182281 at 1.0 mg/kg. Individual recoveries, mean recoveries and %RSD

are summarised in Table 7.1.3-2 (chlorothalonil) and Table 7.1.3-3 (R182281).

Table 7.1.3-2: Summary of procedural recoveries for chlorothalonil in crops

Crop	Fortification level (mg/kg)	Recovery (%)	Mean recovery (%)	RSD (%)
Strawberry	1.0	114, 100, 100, 101, 100, 102, 100, 103, 102, 100	102	4.1
Onion	1.0	92, 96, 100, 109, 95, 92, 96, 95, 103, 106, 89, 95	97	6.1
Broccoli	1.0	89, 90, 93, 92, 101, 99, 101, 103, 85, 88	94	6.8
Cauliflower	1.0	83, 91, 91, 97, 106, 100, 99, 101, 94, 92	95	6.9
Cucumber	1.0	105, 113, 96, 98, 106, 105, 102, 101, 91, 92	101	6.8
Melon	1.0	81, 88, 89, 91, 103, 104, 105, 105, 100, 93	96	9.0
Tomato	1.0	106, 110, 79, 91, 108, 108, 98, 101, 85, 92	98	11.0
Grape	1.0	90, 83, 87, 87, 107, 106, 104, 105, 89, 91	95	9.9
Brussels sprout	1.0	91, 92, 82, 96, 105, 112, 101, 106, 107, 106, 95, 91	99	9.0
Cabbage	1.0	105, 104, 93, 96, 103, 105, 101, 104, 98, 98	101	4.2
Green bean	1.0	99, 103, 94, 99, 109, 103, 101, 103, 96, 105	101	4.4
Pea	1.0	93, 91, 91, 95, 102, 99, 106, 113, 85, 99, 88, 93	96	8.3
Apple	1.0	98, 97, 92, 93, 100, 102, 104, 102, 93, 93	98	4.6
Potato	1.0	114, 110, 115, 109, 96, 94, 108, 107, 86, 81	102	11.7
Carrot root	1.0	94, 98, 91, 95, 102, 102, 97, 99, 92, 87	96	5.2
Leek	1.0	88, 87, 99, 100, 92, 89, 103, 107, 97, 102	96	7.4
Plum	1.0	92, 92, 92, 91, 112, 112, 106, 101, 78, 88	96	11.4
Banana (whole fruit)	1.0	98, 91, 98, 108, 102, 104, 109, 106, 75, 86	98	5.5
Sugar beet root	1.0	95, 97, 89, 94, 101, 98, 94, 109, 101, 99	98	11.2
Olive	1.0	101, 95, 83, 87, 108, 105, 93, 96, 91, 93	95	8.2

Table 7.1.3-3: Summary of procedural recoveries for R182281 in crops

Crop	Fortification level (mg/kg)	Recovery (%)	Mean recovery (%)	RSD (%)
Strawberry	1.0	123, 112, 98, 101, 99, 100, 91, 91, 98, 100	101	9.5
Onion	1.0	117, 116, 102, 105, 102, 95, 105, 109, 91, 94, 108, 112	105	8.0
Broccoli	1.0	114, 110, 113, 109, 103, 95, 96, 96, 97, 102	104	7.3
Cauliflower	1.0	100, 110, 93, 108, 105, 109, 100, 101, 107, 102	104	5.1
Cucumber	1.0	104, 108, 102, 83, 105, 102, 94, 99, 108, 101	101	7.5
Melon	1.0	87, 90, 90, 94, 104, 102, 99, 100, 103, 100	97	6.3
Tomato	1.0	101, 121, 90, 85, 99, 95, 102, 101, 109, 105, 91, 94, 94, 88	98	9.6
Grape	1.0	97, 89, 91, 89, 100, 95, 95, 98, 104, 105	96	5.9
Brussels sprout	1.0	101, 104, 86, 97, 102, 97, 96, 96, 105, 108	99	6.5
Cabbage	1.0	111, 108, 95, 96, 108, 110, 96, 96, 99, 99	102	6.6

Crop	Fortification level (mg/kg)	Recovery (%)	Mean recovery (%)	RSD (%)
Green bean	1.0	101, 89, 104, 109, 114, 104, 97, 94, 77, 81	97	12.2
Pea	1.0	95, 95, 98, 102, 113, 103, 103, 103, 104, 107	102	5.2
Apple	1.0	107, 109, 79, 78, 99, 97, 96, 96, 89, 87	94	11.3
Potato	1.0	108, 102, 106, 97, 110, 102, 97, 94, 95, 82	99	8.3
Carrot root	1.0	102, 102, 92, 93, 106, 103, 99, 94, 93, 91	98	5.6
Leek	1.0	93, 91, 103, 101, 100, 94, 97, 99, 92, 90	96	4.8
Plum	1.0	80, 82, 90, 87, 107, 106, 91, 95, 80, 81, 104, 107	92	11.7
Banana (whole fruit)	1.0	88, 79, 106, 106, 109, 105, 95, 94, 74, 72	98	3.5
Sugar beet root	1.0	94, 99, 97, 97, 105, 100, 93, 96, 98, 97	93	15.3
Olive	1.0	98, 93, 109, 111, 112, 102, 97, 95, 91, 82	99	9.7

Storage Stability of Residues

The recoveries of chlorothalonil and R182281 in various crops stored at ≤ -18°C are summarised in Table 7.1.3-4 and Table 7.1.3-5, respectively. The results are not corrected for freshly fortified recoveries.

Table 7.1.3-4: Freezer storage stability for chlorothalonil at 1.0 mg/kg in crops

Sampling interval (nominal months)	Sampling interval (actual days)	Uncorrected residue (mg/kg)	Mean uncorrected residue (mg/kg)	Mean procedural recovery (%)	Mean recovered uncorrected residue (%), day 0 set at 100 / not corrected for day 0
Strawberry					
0	0	1.06, 0.97, 1.03	1.02	107	100 / 102
3	93	1.02, 1.00, 1.00	1.00	101	99 / 100
6	184	1.00, 0.96, 0.87	0.94	101	93 / 94
12	408	0.88, 0.99, 0.95	0.94	102	92 / 94
24	749	1.00, 0.96, 0.95	0.97	101	95 / 97
Onion					
0	0	0.92, 1.05, 0.94	0.97	94	100 / 97
3	92, 96	0.77, 0.78, 0.72, 0.78, 0.82, 0.70	0.76	99	78 / 76
6	187	0.86, 0.83, 0.82	0.84	95	86 / 84
12	408	0.94, 0.92, 0.91	0.92	104	95 / 92
24	749	0.87, 0.91, 0.90	0.89	92	92 / 89
Broccoli					
0	0	0.86, 0.86, 1.02	0.92	90	100 / 92
3	95	1.09, 1.01, 0.92	1.01	92	110 / 101
6	187	0.91, 0.89, 0.84	0.88	100	96 / 88
12	410	0.96, 0.96, 0.92	0.95	102	103 / 95

Sampling interval (nominal months)	Sampling interval (actual days)	Uncorrected residue (mg/kg)	Mean uncorrected residue (mg/kg)	Mean procedural recovery (%)	Mean recovered uncorrected residue (%), day 0 set at 100 / not corrected for day 0
24	748	0.83, 0.80, 0.83	0.82	87	89 / 82
Cauliflower					
0	0	0.96, 0.79, 0.93	0.89	87	100 / 89
3	105	0.96, 0.90, 1.00	0.96	94	107 / 96
6	189	0.99, 1.17, 0.95	1.04	103	116 / 104
12	408	0.96, 0.94, 0.88	0.92	100	104 / 92
24	750	1.06, 1.02, 1.04	1.04	93	117 / 104
Cucumber					
0	0	0.99, 1.04, 1.03	1.02	109	100 / 102
3	104	0.86, 0.87, 0.94	0.89	97	87 / 89
6	188	1.03, 0.93, 0.98	0.98	105	96 / 98
12	406	0.93, 0.93, 0.89	0.92	102	90 / 92
24	750	0.94, 0.83, 0.78	0.85	91	84 / 85
Tomato					
0	0	0.92, 0.82, 1.02	0.92	108	100 / 92
3	98	0.84, 0.85, 0.82	0.84	85	91 / 84
6	118	0.94, 0.95, 1.01	0.97	108	105 / 97
12	410	0.94, 0.93, 0.83	0.90	100	98 / 90
24	747	0.90, 0.84, 0.86	0.87	88	94 / 87
Melon					
0	0	0.91, 0.84, 092	0.89	84	100 / 89
3	102	0.88, 0.91, 0.82	0.87	90	98 / 87
6	187	0.97, 1.03, 0.97	0.99	104	111 / 99
12	405	0.94, 0.98, 0.96	0.96	105	108 / 96
24	748	0.94, 0.89, 1.02	0.95	97	107 / 95
Grape					
0	0	0.84, 0.89, 0.87	0.87	87	100 / 87
3	101	0.87, 0.88, 0.85	0.87	87	100 / 87
6	185	1.08, 1.01, 1.04	1.04	107	**121** / 104
12	402	1.05, 1.03, 1.01	1.03	104	119 / 103
24	745	0.87, 0.89, 0.82	0.86	90	99 / 86
Brussels sprout					
0	0	0.85, 0.83, 0.89	0.86	91	100 / 86
3	103	0.88, 0.81, 0.82, 0.92, 1.04, 1.02	0.92	99	106 / 92
6	187	0.97, 1.01, 0.93	0.97	104	114 / 97
12	405	0.94, 0.83, 0.83	0.87	107	101 / 87
24	744	0.84, 0.80, 0.85	0.83	93	97 / 83

Sampling interval (nominal months)	Sampling interval (actual days)	Uncorrected residue (mg/kg)	Mean uncorrected residue (mg/kg)	Mean procedural recovery (%)	Mean recovered uncorrected residue (%), day 0 set at 100 / not corrected for day 0
Head cabbage					
0	0	1.00, 0.92, 1.00	0.97	105	100 / 97
3	101	0.91, 0.85, 0.87	0.88	94	90 / 88
6	185	0.89, 0.91, 0.87	0.89	104	92 / 89
12	405	0.91, 0.95, 0.91	0.92	102	95 / 92
24	745	0.94, 0.92, 0.91	0.92	98	95 / 92
French bean					
0	0	1.01, 1.08, 0.97	1.02	101	100 / 102
3	103	0.99, 0.95, 0.96	0.97	96	95 / 97
6	187	0.93, 1.10, 1.07	1.04	106	102 / 104
12	405	1.05, 1.04, 1.10	1.06	102	104 / 106
24	746	1.01, 1.03, 0.97	1.00	101	99 / 100
Pea					
0	0	0.91, 0.90, 0.83	0.88	92	100 / 88
3	102	0.89, 0.91, 0.86	0.89	93	101 / 89
6	187	0.96, 0.85, 1.00	0.93	101	106 / 93
12	405	0.95, 0.98, 0.94	0.95	110	109 / 95
24	746, 759	0.85, 0.86, 0.89, 0.88, 0.82	0.86	92	98 / 86
Apple					
0	0	0.89, 0.90, 0.86	0.88	101	100 / 88
3	100	0.91, 0.93, 0.96	0.93	93	106 / 93
6	185	1.06, 0.97, 1.10	1.05	101	119 / 105
12	403	0.89, 0.91, 1.00	0.93	103	106 / 93
24	735	1.02, 0.93, 0.92	0.96	93	109 / 96
Potato					
0	0	1.07, 1.04, 1.10	1.07	112	100 / 107
3	86	1.08, 1.14, 1.16	1.13	112	105 / 113
6	169	0.93, 0.93, 0.93	0.93	95	87 / 93
12	387	1.05, 1.08, 1.04	1.06	107	99 / 106
24	730	0.96, 0.94, 0.89	0.93	83	87 / 93
Carrot root					
0	0	0.94, 0.91, 0.90	0.92	96	100 / 92
3	99	0.96, 0.94, 0.96	0.95	93	104 / 95
6	183	1.05, 1.03, 1.04	1.04	102	114 / 104
12	401	1.01, 1.02, 0.96	1.00	98	109 / 100
24	740	0.92, 0.96, 1.00	0.96	89	105 / 96
Leek					

Sampling interval (nominal months)	Sampling interval (actual days)	Uncorrected residue (mg/kg)	Mean uncorrected residue (mg/kg)	Mean procedural recovery (%)	Mean recovered uncorrected residue (%), day 0 set at 100 / not corrected for day 0
0	0	0.84, 0.83, 0.82	0.83	87	100 / 83
3	99	0.96, 0.89, 0.91	0.92	99	111 / 92
6	186	0.96, 0.88, 0.87	0.90	90	109 / 90
12	404	0.96, 0.97, 0.96	0.96	105	116 / 96
24	741	1.17, 1.08, 1.00	1.08	100	**130** / 108
Plum (excluding stone)					
0	0	0.92, 0.97, 0.91	0.93	92	100 / 93
3	98	0.90, 0.97, 0.95	0.94	91	101 / 94
6	181	1.13, 1.09, 1.05	1.09	112	117 / 109
12	400	0.99, 0.97, 0.97	0.98	103	105 / 98
24	737	0.94, 0.90, 0.90	0.91	83	98 / 91
Sugar beet root					
0	0	0.95, 0.97, 0.87	0.93	96	100 / 93
3	91	0.99, 0.91, 0.91	0.94	92	101 / 94
6	174	0.99, 0.97, 1.09	1.02	99	109 / 102
12	393	1.04, 1.01, 1.04	1.03	102	111 / 103
24	730	0.86, 1.08, 1.12	1.02	100	110 / 102
Olive (excluding stone)					
0	0	0.98, 0.96, 0.96	0.97	98	100 / 97
3	91	0.88, 0.89, 0.88	0.88	85	91 / 88
6	175	1.08, 1.19, 1.18	1.15	107	119 / 115
12	393	0.95, 0.97, 0.95	0.96	95	99 / 96
24	733	0.82, 0.86, 0.92	0.87	92	90 / 87
Banana (whole fruit)					
0	0	0.94, 0.93, 0.86	0.91	95	100 / 91
3	91	1.03, 1.07, 1.03	1.04	103	114 / 104
6	175	1.08, 1.07, 1.06	1.07	103	117 / 107
12	393	0.99, 1.03, 1.04	1.02	108	112 / 102
24	730	0.93, 0.84, 0.86	0.88	80	96 / 88

Values in bold are outside the acceptable range of 70-120% recovery.

No residues were present above the LOQ in control samples.

Table 7.1.3-5: Freezer storage stability for R182281 at 1.0 mg/kg in crops

Sampling interval (nominal months)	Sampling interval (actual days)	Uncorrected residue (mg/kg)	Mean uncorrected residue (mg/kg)	Mean procedural recovery (%)	Mean recovered uncorrected residue (%) day 0 set at 100 / not corrected for day 0

Sampling interval (nominal months)	Sampling interval (actual days)	Uncorrected residue (mg/kg)	Mean uncorrected residue (mg/kg)	Mean procedural recovery (%)	Mean recovered uncorrected residue (%) day 0 set at 100 / not corrected for day 0
Strawberry					
0	0	1.15, 1.08, 1.08	1.10	117	100 / 110
3	93	0.85, 0.85, 0.84	0.85	100	77 / 85
6	184	0.80, 0.75, 0.72	0.76	99	**69** / 76
12	408	0.70, 0.78, 0.76	0.75	91	**68** / 75
24	479	0.80, 0.78, 0.76	0.78	99	71 / 78
Onion					
0	0	1.11, 1.14, 1.18	1.14	117	100 / 114
3	92, 96	0.66, 0.61, 0.60, 0.60, 0.59, 0.60	0.61	102	**54 / 61**
6	187	0.58, 0.53, 0.61	0.58	107	**50 / 58**
12	408	0.42, 0.47, 0.44	0.44	92	**39 / 44**
24	749	0.76, 0.60, 0.68	0.68	110	**59 / 68**
Broccoli					
0	0	1.16, 1.08, 1.17	1.14	112	100 / 114
3	95	0.98, 1.03, 1.02	1.01	111	89 / 101
6	187	0.89, 0.96, 0.96	0.94	99	82 / 94
12	410	0.86, 0.87, 0.90	0.88	96	77 / 88
24	748	0.97, 1.00, 0.86	0.94	100	83 / 94
Cauliflower					
0	0	1.02, 0.96, 0.92	0.97	105	100 / 97
3	105	0.88, 0.86, 0.91	0.88	101	91 / 88
6	189	0.80, 0.85, 0.81	0.82	107	85 / 82
12	408	0.85, 0.87, 0.98	0.90	101	93 / 90
24	750	0.98, 1.14, 1.12	1.08	105	112 / 108
Cucumber					
0	0	1.01, 1.02, 0.94	0.99	106	100 / 99
3	104	0.83, 0.87, 0.91	0.87	92	88 / 87
6	188	0.91, 0.81, 0.88	0.87	103	88 / 87
12	406	0.79, 0.77, 0.75	0.77	96	78 / 77
24	750	0.93, 0.93, 0.88	0.92	105	92 / 92
Tomato					
0	0	1.00, 0.90, 1.12	1.00	111	100 / 100
3	98	0.74, 0.76, 0.72, 0.83, 0.81, 0.77	0.77	93	77 / 77
3	108	0.96, 0.93, 0.93	0.94	102	94 / 94
6	188	0.84, 0.83, 0.87	0.84	107	84 / 84
12	410	0.79, 0.73, 0.68	0.73	92	73 / 73

Sampling interval (nominal months)	Sampling interval (actual days)	Uncorrected residue (mg/kg)	Mean uncorrected residue (mg/kg)	Mean procedural recovery (%)	Mean recovered uncorrected residue (%) day 0 set at 100 / not corrected for day 0
24	747	0.78, 0.77, 0.82	0.79	91	78 / 79
Melon					
0	0	0.92, 0.85, 0.94	0.90	89	100 / 90
3	102	0.84, 0.89, 0.79	0.84	92	93 / 84
6	187	0.85, 0.88, 0.86	0.86	103	96 / 86
12	405	0.78, 0.81, 0.79	0.79	100	88 / 79
24	748	0.88, 0.82 ,0.87	0.85	101	95 / 85
Grape					
0	0	0.92, 0.94, 0.92	0.93	93	100 / 93
3	101	0.71, 0.72, 0.67	0.70	90	75 / 70
6	185	0.64, 0.59, 0.63	0.62	98	**67 / 62**
12	402	0.55, 0.43, 0.43	0.47	96	**51 / 47**
24	745	0.48, 0.67, 0.59	0.58	104	**62 / 58**
Brussels sprout					
0	0	0.96, 1.01, 0.97	0.98	103	100 / 98
3	103	0.94, 0.87, 0.96	0.92	91	94 / 91
6	187	0.99, 1.00, 0.95	0.98	99	100 / 98
12	405	0.94, 0.90, 0.94	0.93	96	95 / 93
24	744	1.16, 1.10, 1.08	1.11	103	113 / 111
Head cabbage					
0	0	1.01, 1.08, 1.04	1.04	110	100 / 104
3	101	0.90, 0.93, 0.94	0.92	95	88 / 92
6	185	0.95, 0.97, 0.96	0.96	109	92 / 96
12	405	0.85, 0.86, 0.86	0.86	96	83 / 86
24	745	0.93, 0.94, 0.94	0.94	99	90 / 94
French bean					
0	0	0.95, 1.00, 0.97	0.98	95	100 / 98
3	103	1.00, 1.02, 0.95	0.99	107	102 / 99
6	187	0.91, 1.02, 1.00	0.98	109	100 / 98
12	405	0.90, 0.97, 0.92	0.93	96	95 / 93
24	746	0.86, 0.74, 0.88	0.83	79	85 / 83
Pea					
0	0	0.95, 0.91, 0.83	0.90	95	100 / 90
3	102	0.99, 1.07, 1.03	1.03	100	115 / 103
6	187	0.99, 0.82, 0.99	0.93	108	104 / 93
12	405	0.94, 0.95, 0.95	0.94	103	105 / 94
24	746	1.14, 1.18, 1.10	1.14	105	**127** / 114
Apple					

Sampling interval (nominal months)	Sampling interval (actual days)	Uncorrected residue (mg/kg)	Mean uncorrected residue (mg/kg)	Mean procedural recovery (%)	Mean recovered uncorrected residue (%) day 0 set at 100 / not corrected for day 0
0	0	0.98, 0.90, 0.92	0.93	98	100 / 93
3	100	0.73, 0.70, 0.73	0.72	78	77 / 72
6	185	0.83, 0.79, 0.81	0.81	98	87 / 81
12	403	0.69, 0.71, 0.76	0.72	96	77 / 72
24	735	0.77, 0.73, 0.76	0.75	88	81 / 75
Potato					
0	0	1.02, 0.97, 1.05	1.01	105	100 / 101
3	86	0.97, 0.90, 1.00	0.96	101	94 / 96
6	169	0.96, 0.95, 1.00	0.97	106	96 / 97
12	387	0.84, 0.89, 0.86	0.87	95	85 / 87
24	730	0.94, 0.91, 0.91	0.92	88	91 / 92
Carrot root					
0	0	1.01, 0.98, 0.97	1.02	102	100 / 102
3	99	0.94, 0.93, 0.88	0.92	93	90 / 92
6	183	0.98, 0.91, 0.99	0.96	104	94 / 96
12	401	0.91, 0.94, 0.90	0.91	97	90 / 91
24	740	0.91, 0.94, 0.98	0.94	92	93 / 94
Leek					
0	0	0.89, 0.94, 0.90	0.91	92	100 / 91
3	99	0.80, 0.69, 0.74	0.74	102	82 / 74
6	186	0.70, 0.68, 0.66	0.68	97	75 / **68**
12	404	0.68, 0.68, 0.68	0.68	98	75 / **68**
24	741	0.64, 0.68, 0.65	0.66	91	73 / **66**
Plum (excluding stone)					
0	0	0.83, 0.82, 0.82	0.82	81	100 / 82
3	98	0.81, 0.83, 0.77	0.80	89	97 / 80
6	181	0.84, 0.85, 0.84	0.84	106	102 / 84
12	400	0.71, 0.68, 0.70	0.70	93	85 / 70
24	737, 745	0.67, 0.63, 0.43, 0.77, 0.74, 0.79	0.67	93	79 / 67
Sugar beet root					
0	0	0.99, 1.04, 0.93	0.99	96	100 / 99
3	91	0.95, 0.85, 0.91	0.92	97	93 / 92
6	174	0.89, 0.90, 0.97	0.92	103	93 / 92
12	393	0.89, 0.87, 0.88	0.88	94	89 / 88
24	730	0.85, 0.92, 0.93	0.90	97	91 / 90
Olive (excluding stone)					
0	0	0.97, 0.93, 0.99	0.96	95	100 / 96

Sampling interval (nominal months)	**Sampling interval (actual days)**	**Uncorrected residue (mg/kg)**	**Mean uncorrected residue (mg/kg)**	**Mean procedural recovery (%)**	**Mean recovered uncorrected residue (%) day 0 set at 100 / not corrected for day 0**
3	91	1.10, 1.14, 1.14	1.13	110	117 / 113
6	175	0.92, 0.96, 0.95	0.94	107	98 / 94
12	393	0.93, 0.94, 0.93	0.93	96	97 / 93
24	733	0.96, 0.94, 0.97	0.96	87	100 / 96
Banana (whole fruit)					
0	0	0.76, 0.77, 0.72	0.75	83	100 / 75
3	91	0.95, 1.00, 0.95	0.97	106	129 / 97
6	175	0.95, 0.95, 0.92	0.94	107	125 / 94
12	393	0.77, 0.82, 0.81	0.80	95	107 / 80
24	730	0.70, 0.64, 0.64	0.66	73	88 / **66**

Values in bold are outside the acceptable range of 70-120% recovery.

No residues were present above the LOQ in control samples.

III. CONCLUSIONS

Residues of chlorothalonil were stable for at least 24 months in onion, broccoli, cauliflower, cucumber, melon, tomato, Brussels sprout, cabbage, French bean, pea, apple, leek, plum, sugar beet root and banana (high water crop group), olive (high oil crop group), potato, carrot root (high starch crop group) strawberry and grape (high acid crop group) when stored in the freezer at ≤-18°C.

Residues of R182281 were stable for at least 24 months in broccoli, cauliflower, cucumber, melon, tomato, Brussels sprout, cabbage, French bean, pea, apple, ~~leek~~, plum, sugar beet root and banana (high water crop group), olive (high oil crop group), potato, carrot root (high starch crop group) and strawberry (high acid crop group) when stored in the freezer at ≤-18°C. Residues of R182281 were found to stable in grapes (high acid) for up to 3 months and were stable for less than 3 months in onions (high water) and a decerase was observed in residues in leek after 3 months when stored frozen at ≤-18°C.

Decrease of stability in leek/onion matrix can be related to interferences of the residues with matrix of thise crops (*Allium sp*). Hence, the conclusion on stability of metabolite SDS-3701 in all water matrices is limited.

The study demonstrated that residues of chlorothalonil and R182281 remained stable during freezer storage when prepared in the presence of acid.

Limitations

It was noted by the expert peer review meeting, that the individual recoveries for chlorothalonil were not provided and only a mean recovery was reported, which is not in line with OECD recomendations.

B.7.1.4 Storage stability in cereal straw

Previous evaluation	Submitted for the purpose of renewal
RMS remark	Acceptable

Report:	K-CA 6.1/03. Brown D., (2014), Chlorothalonil - Storage stability of residues of chlorothalonil and R182281 in cereal straw for up to 12 months. Eurofins Agroscience Service Chem Ltd, Derbyshire, United Kingdom. Report Number S12-01844. Syngenta File No: R044686_11076.

Guidelines

OECD Guideline for Testing of Chemicals No. 506, Stability of Pesticide Residues in Stored Commodities, adopted 16 October 2007

EPA Residue Chemistry Test Guidelines, OPPTS 860.1380, Storage Stability Data, August 1996

Commission of the European Communities: Storage Stability of Residue Samples (SANCO 7032/V1/95 rev. 5 22/7/1997).

GLP

The study was carried out according to the principles of Good Laboratory Practice.

EXECUTIVE SUMMARY

Samples of cereal straw were fortified at 0.5 mg/kg with chlorothalonil or R182281. Triplicate samples were stored under frozen conditions (≤-18°C) and analysed at intervals up to 12 months (five sampling points). The LOQ for both chlorothalonil and R182281 was 0.01 mg/kg.

There was no significant decrease (>30% compared to the zero time value) in the levels of chlorothalonil in cereal straw over 9 months and no significant decrease in the observed residue levels of R182281 in cereal straw over 12 months when stored at ≤-18°C.

Residues of chlorothalonil in cereal straw were stable for 9 months and residues of R182281 in cereal straw were stable for at least 12 months when stored deep frozen at -18°C.

A1. Test Materials

The purity of the analytical standards used in this study is listed in Table 7.1.4-1.

Table 7.1.4-1: Purity of analytical standards

Analyte	Standard reference no.	Purity (wt. %)
Chlorothalonil	638406	99.6
R 182281	P2	98.7

A2. Test Commodity

The test commodity was untreated homogenised cereal straw taken from other studies conducted by the test facility.

A3. Test Facilities

This study was performed at Eurofins Agroscience Services Chem Ltd, Slade Lane, Wilson, Melbourne, Derbyshire, DE73 8AG, UK

B. STUDY DESIGN AND METHODS

B1. Fortification and Storage of Samples

Homogenised samples were fortified at 0.5 mg/kg with either chlorothalonil in acidified toluene or R182281 in acidified acetone. Triplicate samples were stored under frozen conditions (≤-18°C) and analysed at intervals up to 12 months (five sampling points). Control samples were analysed at zero time and at each time point to ensure that no residues of chlorothalonil or R182281 were present at levels above 30% of the LOQ.

B2. Analytical Method

Analysis of the samples was performed according to analytical method GRM005.01.A at intervals of 0, 1, 3, 6, 9 and 12 months. A full method description and validation data are presented in B.5.2.1 (K-CA 4.2/01-K-CA 4.2/07). The method involved extraction of the samples with acetone/5M sulphuric acid followed by centrifugation. For chlorothalonil determination, aliquots are taken and diluted with water followed by SPE clean up using a C8 cartridge. Chlorothalonil residues were eluted from the SPE cartridges using toluene. The final volume of the extracts was adjusted to 4 mL using acidified toluene. Chlorothalonil is analysed by GC-MSD.

For the determination of R182281, aliquots of the extracts are taken and diluted with acetonitrile: water (50/50, v/v) to a volume of 10 ml. Final determination of R182281 is by LC-MS/MS. The LOQ was 0.01 mg/kg for both analytes.

II. RESULTS AND DISCUSSION

Method Validation

Procedural recoveries were determined using freshly fortified samples at each interval. Samples were fortified with chlorothalonil or R182281 at 0.5 mg/kg. Individual recoveries, mean recoveries and %RSD are summarised in Table 7.1.4-2 (chlorothalonil) and Table 7.1.4-3 (R182281).

Table 7.1.4-2: Summary of procedural recoveries for chlorothalonil in cereal straw

Recovery at zero time (%)	Recovery at 1 month (%)	Recovery at 3 months (%)	Recovery at 6 months (%)	Recovery at 9 months (%)	Recovery at 12 months (%)	Mean recovery (%)	RSD (%)
94, 95	100, 104	94, 99	97, 94	90, 94	91, 87	95	5

Table 7.1.4-3: Summary of procedural recoveries for R182281 in cereal straw

Recovery at zero time (%)	Recovery at 1 month (%)	Recovery at 3 months (%)	Recovery at 6 months (%)	Recovery at 9 months (%)	Recovery at 12 months (%)	Mean recovery (%)	RSD (%)
100, 101	103, 103	102, 101	95, 101	99, 102	95, 91	99	4

Storage Stability of Residues

The recoveries of chlorothalonil and R182281 in cereal straw stored at ≤ -18°C are summarised in Table 7.1.4-4 and Table 7.1.4-5, respectively. The results are not corrected for freshly fortified recoveries.

Table 7.1.4-4: Freezer storage stability for chlorothalonil at 0.5 mg/kg in cereal straw

Sampling interval (nominal months)	Sampling interval (actual days)	Uncorrected residue (mg/kg)	Mean uncorrected residue (mg/kg)	Mean procedural recovery (%)	Mean recovered uncorrected residue (%)
0	0	0.49, 0.49, 0.49	0.49	95	98
1	29	0.48, 0.48, 0.48	0.48	102	96
3	91	0.42, 0.43, 0.43	0.43	97	86
6	182	0.38, 0.38, 0.37	0.38	96	76
9	274	0.35, 0.34, 0.35	0.35	92	70
12	367	0.31, 0.30, 0.31	0.31	89	**62**

Calculations performed on unrounded values.
No residues were present above the LOQ in control samples, however, no control data are presented in the study report.
Values in bold are outside the acceptable range of 70-120% recovery.

Table 7.1.4-5: Freezer storage stability for R182281 at 0.5 mg/kg in cereal straw

Sampling interval (nominal months)	Sampling interval (actual days)	Uncorrected residue (mg/kg)	Mean uncorrected residue (mg/kg)	Mean procedural recovery (%)	Mean recovered uncorrected residue (%)
0	0	0.51, 0.50, 0.52	0.51	100	102
1	29	0.50, 0.51, 0.51	0.51	103	102
3	91	0.50, 0.50, 0.50	0.50	102	100
6	182	0.49, 0.49, 0.47	0.48	98	96
9	274	0.49, 0.50, 0.52	0.51	100	102
12	367	0.50, 0.47, 0.41	0.46	93	92

Calculations performed on unrounded values.
Residue in control sample for 9 month time point was 31% of the LOQ, however, there is no information in the study report to check this statement.

Due to the steady decline of chlorothalonil residues overtime, additional analysis was undertaken at 9 months and 12 months. The sample sets fortified with chlorothalonil were also analysed for R182281 to assess whether the decline of chlorothalonil was due to conversion to R182281.

A total chlorothalonil residue was calculated as the sum of chlorothalonil (from Table 7.1.4-4) and R182281. The residue equivalent to chlorothalonil was calculated using a conversion factor based on the molecular weight of chlorothalonil and metabolite R182281 (265.9/247.5=1.0743). The results are presented in Table 7.1.4-6.

Table 7.1.4-6: Reanalysis of samples fortified with chlorothalonil at 0.5 mg/kg in cereal straw for R182281

Sampling interval (nominal months)	Sampling interval (actual days)	Uncorrected residue as R182281 (mg/kg)	Uncorrected residue as chlorothalonil equivalents (mg/kg)	Chlorothalonil residue from Table 7.1.4-4 (mg/kg)	Total chlorothalonil residues (mg/kg)	Mean recovered uncorrected residue (%)
9	274	0.013, 0.015, 0.013	0.014, 0.016, 0.014	0.35, 0.34, 0.35	0.36, 0.36, 0.36	72
12	367	0.017, 0.013, 0.016	0.018, 0.014, 0.017	0.31, 0.30, 0.31	0.33, 0.31, 0.33	**64**

Values in bold are outside the acceptable range of 70-120% recovery.

III. CONCLUSIONS

There was no significant decrease (>30% as compared to the zero-time value) in the residue levels of chlorothalonil in cereal straw after deep frozen storage for 9 months, however, a significant decrease (>30%) was observed at the 12 month time point. Some of the reduction in levels of chlorothalonil may have been due to degradation to R182281.
No significant decrease (>30% as compared to the zero-time value) was observed in the residue values of R182281 in cereal straw after deep frozen storage for 12 months.

B.7.1.5 Storage stability in various crops

Previous evaluation	Submitted for the purpose of renewal
RMS remark	Acceptable, however, the expert peer review meeting has concluded that for soya bean (high oil crop) results are inclonclusive.

Report:	K-CA 6.1/04. Gasso-Brown D (2015), Chlorothalonil - Storage stability of residues of R611965 in crop matrices stored frozen for up 30 months. Eurofins Agroscience Service Chem Ltd, Derbyshire, United Kingdom. Report Number S12-04611. Syngenta File No: R611965_10041

Guidelines

OECD Guideline for Testing of Chemicals No. 506, Stability of Pesticide Residues in Stored Commodities, adopted 16 October 2007

EPA Residue Chemistry Test Guidelines, OPPTS 860.1380, Storage Stability Data, August 1996

Commission of the European Communities: Storage Stability of Residue Samples (SANCO 7032/V1/95 rev. 5 22/7/1997).

GLP

The study was carried out according to the principles of Good Laboratory Practice.

EXECUTIVE SUMMARY

Samples of wheat grain, tomato, lentil, orange and soy bean were fortified at 0.5 mg/kg with R611965. Duplicate samples were stored under frozen conditions (≤-18°C) and analysed at intervals up to 12 months (four or five sampling points). The LOQ for R611965 was 0.01 mg/kg.

There was no significant decrease (>30% compared to the zero time value) in the levels of R611965 in the crops tested over 30 months when stored at ≤-18°C.

A1. Test Materials

The purity of the analytical standard used in this study is listed in Table 7.1.5-1.

Table 7.1.5-1: Purity of analytical standards

Analyte	Standard reference no.	Purity (wt. %)
R 611965	MES 134/1	96

A2. Test Commodity

The test commodities were untreated homogenised wheat grain and soya beans taken from other studies conducted by the test facility, and organic oranges, tomatoes and lentils purchased from a local supermarket.

A3. Test Facilities

This study was performed at Eurofins Agroscience Services Chem Ltd, Slade Lane, Wilson, Melbourne, Derbyshire, DE73 8AG, UK.

B. STUDY DESIGN AND METHODS

B1. Fortification and Storage of Samples

Homogenised samples were fortified at 0.5 mg/kg with R611965 in methanol. Duplicate samples were stored under frozen conditions (≤-18°C) and analysed at intervals up to 12 months (four or five sampling points). Control samples were analysed at zero time and at all other time points to ensure that no residues of R611965 were present at levels above 30% of the LOQ.

B2. Analytical Method

Analysis of the samples was performed according to analytical method GRM005.06A at intervals of 0, 1, 3, 6, 12, 18, 24 and 30 months of frozen storage for wheat grain, tomato, lentil and whole orange, and of 0, 1, 3, 6, 9, 12, 18 and 30 months for soybean seed. A full method description and validation data are presented in B.5.1.2.5 (K-CA 4.1.2/26). The method involved extraction of the samples with acetone/5M sulphuric acid followed by centrifugation. An aliquot of the extract was taken, evaporated and reconstituted in methyl tert-butyl ether (MTBE). A liquid-liquid partition was performed three times and the organic layer transferred to a concentration tube. Extracts were concentrated and reconstituted in water: methanol 50:50 (v/v). Final determination was by LC-MS/MS, monitoring for the primary transition (m/z 268→224). The LOQ was 0.01 mg/kg.

II. RESULTS AND DISCUSSION

Method Validation

Procedural recoveries were determined using freshly fortified samples at each interval. Samples were fortified with R611965 at 0.5 mg/kg. Individual recoveries, mean recoveries and %RSD are summarised in Table 7.1.5-2.

In order to assess the stability of standard solutions of R611965, the instrument response of stored stock and calibration solutions were compared to those of freshly prepared standards. After 6.5 months refrigerated storage the difference in response was < 10% therefore the standard solutions were shown to be stable for approximately 6.5 months.

Table 7.1.5-2: Summary of procedural recoveries for R611965 in crops

Recovery (%)										RSD (%)
zero time	1 month	3 months	6 months	9 months	12 months	18 months	24 months	30 months	Mean	
Wheat grain										
99, 108	93, 92	117, 103	105, 104	-	92, 98	105, 104	106, 108	88, 90	101	8.0
Tomato										
99, 107	97, 94	99, 96	107, 95	-	104, 107	90, 92	103, 101	88, 94	98	6.1
Lentil										
102, 103	94, 91	108, 110	111, 105	-	99, 105	101, 102	108, 109	86, 89	101	7.7
Orange										
107, 107	89, 95	100, 100	109, 111	-	107, 104	101, 99	100, 96	85, 90	100	7.5
Soya bean										
104, 104	88, 90	108, 99	98, 96	102, 102	99, 96	104, 103	60, 59	93, 93	94	15

Storage Stability of Residues

The recoveries of R611965 in various crops stored at ≤ -18°C are summarised in Table 7.1.5-3 below. The results are not corrected for freshly fortified recoveries.

Table 7.1.5-3: Freezer storage stability for R611965 at 0.5 mg/kg in crops

Sampling interval (nominal months)	Sampling interval (actual days)	Uncorrected residue (mg/kg)	Mean uncorrected residue (mg/kg)	Mean procedural recovery (%)	Mean recovered uncorrected residue (%)
Wheat grain					
0	0	0.52, 0.50, 0.52	0.51	103	102
1	31	0.40, 0.41	0.40	92	80
3	91	0.46, 0.50	0.48	110	96
6	182	0.40, 0.42	0.41	104	82
12	365	0.42, 0.45	0.43	95	86
18	547	0.41, 0.41	0.41	104	79
24	734	0.48; 0.47	0.48	107	93
30	915	0.47; 0.44	0.45	89	88
Tomato					
0	0	0.52, 0.50, 0.54	0.52	103	104
1	31	0.47, 0.46	0.47	96	94
3	91	0.50, 0.48	0.49	98	98
6	181	0.56, 0.54	0.55	101	110
12	367	0.52, 0.55	0.53	105	106
18	546	0.49, 0.49	0.47	91	91
24	733	0.49, 0.50	0.50	102	95
30	916	0.49, 0.48	0.48	91	93
Lentil					
0	0	0.52. 0.53. 0.54	0.53	103	106
1	31	0.47. 0.42	0.44	93	88
3	91	0.47. 0.45	0.46	109	92
6	182	0.45. 0.49	0.47	108	94
12	365	0.50. 0.50	0.50	102	100
18	547	0.42, 0.43	0.43	102	80
24	734	0.48, 0.51	0.49	109	93
30	917	0.49, 0.47	0.48	87	91
Orange					
0	0	0.50, 0.54, 0.52	0.52	107	104
1	31	0.45, 0.46	0.45	92	90
3	94	0.49, 0.46	0.47	100	94
6	181	0.51, 0.55	0.53	110	106
12	367	0.52, 0.50	0.51	106	102

Sampling interval (nominal months)	Sampling interval (actual days)	Uncorrected residue (mg/kg)	Mean uncorrected residue (mg/kg)	Mean procedural recovery (%)	Mean recovered uncorrected residue (%)
18	546	0.50, 0.49	0.50	100	95
24	733	0.33, 0.30	0.31	98	60#
30	916	0.47, 0.45	0.46	87	89
Soya bean					
0	0	0.53, 0.54, 0.52	0.53	104	106
1	31	0.39, 0.41	0.40	89	80
3	94	0.32, 0.34	0.33	104	66
6	186	0.37, 0.38	0.38	97	76
9	276	0.34, 0.39	0.36	102	72
12	367	0.45, 0.42	0.44	97	88
18	546	0.34; 0.35	0.34	103	65#
30	916	0.41; 0.39	0.40	93	75

Calculations performed on unrounded values.
No residues were present above the LOQ in control samples.
#Degradation of more than 30% of R611965 from the zero time point, however, results for later time points are within the accepted range implying residues are stable over the 30 month period.

III. CONCLUSIONS

There was no significant decrease (>30% as compared to the zero-time value) in the observed residue levels of R611965 in wheat grain (high starch), tomato (high water), lentil (high protein) and whole orange (high acid) after deep frozen storage for 30 months. Degradation of more than 30% of R611965 from the zero time point was observed in the 3 and 18 month time-points for soya bean, although the recoveries for later time points are within the accepted range demonstrating that residues are stable over the 30 month period. Therefore, R611965 is considered to be stable in samples of wheat grain, tomato, lentil, whole orange and soybean stored for 30 months at ≤ -18°C.

B.7.1.6 Storage stability in wheat

Previous evaluation	Submitted for the purpose of renewal
RMS remark	Acceptable.

Report:	K-CA 6.1/05. Krainz A. (2007), Chlorothalonil: Frozen storage stability in wheat (grain and straw). RCC Ltd, Switzerland. Report Number A71256. Syngenta File No: R044686_11197.

Guidelines

Commission of the European Communities: Storage Stability of Residue Samples (SANCO 7032/V1/95 rev. 5 22/7/1997).

GLP

The study was carried out according to the principles of Good Laboratory Practice.

EXECUTIVE SUMMARY

Samples of wheat grain and straw were fortified at 0.2 mg/kg with chlorothalonil. Triplicate samples were stored under frozen conditions (-20°C) and analysed at 0, 1, 2 and 3 months (three sampling points). The LOQ for chlorothalonil was 0.01 mg/kg.

Residues of chlorothalonil were stable in wheat grain and straw for at least 3 months when stored in the freezer at -20°C.

A1. Test Materials

The purity of the analytical standard used in this study is listed in Table 7.1.6-1.

Table 7.1.6-1: Purity of analytical standards

Analyte	Standard reference no.	Purity (wt. %)
Chlorothalonil	337-98B	98

A2. Test Commodity

The test commodities were untreated homogenised wheat grain and straw taken from other studies conducted by the test facility.

A3. Test Facilities

This study was performed at RCC Ltd, Analytics, Zelgliweg 1, CH-4452 Itingen, Switzerland.

B. STUDY DESIGN AND METHODS

B1. Fortification and Storage of Samples

Homogenised samples were fortified at 0.2 mg/kg with chlorothalonil in toluene. Triplicate samples were stored under frozen conditions (-20°C) and analysed at intervals up to 3 months (three sampling points). Control samples were analysed at zero time and at all other time points to ensure that no residues of chlorothalonil were present at levels above 30% of the LOQ.

B2. Analytical Method

Analysis of the samples was performed according to analytical method A75813 at intervals of 0, 1, and 3 months. A full method description and validation data are presented in B.5.1.2.5 (K-CA 4.1.2/30). The method involved extraction of the samples with acetone/5M sulphuric acid followed by centrifugation. An aliquot of the extract was taken, diluted and subjected to SPE (C18) clean up, before analysis by gas chromatography with electron capture detection (GC-ECD) using two columns of different polarity for quantification. The LOQ was 0.01 mg/kg.

II. RESULTS AND DISCUSSION

Method Validation

Procedural recoveries were determined using freshly fortified samples at each interval. Samples were fortified with chlorothalonil at 0.2 mg/kg. Individual recoveries, mean recoveries and %RSD are summarised in Table 7.1.6-2.

Table 7.1.6-2: Summary of procedural recoveries for chlorothalonil in wheat grain and straw

Recovery at zero time (%)	Recovery at 1 month (%)	Recovery at 2 months (%)	Recovery at 3 months (%)	Mean recovery (%)	RSD (%)
Wheat grain					
101, 95, 92, 94 (mean 95)	90	81	72	89	11
Wheat straw					
88, 95, 103, 111 (mean 99)	88	83	71	91	14

Storage Stability of Residues

The recoveries of chlorothalonil in cereal crops stored at -20°C are summarised in Table 7.1.6-3 below. The mean recovery of chlorothanonil in wheat grain at 3 months of storage was found to be 64%; the procedural recovery at 3 months was 72%, suggesting there may not be a decline on storage.

Table 7.1.6-3: Freezer storage stability for chlorothalonil at 0.2 mg/kg in cereal crops

Sampling interval (nominal months)	Sampling interval (actual days)	Uncorrected residue (mg/kg)	Mean uncorrected residue (mg/kg)	Mean recovered uncorrected residue (%)	Procedural recovery (%)
Wheat grain					
0	0	0.20, 0.19, 0.19, 0.19	0.19	95	-
1	32	0.15, 0.17, 0.15	0.16	80	90
2	62	0.19, 0.19, 0.19	0.19	95	81
3	99	0.13, 0.13, 0.13	0.13	65	72
Wheat Straw					
0	0	0.18, 0.19, 0.21, 0.22	0.20	100	-
1	32	0.20, 0.19, 0.21	0.20	100	88
2	62	0.21, 0.21, 0.22	0.21	105	83
3	92	0.18, 0.20, 0.22	0.20	100	71

Calculations performed on unrounded values.
No residues were present above the LOQ in control samples.

III. CONCLUSIONS

There was no significant decrease in the observed residue levels of chlorothalonil in cereal grain (high starch) and straw after deep frozen storage for 3 months when results were corrected for procedural recoveries.

B.7.1.7 Storage stability in processed crop commodities

Previous evaluation	Submitted for the purpose of renewal
RMS remark	Acceptable

Report:	K-CA 6.1/06. Heillaut C and Anderson L. (2007), R613636: storage stability of residues in processed crop commodities stored deep frozen for up to two years. ADME Bioanalyses, Vergèze, France. Syngenta Report Number T007198-04-REG. (Syngenta File No: R613636/0003).

Guidelines

Not stated but evaluated as compliant with testing guideline OECD Test No. 506 ~~meets the requirements of Guideline: Commission of the European Communities: Storage Stability of Residue Samples (SANCO 7032/V1/95 rev. 5 22/7/1997).~~

OPPTS 860-1380

GLP

The study was carried out according to the principles of Good Laboratory Practice.

EXECUTIVE SUMMARY

Samples of processed barley (pearl barley and beer), processed wheat (bran and flour), processed peanut (meal and oil), processed tomato (juice, paste and puree) and processed legumes (cooked beans with pods) were fortified with the chlorothalonil metabolite R613636 at 0.10 mg/kg, stored under frozen conditions (≤-18° C) and analysed at intervals up to 24 months (four time points). The LOQ was 0.01 mg/kg.

There was no significant decrease in the levels of R613636 in any processed commodity tested over 24 months. Residues of R613636 were therefore stable for at least 24 months in all processed commodities when stored in the freezer at ≤-18°C.

A1. Test Materials

The purity of the analytical standard used in this study is listed in Table 7.1.7-1.

Table 7.1.7-1: Purity of analytical standards

Analyte	Standard reference no.	Purity (wt. %)
R613636	ASJ10214-01	99

A2. Test Commodity

The peanut meal, tomato paste and tomato puree were prepared by Syngenta to US guidelines. The pearl barley, beer, wheat bran, wheat flour, peanut oil, tomato juice and beans with pods were purchased from UK supermarkets.

A3. Test Facilities

Sample preparation and analysis up to and including the 12 month storage interval was performed at Syngenta, Jealott's Hill International Research Centre, Bracknell, Berkshire, RG42 6EY, UK. Samples for the 24 month analysis were analysed at ADME Bioanalyses, 75, Chemin de Sommières, F-30310 Vergèze, France.

B. STUDY DESIGN AND METHODS

B1. Fortification and Storage of Samples

The peanut meal was homogenised to remove lumps. The beans with pods were boiled in water for 15-20 minutes, drained, cooled, frozen and then chopped with dry ice. The pearl barley was ground in an ultracentrifugal mill.

Samples (10 g) of pearl barley, beer, wheat bran, wheat flour, peanut meal, peanut oil, cooked beans with pods, tomato juice, tomato paste and tomato puree were fortified with R613636 in acetone at 0.10 mg/kg. Triplicate samples were stored under frozen conditions (≤-18°C) and analysed at intervals up to 24 months (four time points). Control samples were analysed at the zero time and at each time point.

B2. Analytical Method

Analysis of the samples was performed according to analytical method RAM 464/01 at intervals of 0, 3, 6, 12 and 24 months. The method involved extraction of the samples by homogenisation with acetone: 5M sulphuric acid (95:5, v: v). After centrifugation, aliquots were diluted with water and cleaned by solid phase extraction. Final determination was by HPLC coupled to a triple quadrupole mass spectrometer in selected reaction monitoring mode (LC-MS/MS). The LOQ was 0.01 mg/kg.

A full method description and validation data are presented in B.5.1.2.5 (K-CA 4.1.2/28).

II. RESULTS AND DISCUSSION

Method Validation

Procedural recoveries were determined using freshly fortified samples at each interval. Samples were fortified with R613636 at 0.1 mg/kg. Individual recoveries, mean recoveries and %RSD are summarised in Table 7.1.7-2.

Table 7.1.7-2: Summary of procedural recoveries for R613636 in processed commodities

Recovery at zero time (%)	Recovery at 3 months (%)	Recovery at 6 months (%)	Recovery at 12 months (%)	Recovery at 24 months (%)	Mean recovery (%)	RSD (%)
Pearl barley						
112, 105	94, 96	95, 90	89, 91	92, 85	95	8
Beer						
101, 99	104, 107	98, 106	95, 100	91, 87	89	3
Wheat bran						
114, 110	77, 80	96, 97	66, 65	92, 93	89	19
Wheat flour						
106, 100	102, 103	88, 95	88, 95	96, 107	98	7
Peanut meal						
99, 102	86, 88	80, 72	88, 80	103, 100	90	12
Peanut oil						
98, 97	86, 98	102, 99	76, 78	94, 90	92	10
Cooked beans with pods						
101, 96	111, 112	99, 105	91, 90	94, 92	99	8
Tomato juice						
95, 113	93, 102	108, 102	99, 95	94, 91	99	7
Tomato paste						
96, 96	101, 108	99, 91	93, 86	86, 90	95	7
Tomato puree						
104, 99	108, 105	102, 100	95, 91	92, 89	99	7

Storage Stability of Residues

The recoveries of R613636 in processed commodities stored at ≤-18°C are summarised in Table 7.1.7-3. The results are not corrected for freshly fortified recoveries.

Table 7.1.7-3: Freezer storage stability for R613636 in processed commodities

Sampling interval (nominal months)	Sampling interval (actual days)	Uncorrected residue (mg/kg)	Mean uncorrected residue (mg/kg)	Mean procedural recovery (%)	Mean recovered uncorrected residue (%)
Pearl barley					
0	0	0.09, 0.10, 0.10	0.10	108	100
3	106	0.09, 0.09, 0.09	0.09	95	90
6	218	0.10, 0.09, 0.09	0.09	92	90
12	359	0.09, 0.09, 0.09	0.09	90	90
24	787	0.10, 0.09, 0.09	0.09	88	90
Beer					

Sampling interval (nominal months)	Sampling interval (actual days)	Uncorrected residue (mg/kg)	Mean uncorrected residue (mg/kg)	Mean procedural recovery (%)	Mean recovered uncorrected residue (%)
0	0	0.10, 0.09, 0.10	0.10	100	100
3	92	0.11, 0.11, 0.10	0.11	105	110
6	215	0.11, 0.10, 0.10	0.10	102	100
12	348	0.10, 0.10, 0.10	0.10	97	100
24	770	0.08, 0.09, 0.09	0.09	89	90
Wheat bran					
0*	0	0.12, 0.11, 0.12	0.11	112	110
3*	73	0.08, 0.09, 0.09	0.09	78	90
6*	196	0.10, 0.09, 0.09	0.09	96	90
12*	330	0.07, 0.07, 0.08	0.07	65	70**
24*	750	0.08, 0.09, 0.10	0.09	93	90
Wheat flour					
0	0	0.10, 0.10, 0.10	0.10	103	100
3	109	0.09, 0.10, 0.10	0.10	103	100
6	221	0.09, 0.10, 0.10	0.10	92	100
12	362	0.09, 0.09, 0.09	0.09	91	90
24	789	0.10, 0.11, 0.11	0.11	101	110
Peanut meal					
0	0	0.08, 0.09, 0.09	0.09	101	90
3	85	0.09, 0.08, 0.08	0.08	87	80
6	208	0.07, 0.07, 0.08	0.07	76	70
12	342	0.08, 0.08, 0.08	0.08	84	80
24	762	0.10, 0.10, 0.10	0.10	101	100
Peanut oil					
0	0	0.10, 0.09, 0.09	0.09	97	90
3	106	0.10, 0.09, 0.08	0.09	92	90
6	217	0.08, 0.08, 0.08	0.08	101	80
12	363	0.09, 0.08, 0.08	0.08	77	80
24	792	0.09, 0.09, 0.09	0.09	92	90
Cooked beans with pods					
0	0	0.09, 0.10, 0.09	0.10	98	100
3	104	0.11, 0.11, 0.11	0.11	111	110
6	224	0.09, 0.10, 0.09	0.10	102	100
12	357	0.09, 0.09, 0.09	0.09	90	90
24	782	0.10, 0.10, 0.08	0.09	93	90
Tomato juice					
0	0	0.12, 0.11, 0.11	0.11	104	110
3	105	0.09, 0.10, 0.09	0.09	98	90

Sampling interval (nominal months)	Sampling interval (actual days)	Uncorrected residue (mg/kg)	Mean uncorrected residue (mg/kg)	Mean procedural recovery (%)	Mean recovered uncorrected residue (%)
6	216	0.11, 0.10, 0.10	0.11	105	110
12	362	0.10, 0.09, 0.09	0.09	97	90
24	790	0.09, 0.10, 0.09	0.09	93	90
Tomato paste					
0	0	0.10, 0.09, 0.10	0.10	96	100
3	92	0.10, 0.10, 0.10	0.10	104	100
6	212	0.09, 0.09, 0.09	0.09	95	90
12	345	0.09, 0.09, 0.09	0.09	90	90
24	772	0.09, 0.09, 0.09	0.09	88	90
Tomato puree					
0	0	0.10, 0.10, 0.09	0.10	102	100
3	92	0.11, 0.11, 0.11	0.11	106	110
6	212	0.10, 0.10, 0.10	0.10	101	100
12	349	0.10, 0.10, 0.11	0.10	93	100
24	770	0.09, 0.09, 0.09	0.09	90	90

Calculations performed on unrounded values.
* Wheat bran residues and recoveries were corrected for any apparent residue in the untreated control samples. No residues were present above the LOQ in other control samples. However, no data are presented in the study report to check the statement that there are no residues in the control samples.
**The mean recovery for wheat bran after 12 months of storage is 70%; for the same day the mean procedural recovery is also low (65%).

III. CONCLUSIONS

All samples showed a degradation <30% compared to the zero time point. Residues of R613636 were stable in pearl barley, beer, wheat bran, wheat flour, peanut meal, peanut oil, cooked beans with pods, tomato juice, tomato paste and tomato puree for at least 24 months when stored in the freezer at ≤-18°C.

B.7.1.8 Storage stability in bovine muscle, fat, liver, kidney and cow's milk

Previous evaluation	Submitted for the purpose of renewal
RMS remark	Acceptable

Report:	K-CA 6.1/07. Krainz A. (2007a), SDS-3701: Frozen storage stability in bovine muscle, fat, liver, kidney and cow's milk. RCC Ltd, Switzerland. Report Number A71278. Syngenta File No: R182281_10018.

Guidelines

Commission of the European Communities: Storage Stability of Residue Samples (SANCO 7032/V1/95 rev. 5 22/7/1997).

GLP

The study was carried out according to the principles of Good Laboratory Practice.

EXECUTIVE SUMMARY

Samples of bovine muscle, fat, liver, kidney and cow's milk were fortified at 0.2 mg/kg with R182281. Triplicate samples were stored under frozen conditions (-20°C) and analysed at intervals up to 3 months (three sampling points). The LOQ for R182281 was 0.01 mg/kg.

Residues of R182281 were stable in bovine muscle, fat, liver, kidney and cow's milk for at least 3 months when stored in the freezer at -20°C.

A1. Test Materials

The purity of the analytical standard used in this study is listed in Table 7.1.8-1.

Table 7.1.8-1: Purity of analytical standards

Analyte	Standard reference no.	Purity (wt. %)
R182281 (SDS-3701)	BJQ-impurity 246-001	95

A2. Test Commodity

The test commodities were untreated homogenised bovine muscle, fat, liver, kidney taken from a local butcher. Cow's milk was obtained from a regional distributor.

A3. Test Facilities

This study was performed at RCC Ltd, Analytics, Zelgliweg 1, CH-4452 Itingen, Switzerland.

B. STUDY DESIGN AND METHODS

B1. Fortification and Storage of Samples

Homogenised samples were fortified at 0.2 mg/kg with R182281 in acetonitrile. Triplicate samples were stored under frozen conditions (-20°C) and analysed at intervals up to 3 months (three sampling points). Control samples were analysed at zero time and at all other time points to ensure that no residues of R182281 were present at levels above 30% of the LOQ.

B2. Analytical Method

Analysis of the samples was performed according to analytical method A71201 at intervals of 0, 1, and 3 months. The method validation is reported in study report number A71201. A full method description and validation data are presented in B.5.2.2 (K-CA 4.2/16 and K-CA 4.2/17).

The method involved extraction of the samples of muscle, kidney and liver with acetone/sulphuric acid (95/5, v/v) followed by centrifugation. Samples of milk were extracted with acetonitrile and centrifuged. Samples of fat were extracted with acetonitrile and partitioned with acetonitrile saturated hexane. Aliquots of all extracts were diluted with methanol/water (1/1, v/v) before analysis by LC-MS/MS monitoring the ion transitions m/z 245→175 (quantitation) and m/z 245→182 (confirmation). The LOQ was 0.01 mg/kg.

II. RESULTS AND DISCUSSION

Method Validation

Procedural recoveries were determined using freshly fortified samples at each interval. Samples were fortified with R182281 at 0.02 mg/kg. Individual recoveries, mean recoveries and %RSD are summarised in Table 7.1.8-2.

Table 7.1.8-2: Summary of procedural recoveries for R182281 in products of animal origin

Recovery at zero time (%)	Recovery at 1 month (%)	Recovery at 2 months (%)	Recovery at 3 months (%)	Mean recovery (%)	RSD (%)
Bovine muscle					
72, 80, 79, 78	77	74	68	75	4
Bovine fat					
93, 95, 95, 95	91	98	98	95	3
Bovine liver					
83, 79, 72, 80	73	94	87	81	8
Bovine kidney					
93, 95, 96, 100	93	91	85	93	5
Cow's milk					
103, 102, 106, 104	101	101	102	103	2

Storage Stability of Residues

The recoveries of R182281 in animal matrices stored at -20°C are summarised in Table 7.1.8-3 below. The results are not corrected for freshly fortified recoveries.

Table 7.1.8-3: Freezer storage stability for R182281 at 0.2 mg/kg in animal products

Sampling interval (nominal months)	Sampling interval (actual days)	Uncorrected residue (mg/kg)	Mean uncorrected residue (mg/kg)	Procedural recovery (%)	Mean recovered uncorrected residue (%)
Bovine muscle					
0	0	0.14, 0.16, 0.16, 0.16	0.16	-	80
1	34	0.17, 0.16, 0.16	0.16	77	80
2	62	0.16, 0.15, 0.15	0.15	74	75
3	92	0.14, 0.14, 0.14	0.14	68	70
Bovine fat					
0	0	0.19, 0.19, 0.19, 0.19	0.19	-	95
1	32	0.18, 0.18, 0.18	0.18	91	90
2	60	0.19, 0.18, 0.20	0.19	98	95
3	90	0.17, 0.18, 0.17	0.17	98	85
Bovine liver					
0	0	0.17, 0.16, 0.15, 0.16	0.16	-	80
1	34	0.17, 0.16, 0.15	0.16	73	80

Sampling interval (nominal months)	Sampling interval (actual days)	Uncorrected residue (mg/kg)	Mean uncorrected residue (mg/kg)	Procedural recovery (%)	Mean recovered uncorrected residue (%)
2	62	0.20, 0.20, 0.20	0.20	94	100
3	92	0.19, 0.18, 0.19	0.18	87	90
Bovine kidney					
0	0	0.19, 0.19, 0.19, 0.20	0.19	-	95
1	29	0.18, 0.18, 0.18	0.18	93	90
2	56	0.19, 0.19, 0.19	0.19	91	95
3	92	0.17, 0.18, 0.19	0.18	85	90
Cow's milk					
0	0	0.21, 0.20, 0.21, 0.21	0.21	-	105
1	36	0.21, 0.20, 0.21	0.21	101	105
2	63	0.19, 0.19, 0.19	0.19	101	95
3	93	0.20, 0.20, 0.20	0.20	102	100

Calculations performed on unrounded values.
No residues were present above the LOQ in control samples. However, there are no data presented in the study report to check this information.

III. CONCLUSIONS

All samples showed a degradation <30% compared to zero time point. Residues of R182281 were stable in bovine muscle, fat, liver, kidney and cow's milk for at least 3 months when stored in the freezer at -0°C.

B.7.1.9 Storage stability in animal matrices

Previous evaluation	Submitted for the purpose of renewal
RMS remark	Acceptable

Report:	KCA 6.1/08. Amic S. (2015), Chlorothalonil - Storage stability of chlorothalonil metabolite R182281 in animal matrices under freezer storage conditions for up to two years. Eurofins Agroscience Service Chem SAS, Vergèze, France. Report Number S12-04421. Syngenta Regulatory Document No: R182281_10047.

Guidelines

OECD Guideline for Testing of Chemicals No. 506, Stability of Pesticide Residues in Stored Commodities, adopted 16 October 2007

EPA Residue Chemistry Test Guidelines, OPPTS 860.1380, Storage Stability Data, August 1996

Commission of the European Communities: Storage Stability of Residue Samples (SANCO 7032/V1/95 rev. 5 22/7/1997).

GLP

The study was carried out according to the principles of Good Laboratory Practice.

EXECUTIVE SUMMARY

Samples of bovine tissues (liver and muscle), bovine milk and poultry eggs were fortified at 0.5 mg/kg with R182281. Duplicate samples were stored under frozen conditions (-20°C) and analysed at intervals up to 24 months (six sampling points). The LOQ for R182281was 0.01 mg/kg.

There was no significant decrease (>30% compared to the zero time value) in the levels of R182281 in bovine liver tested over 24 months when stored at -20°C. There was no significant decrease (>30% compared to the zero time value) in the levels of R182281 in bovine milk and muscle and poultry eggs tested over 18 months when stored at -20°C.

Residues of R182281 were found to be stable in bovine liver for at least 24 months and in bovine muscle, milk and hen's eggs for 18 months when stored in the freezer at -20°C.

A1. Test Materials

The purity of the analytical standard used in this study is listed in Table 7.1.9-1.

Table 7.1.9-1: Purity of analytical standards

Analyte	Standard reference no.	Purity (wt. %)
R 182281	P2	98.7

A2. Test Commodity

The test commodities were untreated homogenised bovine tissues (liver and muscle), bovine milk and poultry eggs purchased from a local supermarket.

A3. Test Facilities

This study was performed at Eurofins Agroscience Services Chem SAS, 75 Chemin de Sommières, 30310 Vergèze, France.

B. STUDY DESIGN AND METHODS

B1. Fortification and Storage of Samples

Homogenised samples were fortified at 0.5 mg/kg with R182281 in acetone/ hydrochloric acid (100:5 v/v). Duplicate samples were stored under frozen conditions (-20°C) and analysed at intervals up to 24 months (six sampling points). Control samples were analysed at zero time and at all other time points to ensure that no residues of R182281 were present at levels above 30% of the LOQ.

B2. Analytical Method

Analysis of the samples was performed according to analytical method GRM005.05.A, renamed as method AGR/MOA/CHL-14 at intervals of 0, 1, 3, 6, 12, 18 and 24 months.

The method involved extraction of the samples of muscle and liver with acetone/5M sulphuric acid followed by centrifugation. Samples of milk were extracted twice with acetonitrile and centrifuged. Samples of egg were extracted with acetonitrile/water and centrifuged. Aliquots of the extracts were taken and diluted in water/ hydrochloric acid. The acidified aliquots were cleaned up by SPE with acetone as

eluting agent. Final determination was by LC-MS/MS, monitoring for the primary transition (m/z 245→182) and the confirmatory transition (m/z 245→210). The LOQ was 0.01 mg/kg.

II. RESULTS AND DISCUSSION

Method Validation

Procedural recoveries were determined using freshly fortified samples at each interval. Samples were fortified with R182281 at 0.5 mg/kg. Individual recoveries, mean recoveries and %RSD are summarised in Table 7.1.9-2.

Table 7.1.9-2: Summary of procedural recoveries for R182281 in animal matrices

Recovery at zero time (%)	Recovery at 1 month (%)	Recovery at 3 months (%)	Recovery at 6 months (%)	Recovery at 12 months (%)	Recovery at 18 months (%)	Recovery at 24 months (%)	Mean recovery (%)	RSD (%)
Bovine milk								
88, 85	90, 82	87, 86	83, 84	89, 90	101,102	76, 73, 76, 79	86	8
Bovine liver								
85, 85	83, 83	84, 97	87, 85	86, 85	99, 100	74, 72	86	8
Bovine muscle								
83, 82	80, 77	98, 92	86, 86	82, 85	98, 100	76, 76	86	8
Poultry eggs								
87, 90	86, 85	85, 84	85, 85	86, 85	103, 99	79, 73, 73, 79	85	8

Storage Stability of Residues

The recoveries of R182281 in various crops stored at -20°C are summarised in Table 7.1.9-3 below. The results are not corrected for freshly fortified recoveries.

Table 7.1.9-3: Freezer storage stability for R182281 at 0.5 mg/kg in animal matrices

Sampling interval (nominal months)	Sampling interval (actual days)	Uncorrected residue (mg/kg, mg/L for milk)	Mean uncorrected residue (mg/kg, mg/L for milk)	Mean procedural recovery (%)	Mean recovered uncorrected residue (%)
Bovine milk					
0	0	0.41, 0.44, 0.42	0.42	87	84
1	32	0.41, 0.40	0.40	86	80
3	95	0.43, 0.46	0.45	87	90
6	180	0.35, 0.37	0.36	83	72
12	364	0.45, 0.43	0.44	89	88
18	544	0.51, 0.50	0.51	101	102
24	731, 843	0.21, 0.27, 0.24, 0.22	0.24	76	**48**

Sampling interval (nominal months)	Sampling interval (actual days)	Uncorrected residue (mg/kg, mg/L for milk)	Mean uncorrected residue (mg/kg, mg/L for milk)	Mean procedural recovery (%)	Mean recovered uncorrected residue (%)
Bovine Liver					
0	0	0.43, 0.43, 0.43	0.43	85	86
1	32	0.39, 0.39	0.39	83	78
3	95	0.46, 0.44	0.45	91	90
6	180	0.38, 0.39	0.39	86	78
12	364	0.42, 0.44	0.43	86	86
18	544	0.51, 0.50	0.50	99	100
24	731	0.29, 0.39	0.34	73	**68**
Bovine Muscle					
0	0	0.42, 0.42, 0.42	0.42	83	84
1	32	0.39, 0.40	0.40	78	80
3	95	0.47, 0.46	0.47	95	94
6	180	0.37, 0.38	0.38	86	76
12	364	0.42, 0.46	0.44	84	88
18	544	0.50, 0.48	0.49	99	98
24	843	0.27, 0.25	0.26	76	**52**
Poultry Eggs					
0	0	0.44, 0.43, 0.42	0.43	89	86
1	32	0.41, 0.42	0.42	86	84
3	95	0.41, 0.43	0.42	84	84
6	180	0.38, 0.38	0.38	85	76
12	364	0.43, 0.42	0.43	85	86
18	544	0.50, 0.50	0.50	101	100
24	731, 843	0.31, 0.27, 0.25, 0.23	0.27	76	**54**

Calculations performed on unrounded values.

No residues were present above the LOQ in control samples.

Values in bold are outside the acceptable range of 70-120% recovery.

III. CONCLUSIONS

There was no significant decrease (>30% as compared to the zero-time value) in the observed residue levels of R182281 after deep frozen storage for 18 months. A decrease >30% was observed in residue levels in bovine milk, muscle and poultry eggs after 24 months.

Residues of R182281 were found to be stable in bovine liver for at least 24 months and in bovine muscle, bovine milk and hen's eggs for 18 months when stored in the freezer at -20°C.

B.7.1.10 Storage stability in sample extracts

Previous evaluation	Submitted for the purpose of renewal
RMS remark	Supplementary study regarding storage stability of sample extracts

Report:	K-CA 6.1/09. Lister, N. (2000), Chlorothalonil: Validation of SOP RAM 320/01 for the determination of residues in crops. Syngenta, Jealott's Hill International Research Centre, Bracknell, United Kingdom. Syngenta Report Number RJ2872B. Syngenta File No: R44686/0099.

Guidelines

~~Not applicable for the storage stability of sample extracts.~~ The study was evaluated as compliant with testing guideline OECD Test No. 506.

GLP

The study was carried out according to the principles of Good Laboratory Practice.

EXECUTIVE SUMMARY

The storage stability of sample extracts was determined as part of the validation of method SOP RAM 320/01. For the validation, see B.5.1.2.5 (K-CA 4.1.2/22). Extracts of homogenised pear fruit, soya bean seed, barley grain and barley straw that were fortified at 0.01, 0.1 and 1.0 mg/kg with chlorothalonil were stored at < 7°C and analysed at intervals up to 35 days after extraction. The final measurement extracts were stored at < -18°C and reanalysed 1, 3 and 7 days after the initial analysis.

Extracts of representative crop matrices stored in extraction solvent (acetone/5M sulphuric acid) at temperatures of < 7°C were shown to be stable for a period of 35 days. The final extracts in toluene were shown to be stable for a period of at least 7 days when stored at < -18°C.

A1. Test Materials

The purity of the analytical standards used in this study is listed in Table 7.1.10-1.

Table 7.1.10-1: Purity of analytical standards

Analyte	Standard reference no.	Purity (wt. %)
Chlorothalonil	ASJ10125-03S	99.6

A2. Test Commodity

The test commodities were pear fruit, soya bean seed, barley grain and barley straw. Samples were purchased from a local supermarket with the exception of the barley samples which were taken from a Syngenta field trial.

A3. Test Facilities

This study was performed at Syngenta, Jealott's Hill International Research Centre, Bracknell, Berkshire, RG42 6EY, UK.

B. STUDY DESIGN AND METHODS

B1. Fortification and Storage of extracts

Samples were fortified at 0.01, 0.10 and 1.0 mg/kg with chlorothalonil in acetone and extracted according to method RAM 320/01. Duplicate samples were used, except for the 0.01 mg/kg fortification, which was done in quadruplicate. Sample extracts in acetone/5M sulphuric acid taken immediately after homogenisation were stored in a cold room at < 7°C and analysed at intervals up to 35 days. The stability of the final measurement extracts in toluene (from SPE clean up) was assessed by retaining the samples in the vials at temperatures <-18°C and reanalysing them 1, 3 and 7 days after the initial analysis.

B2. Analytical Method

Analysis of the samples was performed according to analytical method SOP RAM 320/01. A full method description and validation data are presented in B.5.1.2.5 (K-CA 4.1.2/22). In summary, the toluene aliquots obtained from SPE clean up were measured with GC-MS monitoring for the primary transition (m/z 266→264). The LOQ was established to be 0.01 mg/kg.

II. RESULTS AND DISCUSSION

Storage Stability of Extracts

The recoveries of chlorothalonil in sample extracts stored at < 7°C and ≤ -18°C are summarised in Table 7.1.10-2 and Table 7.1.10-3, respectively.

Table 7.1.10-2: Storage stability of chlorothalonil in crop extraction solvent stored at <7°C

Crop Matrix	Recovery level (mg/kg)	Average Chlorothalonil recovery							
		Day 0		Day 7-10		Day 13-17		Day 28-35	
		mg/kg	%	mg/kg	%	mg/kg	%	mg/kg	%
Pear fruit	0.01	0.010	100	0.010	100	0.009	90	nd	nd
	0.1	0.091	100	0.090	99	0.081	89	nd	nd
	1.0	0.98	100	0.94	96	0.82	84	nd	nd
Pear fruit	0.01	0.008	100	nd	nd	nd	nd	0.009	113
	0.1	0.077	100	nd	nd	nd	nd	0.095	**123**
	1.0	0.86	100	nd	nd	nd	nd	1.05	**122**
Soya bean seed	0.01	0.009	100	0.008	89	0.008	89	0.008	89
	0.1	0.093	100	0.076	82	0.080	86	0.070	75
	1.0	1.02	100	0.82	80	0.83	81	0.81	79
Barley grain	0.01	0.008	100	0.009	113	0.008	100	0.008	100
	0.1	0.074	100	0.079	107	0.076	103	0.084	114
	1.0	0.86	100	0.89	103	0.80	93	1.01	117
Barley straw	0.01	0.009	100	0.009	100	0.009	100	0.009	100
	0.1	0.073	100	0.088	**121**	0.091	**125**	0.092	**126**
	1.0	0.79	100	0.88	111	1.00	**127**	0.90	114

nd = not determined
Values in bold are outside the acceptable range of 70-120% recovery.

Table 7.1.10-3: Storage stability of chlorothalonil in toluene stored at <-18°C

Crop Matrix	Recovery level (mg/kg)	Average Chlorothalonil recovery (mg/kg)							
		Day 0		Day 1		Day 3		Day 7	
		mg/kg	%	mg/kg	%	mg/kg	%	mg/kg	%
Pear fruit	0.01	0.01	100	0.010	100	0.009	90	0.010	100
	0.1	0.091	100	0.089	98	0.081	89	0.089	98
	1.0	0.98	100	0.99	101	0.88	90	0.96	98
Soya bean seed	0.01	0.009	100	0.009	100	0.009	100	0.009	100
	0.1	0.091	100	0.088	97	0.089	98	0.091	100
	1.0	1.02	100	1.04	102	1.00	98	1.07	105
Barley grain	0.01	0.008	100	0.008	100	0.008	100	0.008	100
	0.1	0.074	100	0.073	99	0.072	97	0.070	95
	1.0	0.86	100	0.83	97	0.82	95	0.87	101
Barley straw	0.01	0.009	100	0.008	89	nd	nd	0.010	111
	0.1	0.073	100	0.073	100	nd	nd	0.085	116
	1.0	0.79	100	0.79	100	nd	nd	0.93	118

nd = not determined

III. CONCLUSIONS

Extracts of representative crop matrices stored in extraction solvent at temperatures of < 7°C were shown to be stable for a period of at least 35 days. The final extracts in toluene were shown to be stable for a period of at least 7 days when stored at < -18°C.

B.7.1.11 Storage stability in sample extracts

Previous evaluation	Submitted for the purpose of renewal
RMS remark	Supplementary study regarding storage stability of sample extracts

Report:	K-CA 6.1/10. McGill, C. and Robinson, N. (2002), Chlorothalonil metabolite R182281 (SDS-3701): Validation of analytical method 384/01 for the determination of residues in bovine muscle, fat, kidney, liver, milk and hen's eggs. Syngenta, Jealott's Hill International Research Centre, Bracknell, United Kingdom. Syngenta Report Number RJ3312B. Syngenta File No: R44686/3317, including amendment issued in 2015.

Guidelines

~~Not applicable.~~ The study was evaluated as compliant with testing guideline OECD Test No. 506.

GLP

The study was carried out according to the principles of Good Laboratory Practice. The amendment was not performed under GLP.

EXECUTIVE SUMMARY

The storage stability of sample extracts was determined as part of the validation of method SOP RAM

384/01. Extracts of homogenised muscle, fat, liver, kidney, milk and egg, that were fortified at 0.01 and 0.1 mg/kg with R182281 before extraction, were stored at < 7°C and analysed at intervals up to 35 days after extraction. The final measurement extracts were stored at < 7°C and reanalysed 7 days after the initial analysis.

The applicant concluded that extracts of representative matrices stored in extraction solvent at temperatures of < 7°C were shown to be stable for 30-35 days. The final extracts in acetone/water were shown to be stable for 7 days.

A1. Test Materials

The purity of the analytical standards used in this study is listed in Table 7.1.11-1.

Table 7.1.11-1: Purity of analytical standards

Analyte	Standard reference no.	Purity (wt. %)
R182281	ASJ10209-02	Not stated

A2. Test Commodity

The test commodities were bovine muscle, fat, kidney, liver, milk and hen's eggs. Samples were taken from pre-prepared control samples available at Syngenta with the exception of hen's eggs which were purchased from a local supermarket.

A3. Test Facilities

This study was performed at Syngenta, Jealott's Hill International Research Centre, Bracknell, Berkshire, RG42 6EY, UK.

B. STUDY DESIGN AND METHODS

B1. Fortification and Storage of extracts

Samples were fortified at 0.01 and 0.1 mg/kg with R182281 in acidified acetone and extracted according to method RAM 384/01 at 5 replicates. Sample extracts taken immediately after homogenisation were stored at < 7°C and analysed 30 to 35 days after extraction. The stability of the final measurement extracts in acetonitrile/water (50:50) was assessed by retaining the samples in the vials at temperatures < 7°C and reanalysing 7 days after the initial analysis.

B2. Analytical Method

Analysis of the samples was performed according to analytical method RAM 384/01. A full method description and validation data are presented in Addendum 13 to the DAR.

II. RESULTS AND DISCUSSION

Storage Stability of Extracts

The recoveries of chlorothalonil in sample extracts stored at < 7°C are summarised in Table 7.1.11-2 and Table 7.1.11-3 below.

Table 7.1.11-2: Storage stability of R182281 in extraction solvent stored at <7°C[a]

Matrix	Recovery level (mg/kg)	Storage interval (days)	R182281 recovery (mean of 5 replicates)	
			mg/kg	%
Muscle	0.01	0	0.010	100
		30	0.009	90
	0.1	0	0.094	94
		30	0.092	92
Fat	0.01	0	0.009	90
		35	0.009	90
	0.1	0	0.086	86
		35	0.084	84
Kidney	0.01	0	0.010	100
		34	0.010	100
	0.1	0	0.097	97
		34	0.096	96
Liver	0.01	0	0.009	90
		34	0.010	100
	0.1	0	0.091	91
		34	0.083	83
Milk	0.01	0	0.008	80
		30	0.011	110
	0.1	0	0.090	90
		30	0.100	100
Egg	0.01	0	0.008	80
		31	0.007	70
	0.1	0	0.081	81
		31	0.094	94

[a] For muscle, fat, liver and kidney the extraction solvent used was acetone/5 M H_2SO_4 95:5 (v/v), for milk acetonitrile was the extraction solvent and for egg the extraction solvent was acetonitrile/water, 3:1 (v/v).

Table 7.1.11-3: Storage stability of R182281 in acetone/water 50/50 v/v stored at <7°C

Matrix	Recovery level (mg/kg)	Storage interval (days)	R182281 recovery (mean of 5 replicates)	
			mg/kg	%
Muscle	0.01	0	0.010	100
		7	0.011	110
	0.10	0	0.094	94
		7	0.095	95
Fat	0.01	0	0.009	90
		7	0.009	90
	0.10	0	0.086	86
		7	0.084	84

Matrix	Recovery level	Storage interval	R182281 recovery (mean of 5 replicates)	
Kidney	0.01	0	0.010	100
		7	0.010	100
	0.10	0	0.097	97
		7	0.097	97
Liver	0.01	0	0.009	90
		7	0.008	80
	0.10	0	0.091	91
		7	0.079	79
Milk	0.01	0	0.008	80
		7	0.008	80
	0.10	0	0.092	92
		7	0.085	85
Egg	0.01	0	0.008	80
		7	0.008	80
	0.10	0	0.081	81
		7	0.088	88

III. CONCLUSIONS

The applicant concluded that R182281 stored in extraction solvent at temperatures of < 7°C was shown to be stable for 30-35 days based on the amended data. The final extracts in acetone /water (50/50 v/v) were shown to be stable for 7 days for all matrices.

B.7.1.12 Storage stability of SDS-3701 in cow's milk and tissues

Previous evaluation	In Addendum 14 to DAR
RMS remark	Acceptable

Characteristics

reference	:	King C. and Prince P., 1995	GLP statement	:	Yes
type of study	:	stability of residues in cow's milk and tissues	guideline	:	EPA 171-4e
year of execution	:	1994-1995	fortification level	:	0.2-1.0 mg/kg
test substance	:	SDS-3701, analytical standard, purity 100%.	storage conditions		ca. –23°C in the dark
specimen	:	cow's milk and tissues	storage period	:	1 year

Study design

Ten gram samples of locally obtained cow's whole milk, muscle, liver and body fat were fortified with SDS-3701 at a rate of 0.2 mg/kg (muscle and fat) or 1.0 mg/kg (liver and milk). Fortified and control samples were stored in a freezer between -2°C and -23°C (median -23°C) for up to 1 year. Samples were removed for analysis after 0 days (prior to freezing), 1, 7 and 14 days, and after 1, 2, 3, 6, 9 and 12 months. At each sampling point four fortified stored samples and three controls were analyzed. Two controls were used for concurrent recoveries. Residues of SDS-3701 were extracted with acidified acetone and partitioned into diethyl ether. After derivatisation with diazomethane and alumina column chromatography clean up, the residues were quantified by GC-ECD.

Results

Residues of SDS-3701 in control samples of milk, muscle, liver and body fat were not detectable (<0.01 mg/kg) at all sampling intervals. Concentrations and recoveries of stored fortified and freshly fortified samples at each sampling interval are shown in Table 7.6.3.2. Values for concurrent recoveries of all 4 matrices ranged between 70 and 120%, with means for each matrix ranging between 86 and 103% and coefficients of variation between 7% an 13%.

Statistical analysis (regression analysis for downward trend) was performed on the residue values of SDS-3701 recorded during the study. This analysis was based on corrected residues, in case day-to-day variation in concurrent recovery was significantly higher than within-day variation. Otherwise, uncorrected residues were used. This analysis demonstrated that residues of SDS-3701 were stable in milk, but that there was a significant downward trend in muscle, body fat and liver, with a loss over the 1 year study period of 8%, 9% and 17%, respectively.

Table 7.6.3.2 Recovery of SDS-3701 from cow's milk and tissues fortified with 0.2-1.0 mg/kg SDS-3701 after storage at -23°C

	muscle		body fat		liver		milk	
storage period	**residue stored sample (mg/kg)[A]**	**inconcur-rent recovery (%)[B]**	**residue stored sample (mg/kg)[A]**	**inconcur-rent recovery (%)[B]**	**residue stored sample (mg/kg)[A]**	**inconcur-rent recovery (%)[B]**	**residue stored sample (mg/kg)[A]**	**inconcur-rent recovery (%)[B]**
0 day (unfrozen)	0.18	85-90	0.20	90-100	0.95	91-99	0.91	88-89
1 day	0.20	100-105	0.20	90-115	0.87	73-96	0.97	102-106
7 days	0.20	95	0.21	85-100	0.72	73-82	0.95	97-101
14 days	0.17	80-90	0.20	95-105	0.89	99-109	1.04	98-107
1 month	0.18	80-100	0.17	90	0.84	79-88	1.05	113-116
2 months	0.19	85-100	0.17	90	0.73	77-82	0.96	95
3 months	0.19	90-95	0.16	80	0.80	99-100	1.11	100-104
6 months	0.16	70-80	0.16	75	0.76	71-75	1.17	105-109
9 months	0.21	110	0.19	100-105	0.63	77-80	1.04	105-108
12 months	0.16	85-90	0.22	115-120	0.67	86-88	1.07	103-110

(A) Average of four samples.

(B) Range for two samples.

Conclusions

Under frozen conditions, residues of SDS-3701 are stable in milk for one year, but in muscle, body fat and liver a loss over the one year study period occurs of 8%, 9% and 17%, respectively.

Guidelines and limitations

The study complied with the guideline OECD Test No. 506 ~~(Lundehn document, Appendix H)~~ and is acceptable.

B.7.2 Metabolism, distribution and expression of residues

B.7.2.1 Plants

For the initial peer review, the metabolism of chlorothalonil has been studied in lettuce, tomato, carrot, celery, snap beans (French beans), wheat and peas using ^{14}C-chlorothalonil labelled in the phenyl position. No additional studies on plant metabolism have been submitted within the framework of the renewal of chlorothalonil.

B.7.2.1.1 Plant metabolism in lettuce

Previous evaluation	In DAR
RMS remark	~~Acceptable~~ Not conclusive; It was noted during the expert Peer Review Meeting (# 164), that for some of the metabolism studies available for the evaluation, the proportions for the identified metabolites were (probably) calculated based on the total residues in the extracted fractions and not based on the total residues in extracted commodity. A lot of inconsistencies across of all the studies have been observed and it was agreed by the experts that the studies cannot be considered as reliable.

Characteristics

reference	:	Zeneca, Nelsen, 1985	application rate	:	equivalent to 1.75 kg ai/ha (3 pints of Bravo 500/acre)
type of study	:	plant metabolism	concentration	:	9 g ai/l 80-fold diluted Bravo 500
year of execution	:	1984	frequency	:	4 applications
test substance	:	[^{14}C-U-phenyl]-chlorothalonil (radiochem. pur. 99%) and chlorothalonil (pur. >97%)	interval	:	4-5 d
plant	:	lettuce	total rate	:	7 kg ai/ha
GLP statement	:	yes	exposure	:	foliar by syringe
guideline	:	not applicable	PHI	:	1, 3, 7, 10, 14, 21 d

Study design

Lettuce plants, grown in pots in growth chambers, received four foliar applications of [^{14}C-U-phenyl]-labelled chlorothalonil, applied as a Bravo® 500 spray suspension by syringe. These plants were dosed 4 times at 4-5 day intervals at a rate equivalent to approximately 1.75 kg ai/ha (i.e., 3 pints Bravo 500/acre) per application (in total equivalent to 7.0 kg ai/ha). Applications started 22 days post-emergence. Plants were harvested at 1, 3, 7, 10, 14 and 21 days PHI, ground in the presences of dry ice and processed for analysis. The total ^{14}C-residue from plant material from each pot was determined by combustion/LSC. Individual plant samples were blended/extracted with a mixture of acetone/hydrochloric acid. Acetone was removed and the resulting aqueous sample was partitioned with diethyl ether. Both phases were monitored for total radiolabel. The organosoluble fraction was analysed by HPLC.

Results

The mass balance is shown in the following table:

Mass Balance from Lettuce

Sample Day	Replicates	% Extracted[1]	% Post Extraction Solids (PES) [1]	% Total[1]
1	1	102.6	1.1	103.7
3	1	101.7	4.7	106.4
7	1	91.7	1.9	93.6
10	1	99.1	2.5	101.6
14	2	95.4	2.4	97.8
21	2	97.2	3.6	100.8

[1] Percentage of total as determined by combustion

The ^{14}C-residue determined in ^{14}C-chlorothalonil treated lettuce at the various days after last treatment (DALT) was as follows (standards were also available for SDS-19221, SDS-47524, SDS-47525, SDS-46851):

Table 7.1.1: Distribution of ^{14}C-residues

DALT	Total [1]	Organosoluble [2]	Water soluble [2]	PES [3]	Chlorothalonil [3, 4]	SDS-3701 [3, 4]	Polar nonextractable [3, 5]
days	mg/kg	%	%	%	%	%	%
LETTUCE							
1	118	94	5.7	1.0	89	1.5	5.6
3	170	94	5.9	4.5	87	0.9	5.6
7	147	93	7.0	2.0	88	1.4	7.0
10	139	95	4.8	2.4	90	1.5	4.7
14	153	95	5.4	2.4	89	1.8	5.3
21	158	95	5.1	3.6	87	2.0	4.9

[1] Expressed as chlorothalonil equivalents.
[2] Relative to the total extracted residue (organosoluble + watersoluble).
[3] Relative to ^{14}C recovered ~~the amount of radiolabel determined by combustion LSC~~.
[4] Determined by HPLC and confirmed by GC-MS in the organosoluble extract.
[5] Radioactivity remaining in the aqueous phase after partitioning

Conclusion

Total radiolabel residue levels in lettuce, treated 4 times with 1.75 kg ai/ha, varied from 118 to 170 mg/kg equivalents/kg lettuce and was independent of the DALT. Chlorothalonil metabolism in lettuce resulted in a single, identifiable and quantifiable metabolite, SDS-3701, accounting for maximally 2% of the total residue recovered. The amount of residue that was not extracted varied from 1.0-4.5% of the total residue in lettuce (calculated by the rapporteur to be approx. 1-7 mg equivalents/kg) and was independent of the PHI. The amount of polar, watersoluble residue was 4.7-7.0% (calculated by the rapporteur to be approx. 7-10 mg equivalents/kg) and independent of the PHI. The major identified and quantified residue in lettuce was chlorothalonil, accounting for over 85% of the total residue.

Limitations

It should be noted that the total amounts of unidentified organosoluble and watersoluble residues exceeded 0.05 mg equivalents/kg. However, since approx. 90% of the total residue was identified and only 4.5% remained not extracted, the study is considered acceptable for the overall evaluation.

B.7.2.1.2 Plant metabolism in tomato

Previous evaluation	In DAR
RMS remark	~~Acceptable~~ Not conclusive; It was noted during the expert Peer Review Meeting (# 164), that for some of the metabolism studies available for the evaluation, the proportions for the identified metabolites were (probably) calculated based on the total residues in the extracted fractions and not based on the total residues in extracted commodity. A lot of inconsistencies across of all the studies have been observed and it was agreed by the experts that the studies cannot be considered as reliable.

Characteristics

reference	:	Zeneca, Nelsen, 1988	application rate	:	equivalent to 2.33 kg ai/ha (4 pints of Bravo 500/acre)
type of study	:	plant metabolism	concentration	:	1.47 g ai/l 200-fold diluted Bravo 500
year of execution	:	1985	frequency	:	3 applications
test substance	:	[^{14}C-U-phenyl]-chlorothalonil (radiochem. pur. 98%) and chlorothalonil (pur. >99.9%)	interval	:	7 d
plant	:	tomato	total rate	:	7 kg ai/ha
GLP statement	:	yes	exposure	:	foliar
guideline	:	not applicable	PHI	:	1, 7, 14 days

Study design

Tomato plants (1/pot), grown in greenhouses, received three weekly applications of [^{14}C-U-phenyl]-labelled chlorothalonil. The test material was applied as a Bravo 500 spray suspension at a rate of 2.33 kg ai/ha/application (i.e. 4.0 pints of Bravo 500/acre) with an air brush. The total amount of active ingredient applied was equivalent to 7.0 kg/ha. At 1, 7 and 14 days PHI, tomato fruit and vines were harvested. Vine samples were chopped and then ground with dry ice. The dry ice was allowed to sublime and the remaining sample was stored frozen (-20 ºC). Tomato (fruit) samples were either immediately subjected to an organic solvent surface strip and macerated as above or frozen whole. With the exceptions noted and discussed below, the procedures and analytical methods used in this study were identical to those described in study 1 (Zeneca, Nelsen, 1985). Organosoluble residue was analysed by HPLC. Since considerable amounts of polar, water-soluble ^{14}C-residues were present in the fruit, considerable effort was directed to the characterization of these residues. Techniques employed in this study included gel permeation chromatography (GPC), enzyme hydrolysis, base solvolysis, TLC, GC-MS, alcoholic acid hydrolysis, diazomethane derivatisation and appropriate chromatographic comparison to authentic molecular weight calibration standards for GPC. Furthermore, in an effort to distinguish between surface and in-fruit residues, two or three tomatoes (fruit) were individually subjected to a

dichloromethane rinse to remove surface residue. The surface stripped sample was then ground with dry ice and the dry ice allowed to sublime. The ground sample was held frozen until analysis.

Results

The distribution of radiolabel residues from ^{14}C-chlorothalonil treated tomatoes was as follows:

Table 7.1.2. Distribution of ^{14}C-residues

PHI	Total [1]	Rinse [2]	Organo-soluble [2]	Water-soluble [2]	Not extracted [2]	Chlorotha-lonil [3]	SDS-3701 [3]	Largest unknown(s) [3]		Total unknown [3]
								Rinse	Organo soluble	
d	mg/kg	%	%	% [mg/kg] [4]	%	% [mg/kg] [4]	% [mg/kg] [4]	%	%	%
FRUIT										
1	2.6	75	4.2	19 [0.48]	2.0	95 [2.0]	2.0 [0.04]	0.62		3.4
7	0.7	56	3.5	32 [0.22]	9.1	94 [0.36]	3.3 [0.01]	0.57		4.1
14	0.6	61	3.3	32 [0.19]	4.2	91 [0.35]	4.6 [0.02]	0.70		4.8
VINES										
1	21	na [5]	80	13 [2.7]	6.8	87 [15]	4.2 [0.7]	0.9	0.4	4.3
7	13	na	67	19 [2.5]	14	81 [6.9]	11 [1.0]	1.5	1.2	8.1
14	14	na	56	30 [4.2]	15	75 [5.8]	14 [1.1]	1.8	2.1	12

[1] Expressed as chlorothalonil equivalents (determined by combustion LSC).

[2] Two or three tomatoes were rinsed with dichloromethane at each sampling time point. The organosoluble, watersoluble, and not extracted levels refer to levels determined following rinsing with dichloromethane. Residues were determined by LSC.

[3] Expressed as percentage of the combined organic rinse and organosoluble extract.

[4] Expressed in mg/kg of tomato or vine.

[5] na = not applicable

The major identified component of the residue in/on tomato fruit and vines was the parent compound chlorothalonil, which was calculated by the rapporteur to account for 50-76% and 41-73% of the total residue, respectively. The minor identified residue component was SDS-3701, calculated to account for less than 5% of the total residue in/on fruit. The SDS-3701 level in tomato vines was calculated to be maximally 8% of the total residue. Since tomato vines do not represent a food or feed commodity, no further work was performed to identify/characterize vine residues. Furthermore, since the not extracted residue in the fruit represented less than 25% of the total radioactive residue at 7 days PHI, no further work was carried out to characterize this fraction. However, since the polar, watersoluble ^{14}C-residue in tomato fruit represented up to 32% of the total residue at 7 and 14 days PHI, considerable additional effort was made in an attempt to characterize/identify these residues. Attempts to further identify the

watersoluble residue were only partly successful. Indications were provided that at least one-half of the watersoluble extract may have been methylated by diazomethane derivatisation to SDS-3316 (5-chloro-2,4,6-trimethoxyisophthalonitrile).

Conclusion

Total residue levels on tomato fruit decreased from 2.6 to 0.6 mg/kg whole product 1 and 14 days PHI after treatment with 3 subsequent applications of 2.33 kg ai/ha. The major identified component of the residue in/on tomato fruit and vines was the parent compound chlorothalonil, accounting for 50-76% and 41-73% of the total residue, respectively. The second major identified residue component was SDS-3701, accounting for less than 5% of the total residue in/or fruit. The SDS-3701 level in/on tomato vines was maximally 8% of the total residue. Around or below 10 and 15% of the total residue in/on fruit and vines, respectively, remained not extracted. Watersoluble residues present at levels corresponding to up to approx. 0.5 and 4.2 mg equivalents/kg in fruit and vines, respectively, remained unidentified.

Limitations

Total amounts of unidentified organosoluble and watersoluble residues exceeded 0.05 mg equivalents/kg. However, around or more than 95% and 90% of the organosoluble residue in /on fruit and vines, respectively, was identified. Watersoluble residues were only poorly characterised, despite much effort. In total, around or over 50% of the total residue was identified and below 10 and 15% of the total residue in/on tomato fruit and vines remained not extracted, respectively. Therefore, the study is considered acceptable for the overall evaluation. Because of the limited proof, a possible *in vivo* metabolite being derivatised to SDS-3316 was not included as identified metabolite.

B.7.2.1.3 Plant metabolism in carrot

Previous evaluation	In DAR
RMS remark	~~Acceptable~~ Not conclusive; It was noted during the expert Peer Review Meeting (# 164), that for some of the metabolism studies available for the evaluation, the proportions for the identified metabolites were (probably) calculated based on the total residues in the extracted fractions and not based on the total residues in extracted commodity. A lot of inconsistencies across of all the studies have been observed and it was agreed by the experts that the studies cannot be considered as reliable.

Characteristics

reference	:	Zeneca, Nelsen, 1987	application rate	:	equivalent to 1.6 kg ai/ha (2.75 pints of Bravo 500/acre)
type of study	:	plant metabolism	concentration	:	1.01 g ai/l 200-fold diluted Bravo 500
year of execution	:	1986	frequency	:	3 applications
test substance	:	[^{14}C-U-phenyl]-chlorothalonil (radiochem. pur. 99.4%) and chlorothalonil (pur. >99.9%)	interval	:	7 d
plant	:	carrot	total rate	:	4.8 kg/ha
GLP statement	:	yes	exposure	:	foliar
guideline	:	not applicable	PHI	:	1, 7, 14, 21 days

Study design

Carrot plants (4 carrot seeds/pot), grown in growth chambers, received three applications of [^{14}C-U-phenyl]-chlorothalonil at 7-day intervals. This material was applied to the carrot foliage as a Bravo 500 formulation at a rate of 1.6 kg ai/ha for each application with an air brush. At 1, 7, 14 and 21 days PHI, plants were harvested, separated into foliage and roots, and analysed for ^{14}C-residues. With the exceptions noted and discussed below, the procedures and analytical methods used in this study were identical to those described in study 1 (Zeneca, Nelsen, 1985). Immediately after harvest, two of the three carrots were subjected to a dichloromethane rinse to remove surface residue. At the first harvest, all three root samples were subjected to the organic solvent rinse. The surface stripped sample was then ground with dry ice and the dry ice allowed to sublime. The ground sample was held frozen until analysis. A similar organic solvent stripping procedure was performed on each of the corresponding foliage samples. These samples were then ground and stored as described for the root samples. The total residue remaining in the root and foliage samples after removal of the surface residue was determined by combustion of the ground samples.

Results

The total residues for ^{14}C-chlorothalonil treated carrots, both roots and foliage, were as follows:

Table 7.1.3: Distribution of ^{14}C-residues [1]

PHI	Total	Rinse	Organosoluble	Water soluble	Not extracted	Chlorothalonil	SDS-3701
days	mg/kg	mg/kg	% [mg/kg]	% [mg/kg]	% [mg/kg]	% [mg/kg]	% [mg/kg]
ROOT							
1	0.07	0.02					
7	0.02	0.01					
14	0.01	<0.01					
21	0.04	<0.01	53 [0.024]	18 [0.008]	29 [0.013]	79 [2] [0.019]	6.2 [2] [0.0015]

PHI	Total	Rinse	Organosoluble	Water soluble	Not extracted	Chlorothalonil	SDS-3701
days	mg/kg	mg/kg	% [mg/kg]	% [mg/kg]	% [mg/kg]	% [mg/kg]	% [mg/kg]
FOLIAGE							
1	36	12	27[3] [6.48]	31[3] [7.44]	42[3] [10.08]	14 [3] [3.36]	3.4 [3] [0.82]
7	20	2.6	27[3] [4.70]	30[3] [5.22]	44[3] [7.66]	15 [3] [2.61]	2.4 [3] [0.42]
14	36	2.0	24[3] [8.16]	31[3] [10.54]	46[3] [15.64]	9.0 [3] [3.06]	4.3 [3] [1.46]
21	13	2.7	32[3] [3.30]	29[3] [2.99]	39[3] [4.02]	4.0 [3] [0.41]	12 [3] [1.24]

[1] Expressed in percentages or in mg/kg ^{14}C-chlorothalonil equivalents in whole product, unless otherwise stated.
[2] Percentage from the organosoluble fraction, including rinse.
[3] Percentage in the rinsed foliage.

Because of the rather low levels of residue in the treated roots, only one (21 days PHI) sample was successfully processed and analysed by HPLC. The residue levels in the watersoluble and not extracted fractions were around or below 0.01 mg eq/kg and about 80% of the organosoluble fraction was identified as chlorothalonil and 6.2% as SDS-3701. The major components of the foliage rinse were chlorothalonil (76-97%) and SDS-3701 (1.6-15%). In the rinsed foliage residue, 24-32% was organosoluble, 29-31% was watersoluble, and 39-46% was not extracted. HPLC analysis of the organosoluble fraction showed that chlorothalonil was the major component with varying amounts of the hydroxy metabolite (SDS-3701) present (2.4-12% of the rinsed foliage residue). Furthermore, radiolabel was recovered at retention times on HPLC that are associated with SDS-46851, SDS-19221, and SDS-47524. Attempts to further characterize the residues in the watersoluble and not extracted part by acid hydrolysis/HPLC were unsuccessful. No further work was done with the foliage, since this does not represent a food commodity.

Conclusion

Total residue levels in/on root and foliage, respectively, were 0.01-0.07 and 13-36 mg equivalents/kg whole product 1-21 days after treatment with 3 applications of 1.6 kg ai/ha. The only compounds in the residue that were identified were chlorothalonil (79% of rinse and organosoluble residue in roots) and SDS-3701 (6.2% of rinse and organosoluble residue in roots). The levels at 21 days PHI were approx. 0.019 and 0.0015 mg/kg for chlorothalonil and SDS-3701, respectively. Also some minor, not completely identified compounds were found at very low levels. Translocation of ^{14}C-residue from the site of application (foliage) to the edible portion (root) is very small. Because of the limited proof, the potential minor metabolites SDS-46851, SDS-19221 and SDS-47524 were not included as identified metabolite.

Limitations

Not applicable.

B.7.2.1.4 Plant metabolism in celery

Previous evaluation	In DAR
RMS remark	~~Acceptable~~ Not conclusive; It was noted during the expert Peer Review Meeting (# 164), that for some of the metabolism studies available for the evaluation, the proportions for the identified metabolites were (probably) calculated based on the total residues in the extracted fractions and not based on the total residues in extracted commodity. A lot of inconsistencies across of all the studies have been observed and it was agreed by the experts that the studies cannot be considered as reliable.

Characteristics

reference	:	Zeneca, Huhtanen, 1992	application rate	:	equivalent to 2.5 kg ai/ha (2.25 lbs of Bravo 720/acre)
type of study	:	plant metabolism	concentration	:	not specified
year of execution	:	1990	frequency	:	12 applications
test substance	:	[^{14}C-U-phenyl]-chlorothalonil (radiochem. pur. >99%) and chlorothalonil (pur. 99.6%)	interval	:	6-8 days
plant	:	celery	total rate	:	30 kg/ha
GLP statement	:	yes	exposure	:	by spray on foliage, stalks, and soil
guideline	:	not applicable	PHI	:	7, 21 days

Study design

Celery plants, grown outdoors in a muck soil, were treated with 12 applications of [^{14}C-U-phenyl]-chlorothalonil, formulated as Bravo 720. The application rate was 2.5 kg ai/ha (2.23 lbs. ai/acre) per treatment for a total of 30 kg ai/ha. The interval between applications was 6-8 days. The test substance was applied to foliage, stalks and soil. Plants were harvested at 7 and 21 days PHI, separated into foliage and stalks and analysed for total ^{14}C-residues and metabolites. Twelve plants were harvested at 7 days PHI. The remaining twelve plants were harvested at 21 days PHI. Three control plants were harvested at the same time as the 7 day and 21 day post treatment harvests. In general, the procedures and analytical methods used in this study were identical to those described in study 1 (Zeneca, Nelsen, 1985). Samples were extracted three times with either 5 mL/g acetone/1M HCl (4:1), or 5 mL/g acetone/0.3M phosphate buffer pH 6-7 (4:1). Material was centrifuged and the phases separated. The acetone was then removed and partitioned with ether. The method of analysis required 2 solvent systems to elute the non-polar residues.

The HPLC procedure was capable of separating and quantifying chlorothalonil and its five known soil metabolites (among others SDS-3701, SDS-46851) as identified in study 1 (Zeneca, Nelsen, 1985). The watersoluble and not extracted fractions were treated by acid hydrolysis and hydrolytic enzymes, and analysed by TLC, HPLC and liquid scintillation or flow-trough detection. Analytical results were routinely compared to authentic standards of chlorothalonil, chlorothalonil soil metabolites or chlorothalonil mono- and diglutathione derivatives.

Results

The distribution of ^{14}C-residues was as follows:

Table 7.1.4: Distribution of residues [1]

PHI	Total	Organosoluble	Water soluble	Not extracted	Chlorothalonil [2]
days	mg/kg	% [mg/kg]	% [mg/kg]	% [mg/kg]	% [mg/kg]
FOLIAR					
7	161-263	72-80 [117-209]	10-14 [22-34]	8-14 [14-23]	72-80 [116-206]
21	52-78	47-60 [24-47]	21-30 [15-16]	19-24 [11-15]	42-58 [22-45]
STALK					
7	1.0-4.6	29-59 [0.29-2.7]	21-36 [0.25-0.95]	21-35 [0.33-0.95]	27-55 [0.28-2.57]
21	0.7-1.4	18-46 [0.13-0.60]	30-53 [0.39-0.65]	24-29 [0.22-0.38]	10-42 [0.08-0.55]

[1] Expressed as percentages or as mg/kg chlorothalonil equivalents in whole product; results are given as a range and not as mean.

[2] Determined by HPLC in the organosoluble fraction.

Summary of Characterization and Identification of Residues in Celery Commodities Following Foliar Treatment with ^{14}C-Chlorothalonil at a Rate of 12 x 2.5 kg/ha.

Crop and Commodity		Celery Stem	
TRR mg/kg		Day 7 (1.0-4.6 mg/kg)	Day 21 (0.7-1.4 mg/kg)
Initial extraction applied to chromatography, %TRR		NP 44.9; P 26.9; PES 28.2	NP 28.8; P 44.2; PES 27.0
Origin of component	Component (code or structure)	% TRR	% TRR
Chromatography of extracted residue	Chlorothalonil	43.1	26.2
	SDS 3701	ND	ND
	Unknowns[2]	1.8	2.6
Aqueous		26.9[A]	44.2
Unextractable		28.2[B]	27.0

NP – non polar; P – polar; PES– post extracted solids

Aqueous phase in foliage was investigated by 6M HCl reflux, but radioactivity evenly spread interpretation difficult.

A – chromatographed using anion chromatography and found to be present in several fractions, no further id possible

B- 10% released using enzymes, but no further ID possible

Based on TLC and HPLC analyses, the principal component of both the foliar and stalk organo-soluble ^{14}C-residue was chlorothalonil and accounted for over 95% of the organosoluble residue in these plant parts ~~and for 72-80% of the total residue~~. Neither SDS-3701 nor SDS-46851 was detected in these analyses. Only numerous, very minor components were observed in addition to chlorothalonil, none of which could be present at greater than 0.01 mg/kg. Polar watersoluble residue levels in foliar samples were higher than in stalk samples. The stalk tissues contained less than 1 mg/kg equivalent of polar residues,

which did not provide enough material for isolation and characterisation of any single component. These polar watersoluble samples were further analysed for mono- and diglutathione conjugates of chlorothalonil. However, none of the many minor components in the aqueous soluble fractions of stalks or foliar samples could be identified. They were shown by direct analysis to contain many minor components of varying polarity. Fractionation using anion chromatography gave four fractions ranging from weakly polar to strongly acid. Analysis of each fraction by HPLC and TLC confirmed each fraction was a complex mixture of components (each <8% TRR of the stalk residue). The largest single fraction represented 0.22 mg/kg, comprised of up to 20 or more components. Therefore, isolation and identification of a single component was not possible. Further chemical and enzymatic hydrolysis of foliar samples did not release identifiable organosoluble components. For foliar samples, sequential hydrolysis of the not extracted fraction produced complex, multi-component mixtures which could not be further identified. The distribution of radiolabel in leaf laminae residues was studied in an additional experiment. The result indicated that the polar watersoluble and the not extracted residues increased slowly over the application period (with increasing number of applied doses).

Conclusions

Celery stalks and foliar samples harvested 7 days PHI contained total residue levels of 1.0- 4.6 and 161-263 mg equivalents/kg, respectively. On day 21, the total residue levels were 0.7-1.4 and 52-78 mg equivalents/kg, in stalk and foliar samples, respectively. Chlorothalonil was the only identifiable constituent from the celery stalk (0.08-2.57 mg/kg) and foliage (22-210 mg/kg) and accounted for over 95% of the organosoluble residue in these plant parts ~~and for 72-80% of the total residue~~. Up to 0.95 and 34 mg equivalents/kg in stalk and foliage, respectively, was unidentified watersoluble and not extracted residue.

Limitations

Total amounts of organosoluble, watersoluble as well as not extracted residues in foliage and stalk exceeded 0.05 mg equivalents/kg. Although over 95% of the organosoluble residue was identified to be chlorothalonil, the watersoluble residue was only poorly characterised, despite many efforts. Particularly for stalk, the watersoluble residue accounted for 21-53% of the total residue while 21-35% of the total residue remained not extracted. However, because attempts to identify the watersoluble and not extracted residue indicated the presence of many minor components which could not be further identified despite many efforts, the study is considered acceptable for the overall evaluation.

B.7.2.1.5 Plant metabolism in snap beans

Previous evaluation	In DAR
RMS remark	~~Acceptable~~ Not conclusive; It was noted during the expert Peer Review Meeting (# 164), that for some of the metabolism studies available for the evaluation, the proportions for the identified metabolites were (probably) calculated based on the total residues in the extracted fractions and not based on the total residues in extracted

	commodity. A lot of inconsistencies across of all the studies have been observed and it was agreed by the experts that the studies cannot be considered as reliable.

Characteristics

reference	:	Zeneca, Huhtanen, 1993	application rate	:	equivalent to 2.46 kg ai/ha (2.25 lb ai/acre)
type of study	:	plant metabolism	concentration	:	Bravo 720 [1]
year of execution	:	1992	frequency	:	4 applications
test substance	:	[^{14}C-U-phenyl]-chlorothalonil (radiochem. pur. 99%) and chlorothalonil (pur. 99%)	interval	:	7 days
plant	:	snapbeans	total rate	:	10 kg/ha
GLP statement	:	yes	exposure	:	spray
guideline	:	not applicable	PHI	:	7, 28 days

[1] Concentration not presented

Study design

Snapbean plants (Blue Lake variety) were treated with [^{14}C-U-phenyl]-chlorothalonil by spraying the test material prepared for application as the Bravo® 720 formulation at levels to achieve the maximum recommended rate. Four applications each at the nominal rate of 2.46 kg ai/ha (2.25 lb ai/acre) ^{14}C-chlorothalonil were made at weekly intervals using methods to simulate a normal field application. The first application was at the early bloom stage of plant development. The fourth application was made when immature beans had grown to a length of 7.5 to 8 cm. The plants were harvested at 7 and 28 days PHI by cutting the main stalk close to the ground. Beans, leaves and combined stems and stalk were separated. Beans (fresh) from three plants at each harvest were immersed in acetone and analysed to determine the surface ^{14}C-residue. The samples were refrigerated (4 ºC) for no longer than 24 hours prior to the acetone wash. All other samples were stored frozen below -5 ºC prior to analysis. The procedures and analytical methods used in this study were identical to those described in study 1 (Zeneca, Nelsen, 1985) and study 4 (Zeneca, Huhtanen, 1992). Considerable emphasis was placed on the characterization and identification of polar watersoluble and not extracted residues. Methodologies employed included: Extraction, partitioning, TLC, HPLC analysis and HPLC separation/collection of eluent of anion chromatography, acid, base and enzyme hydrolysis and derivatisation/TLC.

Samples were extracted three times with 5 mL/g acetone/1M HCl (4:1), or 5 mL/g acetone/ 0.3M phosphate buffer pH 6-7 (4:1). Material was centrifuged and the phases separated. The acetone was then removed and partitioned with ether. The ether phase was defined as non-polar and residues remaining in aqueous phase defined as polar, solids defined as PES.

Results

The results obtained in this study were as follows:

Table 7.1.5: Distribution of ^{14}C-residues [1]

PHI	Total	Acetone wash	Organosoluble [2]	Watersoluble [2]	Not extracted [2]	Chlorothalonil [3]
days	mg/kg	% (mg/kg)	% (mg/kg)	% (mg/kg)	% (mg/kg)	% (mg/kg)
BEANS						
7	0.90-1.2	26 (0.24)	28-35 (0.25-0.38)	50-54 (0.46-0.59)	14-19 (0.12-0.23)	20-31 (0.18-0.30)
28	1.0-3.1	8.0 (0.09)	14-19 (0.15-0.60)	57-60 (0.58-1.8)	23-28 (0.26-0.75)	3.3-14 (0.03-0.43)
FOLIAR						
7	110-220	na [4]	77-85 (82-190)	13-18 (20-28)	1.9-4.2 (4-6)	77-80 (82-170)
28	31-160	na	33-75 (10-120)	19-50 (15-31)	5.9-17 (5.2-9.4)	33-70 (10-110)

[1] Expressed as percentage relative to total residue and as mg/kg chlorothalonil equivalents in whole product as determined by combustion and LSC, unless stated otherwise.
[2] Expressed as % or in mg equivalent/kg acetone washed beans, or whole product in the case of foliar samples.
[3] Chlorothalonil was determined by HPLC in the organosoluble fraction.
[4] na = not applicable

Summary of Characterization and Identification of Residues in Snapbean Commodities Following Foliar Treatment with ^{14}C-Chlorothalonil at a rate of 4 x 2.5 kg/ha.

Crop and Commodity		Snapbean beans		Snapbean foliage	
TRR mg/kg		Day 7 (0.9-1.2)		Day 7 (106-217)	Day 28 (30.5-159)
Initial extraction applied to chromatography, %TRR		NP 27-35; P 50-54; PES 14-19		NP 77-85; P 13-18; PES 2-4	NP 33-75; P 19-50; PES 6-17
Origin of component	Component (code or structure)	mg/kg	%TRR	%TRR	
Chromatography of extracted residue	Chlorothalonil	0.26	25.3	77-80	33-70
	SDS 3701		ND	ND	ND
	Unknowns[2]	0.06	6.3		
Aqueous		0.52	51.2[A]	13-18	19-50
Unextractable		0.18	17.2[B]	2-4	6-17

A – chromatographed using anion chromatography and found to be present in several (50) fractions, no further id possible, albeit there was HPLC and TLC conducted on fractions and containing numerous components. Non-greater than 0.02 mg/kg.
B- 8% released using enzymes but no further ID possible
The initial extraction applied to chromatography for day 28 beans was 1.02-3.10 %TRR with NP 14-19; P 57-60; PES 23-28

The components identified in the non polar organosoluble residue fraction were chlorothalonil, SDS-3701, and SDS-46851. Only chlorothalonil could be quantified. In total (acetone wash plus organosoluble fraction of washed beans), the level of chlorothalonil was calculated by the rapporteur to vary between 0.42-0.52 mg/kg and 0.12-0.52 mg/kg whole (unwashed) beans for 7 and 28 days PHI, respectively. The two other compounds were below the limit of determination (LOD: 0.02 mg/kg and 0.03 mg/kg for SDS-3701 and SDS-46851, respectively). Processing and analysis of the watersoluble fractions by a variety of techniques (including TLC, HPLC, hydrolysis) produced complex, multi-component mixtures. Analysis by TLC showed more than 50 minor components of varying polarity. These were separated by anion column chromatography into 5 fractions ranging from weakly polar to strong acidic. Analysis of each of these fractions by HPLC and TLC showed that each was a complex mixture of components. Following acid and base hydrolysis 3-25% of the residues were organosouble. However, TLC analysis of these components did not identify them. Each of these components was estimated to be <0.02 mg/kg (expressed as chlorothalonil equivalents). No further efforts were made to identify these residues. Similar efforts were applied to the not extracted fraction in an attempt to characterize and identify these residues. Only small amounts (i.e. 17-26% of the initial not extracted residue) could be released, but appeared not organosoluble: post-extraction solids were sequentially tretaed with cellulase and 6M HCl, which solubilised ca 45% of the PES. These soluble residues accounted for less than 10% of the TRR. After acid treatment, the remaining samples still contained >50% of the original ^{14}C-residues. Since these rigid chemical treatments produced limited water-solubilization of not extracted ^{14}C-residues which were not organosoluble, no further analyses of these samples were conducted.

The results of the present study are consistent with those reported in the ^{14}C-chlorothalonil celery metabolism study (study 4 (Zeneca, Huhtanen, 1992)). In that study, the polar ^{14}C-residues extracted from celery leaf laminae were analysed by anion chromatography which showed that both weak and strong anionic compounds were present. The small amount of ^{14}C-chlorothalonil which penetrated the leaf cuticle apparently was rapidly metabolized and/or bound. Based on these prior results with treated celery leaves, no additional analyses were performed on polar extracts of bean leaves.

Conclusions

Total residue levels in beans and leaves 7 days after treatment were 0.90-1.2 and 110-220 mg equivalents/kg, respectively. At 28 days PHI, these levels were 1.0-3.1 and 31-160 mg equivalents/kg, respectively. The major component of the organosoluble fraction in acetone washed beans was identified as the parent compound chlorothalonil, accounting for 20-31% (0.18-0.30 mg/kg) and 3.3-14% (0.03-0.43 mg/kg) of the total residue in washed beans, at 7 and 28 days PHI, respectively. In total (acetone wash plus organosoluble fraction of washed beans), the level of chlorothalonil was calculated by the rapporteur to vary between 0.42-0.52 mg/kg and 0.12-0.52 mg/kg whole (unwashed) beans for 7 and 28 days PHI, respectively. In the foliar samples, these values were 77-80% (82-170 mg/kg) and 33-70 (10-110 mg/kg), at 7 and 28 days PHI, respectively. Watersoluble and not extracted residue components in washed beans accounted for up to 50-54% and 14-19% at 7 days PHI, respectively, corresponding to 0.46-0.59 and 0.12-0.23 mg equivalents/ kg, respectively. At 28 days PHI, these values were 57-60 and 23-28%, and

0.58-1.8 and 0.26-0.75 mg/kg, respectively. Despite some efforts made, these residues could no further be identified. Minor identified components were SDS-3701 and SDS-46851. Both were below the LOD.

Limitations

Total amounts of unidentified watersoluble residues significantly exceeded 0.05 mg equivalents/kg and accounted for up to 60% of the total residue in/on beans. These residues were not further identified. However, attempts to identify these watersoluble residue components indicated multiple components. In addition, 28% or less of the total residue remained not extracted. However, since analyses indicated multiple components for this fraction of the residue as well, the study is considered acceptable for the overall evaluation.

B.7.2.1.6 Plant metabolism in tomato

Previous evaluation	In DAR The paragraph 'limitations' is amended, since the intended GAP for the renewal is different from the GAP during the original approval of chlorothalonil.
RMS remark	Supportive study

Characteristics

reference	:	Vischim, Mayo 1996a	application rate	:	1.6 kg ai/ha
type of study	:	uptake, distribution and metabolism	concentration	:	733 mg/180 ml, equiv. to 0.41 kg/hl (WG formulation, 0.275% a.i.)
year of execution	:	1994-1995	frequency	:	1
test substance	:	chlorothalonil batch no. 14/09/93/1, chem. pur. 99.5%; [ring-^{14}C]-labelled chlorothalonil batch no. CFQ7386, chem pur. unknown, radiochem. pur. >98%)	interval	:	not applicable
plant	:	tomato	total rate	:	ditto application rate
GLP statement	:	yes	exposure	:	foliar and whole plant
guideline	:	no guideline in force	PHI [1]	:	DAT 2h, 2 w, 3 w, 4 w

[1] PHI is not applicable. Instead, DAT (days after treatment) are reported.

Study design

Tomato plants (variety Moneymaker), grown to maturity (fruits at various stages of ripening) in pots in a polyethylene tunnel located outside (Cambridgeshire, UK) for two months, were treated with a single application of [ring-^{14}C]-chlorothalonil (spec. activity 106 kBq/mg; 2.87 µCi/mg) by spraying at a rate of 1.6 kg ai/ha. Three and six plants were used for foliar application (fruits covered by plastic bags and removed 1-2 h after spraying) and whole plant application, respectively. Samples of fruit and leaves were taken at 2 h and at two, three and four (final harvest) weeks. Total ^{14}C-residues were determined by combustion and liquid scintillation counting (LSC). TLC and HPLC were used for the quantification and identification of residue components.

Results

Although no metabolites were identified in this study, the overall results with respect to characterization

were found to be comparable to those of the tomato study (Study 2) submitted by Zeneca (Nelsen, 1988) and therefore not summarised in detail here.

Between 1.2% and 7.6% of the radioactivity was accounted for in the solvent extracts and up to 4.1% remained unextracted. The major component of the radioactive residue in tomato fruit was chlorothalonil at all samples times. Up to five unknown components were detected, none of which represented greater than 2.8% of the residue. Polar components, eluted at the solvent front, accounted for 3.4% - 4.6% of the total residue. The level of unknown was increased from 3.0% to 5.3% (0.10 µg equiv./g) but no chlorothalonil was detected in the hydrolysate. The major component of the radioactive residue in tomato leaves was chlorothalonil at all samples times. This represented to 93.2% (134.7 ug equiv./g) of the total radioactive residue after two hours and this decreased to 93.2% (59.9 ug equiv./g) after four weeks. Up to three other unknown components were detected, none of which represented greater than 1.7% of the residue.

Mean recovery in tomato and leaf samples

Sample	Mean % radioactivity in surface washes	Mean % radioactivity in solvent extracts	Mean % unextracted radioactivity	TRR (µg equiv./g)
Tomato				
2 hours (zerotime)	98.8	1.2	<0.1	2.18
2 weeks	93.7	4.9	1.4	3.80
3 weeks	92.5	4.9	2.5	2.95
Harvest (4 weeks)	90.4	7.6	2.0	1.83
Leaf				
2 hours (zerotime)	98.1	1.3	0.6	139.0
2 weeks	94.9	2.3	2.9	55.1
3 weeks	94.8	2.7	2.7	67.5
Harvest (4 weeks)	93.1	2.7	4.1	64.3

Major component of the radioactive residue in tomato fruit at different time

Component	Two hours after application		Two weeks after application		Three weeks after application		Harvest, four weeks after application	
	%	µg equiv./g[1]	%	µg equiv./g[1]	%	µg equiv./g[1]	%	µg equiv./g[1]
Parent	94.0	2.05	91.5	3.48	89.4	2.63	88.4	1.62
Unknowns	0.3	<0.01	3.5	0.13	4.1	0.12	3.7	0.07
Polar	4.6	0.10	3.6	0.14	3.4	0.10	4.0	0.07
Other	1.2	0.03	-	-	0.6	0.02	1.9	0.03
Unextracted	<0.01	0.01	1.4	0.05	2.5	0.07	2.0	0.04
Total	100.1	2.18	100	3.80	100	2.95	100	1.83

[1]: Calculated as a percentage of the total concentration (µg equiv./g)

Conclusions

Total residue levels in/on fruit declined from 3.8 to 1.8 mg eq/kg (from 2 to 4 w after treatment). More than 90% of the total residue in/on the fruit and the leaves was present in the surface washes at all sampling times, and more than 88% of the surface washes consisted of unchanged chlorothalonil at all sampling times, including harvest. Less than 10% of the total residue remained in/on the fruit after surface washing.

Limitations

It is to be noted that in the present study 1 application of 1.6 kg ai/ha was used and analyses were performed between 14 and 28 days later, while it is intended to apply the a.i. with a PHI of 3 days. As chlorothalonil has hardly been metabolised after 28 days, the deviation in PHI is not of relevance. ~~As such, it is questioned whether the results sufficiently reflect the nature and levels of residues to be expected under field conditions.~~ As the general results are in line with those of the tomato study submitted by Zeneca (Nelsen, 1988), the present results are considered supportive for the findings in other studies.

B.7.2.1.7 Plant metabolism in wheat

Previous evaluation	In DAR The paragraph 'limitations' is amended, since the intended GAP for the renewal is different from the GAP during the original approval of chlorothalonil.
RMS remark	~~Based on~~ ~~Although the intended GAP for the renewal is more critical, the study can be considered sufficiently acceptable~~ Not acceptable; It was noted during the expert Peer Review Meeting (# 164), that for some of the metabolism studies available for the evaluation, the proportions for the identified metabolites were (probably) calculated based on the total residues in the extracted fractions and not based on the total residues in extracted commodity. A lot of inconsistencies across of all the studies have been observed and it was agreed by the experts that the studies cannot be considered as reliable. Moreover, the study is underdosed (0.6N) compared to proposed cGAP for cereals (wheat, barley) as representative crops.

Characteristics

reference	:	Vischim, Mayo 1996b	application rate	:	1.0 kg ai/ha
type of study	:	uptake, distribution, and metabolism	concentration	:	217.5 mg/80 ml, equivalent to 0.27 kg/hl (WG formulation)
year of execution	:	1994	frequency	:	1
test substance	:	chlorothalonil batch no. 14/09/93/1, chem. pur. 99.5%; [ring-^{14}C]-chlorothalonil batch no. CFQ7386, chem pur. unknown, radiochem. pur. >96%)	interval	:	not applicable
plant	:	wheat	total rate	:	idem application rate
GLP statement	:	yes	exposure	:	foliar
guideline	:	no guideline in force	PHI [1]	:	2 h after treatment (6 May 1994), 4 w after treatment, 4 w prior to harvest and at maturity (3 August 1994)

[1] PHI is not applicable.

Study design

Winter wheat (variety Riband), grown in containers outside (Cambridgeshire, UK), was treated with a single spray application of [ring-^{14}C]-chlorothalonil and unlabelled chlorothalonil (final spec act. 673 kBq/mg) at a rate of 1.0 kg ai/ha. Samples of immature wheat were taken 2 h after dose application and after 4 weeks. A further sample of wheat was taken 4 weeks prior to harvest with the remaining mature wheat taken at harvest (there is almost 13 weeks time between application and final harvest). Wheat samples were separated into grain and straw, and stored frozen until analysis. Total ^{14}C-residues were determined by combustion and liquid scintillation counting. TLC and HPLC were used for the quantification of residue components. Radiolabelled components were identified by co-chromatography of standard reference compounds, and/or by LC-MS and LC-MS/MS.

Results

The mean total level of radioactive residues (TRR) amounted to 0.10 mg eq/kg in immature heads at 4 weeks after treatment, to 0.05 mg eq /kg at 1 month before harvest, and decreased to <0.01 mg eq/kg in mature grain at harvest. Characterisation and identification of residues in mature grain were not performed because of the low total residue levels.

In forage, mean TRR levels were 51 mg eq/kg at 2 h after application. In wheat straw, mean TRR levels were 6.8 mg eq/kg at 4 weeks after application, 1.7 mg eq/kg at 1 month before maturity (harvest) and 8.4 mg eq/kg at maturity (harvest). According to the authors, the observed decrease and increase in TRR was due to growth and loss of moisture on ripening, respectively. The proportion of extractable residue decreased from 93% in forage, 2 h after application, to 48% at maturity in wheat straw. Chlorothalonil represented 89% TRR (45 mg/kg) in immature wheat at 2 h after application, and 2.1% TRR (<0.1 mg/kg) at 1 month before maturity.

In mature straw, 4.4% TRR (0.35 mg eq/kg) of the total residue was hexane soluble, 44% TRR (3.5 mg eq/kg) was soluble in acetonitrile/water, and 52% TRR (4.1 mg eq/kg) was unextracted. Following

hydrolysis of the unextracted residue with base, and partitioning with organic solvents, a further 40% TRR (3.1 mg eq/kg) was extracted and 8.7% TRR (0.70 mg eq/kg) remained unextracted. In the hexane soluble and acetonitrile/water fractions, a total of 9 components were found representing 32% TRR, in addition to polar material (13% TRR) and other material (3.3% TRR). Identification of the components showed that chlorothalonil and the metabolite 4-hydroxy-2,5,6,-trichloro-1,3-dicyanobenzene (in the Zeneca studies coded as SDS-3701) were minor components at levels of 0.02 mg/kg (0.2% TRR), and 0.04 mg/kg (0.5% TRR), respectively. The major component was a di- and/or triglutathione conjugate of chlorothalonil at 0.73 mg/kg (9.3%TRR). Several other unknown components were present at levels below 7% TRR. The polar material was reduced to 4.4% TRR following treatment with β-glucosidase with a corresponding increase in glutathione conjugates (20% TRR). The extracts following base treatment of the unextracted material consisted of a total of 10 polar components, none of which represented more than 8.1% TRR.

Conclusions

Following application of chlorothalonil, significant levels of residue (0.1 mg eq/kg) are found in immature heads at 4 w after treatment. Residue levels were <0.01 mg/kg in mature grain at the time of harvest. No data are available on the nature of the residue in grain.

In total immature plants, chlorothalonil was the main residue component from 2 h after application to 4 w after application at 89% and 52% of the TRR, respectively. At 1 month before harvest, chlorothalonil levels had decreased significantly (2.1% TRR, <0.1 mg/kg), while levels of other (unknown) components, and unextracted radioactivity had increased to in total 37% and 47% TRR, respectively.

Mature straw contained residue levels up to 7.9 mg eq/kg. Chlorothalonil and 4-hydroxy-2,5,6,-trichloro-1,3-dicyanobenzene (SDS-3701) appeared to be minor components at levels of 0.02 mg/kg (0.2% TRR) and 0.04 mg/kg (0.5% TRR), respectively. The major component appeared to be a (di- and/or tri)glutathione conjugate of chlorothalonil at levels of 0.73 mg/kg (9.3% TRR). In addition, a large number of other components are present, none of which represents more than 8.1% TRR.

Limitations

The chemical purity of radiolabelled chlorothalonil was not indicated. It is noted that the grain metabolism study has been performed with less than the maximum total intended rate, i.e. one application of 1.0 kg ai/ha instead of 2 applications of 0.75 kg ai/ha. Furthermore, it is not clear whether the substance was applied at the intended growth stage for application. It is only known from the study report that there is almost 13 weeks time between application and final harvest.

B.7.2.1.8 Plant metabolism in peas

Previous evaluation	In DAR The paragraph 'limitations' is amended, since the intended GAP for the renewal is different from the GAP during the original approval of chlorothalonil.
RMS remark	~~Acceptable~~

	Not conclusive; It was noted during the expert Peer Review Meeting (# 164), that for some of the metabolism studies available for the evaluation, the proportions for the identified metabolites were (probably) calculated based on the total residues in the extracted fractions and not based on the total residues in extracted commodity. A lot of inconsistencies across of all the studies have been observed and it was agreed by the experts that the studies cannot be considered as reliable.

Characteristics

reference	:	Vischim, McEwen 1997	application rate	:	1.4 kg ai/ha
type of study	:	uptake, distribution, and metabolism	concentration	:	262 mg/150 ml, equivalent to 0.174 kg/hl (WG formulation, 0.17% a.i.)
year of execution	:	1995	frequency	:	1
test substance	:	chlorothalonil batch nos. 14/09/93/1 & 9/5/94/7, chem. pur. ≥99.2%; [ring-^{14}C]-chlorothalonil batch no. CFQ7386, chem pur. unknown, radiochem. pur. >96%)	interval	:	not applicable
plant	:	peas	total rate	:	idem application rate
GLP statement	:	yes	exposure	:	foliar and whole plant application
guideline	:	no guideline in force	PHI [1]	:	DAT 1 h, 7, 14, 30, 41 d

[1] PHI is not applicable. Instead, DAT (days after treatment) are reported.

Study design

Pea plants (variety Scout) grown in pots outside (Cambridgeshire, UK) and estimated to be at a growth stage (developing pods) two weeks before the green harvest, were treated with a single spray application of [ring-^{14}C]-chlorothalonil (final spec act. 540 kBq/mg; 14.6 µCi/mg) at a rate of 1.4 kg ai/ha. Foliage as well as whole plant application was performed. Pea vines, pods and seeds (peas) were harvested one hour, 7, 14 (mature green crop), 30 and 41 (mature dry crop) days after dose application. Immediately after harvesting, samples of pods and vines were surface washed with acetonitrile. The peas were removed from the pods, and stored frozen or were directly extracted with acetonitrile/water mixtures. Total ^{14}C-residues were determined by combustion LSC. TLC and HPLC were used for the quantification of residue components. Radiolabelled components were identified by co-chromatography of standard reference compounds and/or by LC-MS.

Results

Although the experimental setup (number of applications; dose rate) was different in this study, the overall results with respect to characterization were found to be comparable to those of the snap bean study (Study 5) as submitted by Zeneca (Huhtanen, 1993) and therefore not summarised in detail here. Similar results were found with respect to the major identified component (chlorothalonil) and some metabolites (4-hydroxy-2,5,6,-trichloro-1,3-dicyanobenzene or SDS-3701 and a diglutathione conjugate). In addition,

these metabolites, that were found at levels below the limit of quantification (< LOD) in the Zeneca study on snapbeans, were found in this study in quantifiable amounts (> LOD). 4-Hydroxy-2,5,6,-trichloro-1,3-dicyanobenzene (SDS-3701) and a diglutathione conjugate accounted for 1.5 and 0.9 mg eq/kg, respectively, in pods and for 3.1 and 2.0 mg eq/kg, respectively in vines. However, in this Vischim study by McEwen, no 3-carboxy-2,5,6-trichlorobenzamide (coded SDS-46851 in Zeneca, Huhtanen, 1993) was reported.

Conclusions

Upon whole plant spraying, total residues at 14 DAT were up to 0.04 mg eq/kg in peas, ca. 10 mg eq/kg in pods, and 56 mg eq/kg in vines. Total residues at maturity (41 DAT) were up to 0.07 mg eq/kg in peas, ca. 28 mg eq/kg in pods, and 71 mg eq/kg in vines. Peas contained 0.003 mg/kg of chlorothalonil, and several other minor components at levels <0.01 mg/kg. About 80% of the TRR on mature vines and pods was removed by organic surface washes and chlorothalonil accounted for 75% (21 mg eq/kg) to 78% (55 mg eq/kg) of the TRR in these surface washes, respectively.
Furthermore, 4-hydroxy-2,5,6,-trichloro-1,3-dicyanobenzene (SDS-3701) and a diglutathione conjugate accounted for 1.5 and 0.9 mg eq/kg, respectively, in pods and for 3.1 and 2.0 mg eq/kg, respectively in vines.

Data for foliar application indicate that systemic transport to the pods is very limited (<10% of the levels observed upon whole plant application) whereas transport to peas is significant (comparable levels to whole plant application).

Limitations

Some discrepancies were noted in the various parts of the study report with respect to batch numbers. Also, the chemical purity of radiolabelled chlorothalonil was not indicated.
Results are considered a first indication with respect to the expression of residues upon intended use application. As the general results are in line with those of other metabolism studies in primary crops, the present results are considered supportive for the findings in other studies.

B.7.2.2 Poultry

During the initial peer review, the metabolism of chlorothalonil and SDS-3701 has been studied in laying hens using ^{14}C-chlorothalonil and ^{14}C-SDS-3701 labelled uniformly in the phenyl ring. In addition, a new metabolism study has been conducted with hens for the sake of the renewal of chlorothalonil.

B.7.2.2.1 Metabolism of chlorothalonil in hens

Previous evaluation	In DAR
RMS remark	Acceptable, however, further characterization of the TRR is recommended

Characteristics

reference	:	Zeneca, Capps, 1983	exposure	:	21 daily doses
type of study	:	distribution, metabolism and excretion	doses	:	0, 2, 6, and 20 mg/kg food
year of execution	:	1982	vehicle	:	none (directly in capsule)
test substance	:	[^{14}C]-chlorothalonil (radiochem. pur. >98 %) and chlorothalonil (pur. 96.0%)	GLP statement	:	no
route	:	oral	guideline	:	not applicable
species	:	laying hen			
group size	:	10/dose			

[1] equal to 0, 0.22, 0.65, and 2.18 mg/kg bw/day

Study design

Groups of 10 laying hens were administered 21 daily doses of [^{14}C]-chlorothalonil in capsules at dose levels of 0.22, 0.65, and 2.18 mg/kg bw/day bw/day ~~(estimated dose rates 36 N, 107 N and 357 N, respectively)~~. Control animals received placebo capsules at 21 successive days. The eggs from each test group laid on all test days were collected and separated into whites and yolk, pooled according to dose level and test day and frozen until analysis. Following the end of the dosing regimen (day 21), the hens were sacrificed at scheduled intervals and combined samples of muscle (adductor, cardiac, and pectoral), liver, skin, and fat were collected and quick-frozen for subsequent residue analysis. The scheduled intervals for sacrifice were as follows: within 6 h after the final dose (4 hens/group), 3 days after final dose (3 hens/group), 7 days after final dose (the remaining 3 hens/group). Samples of egg yolks, egg whites and tissues were homogenized, combusted and analysed by LSC.

Results

The LOQ in egg white, egg yolk and tissues varied between 0.0227 and 0.0476 mg/kg. The radiolabel residues were below the LOD at all dose levels for egg white and at dose levels of 0.22 and 0.65 mg/kg bw/day for egg yolk. Total residue levels in egg yolk were 0.035-0.047 mg/kg in the high dose group during treatment days 13-17 and below the LOD on the other days. Therefore, a plateau cannot be clearly defined for egg yolk. It can only be concluded that there were quantifiable residues at days 13-17 in the high dose group. Total residue levels in tissue were all below the LOD except in liver of the middle and high dose hens. Maximal total residue levels in livers were determined at 6 h after the last dose and were

0.098 and 0.050 mg/kg for the 0.65 and 2.18 mg/kg bw/day dosed hens, respectively. The levels were below the LOD again on day 24. The fact that in the 0.65 mg/kg bw/day dose group, liver residue level was higher than in the 2.18 mg/kg bw/day group, was hypothesized by the authors to be due to mis-labelling or switching at the time of sacrifice.

Conclusion

The results of this study demonstrate that in general only very low residue levels occur in eggs and tissues from hens dosed with chlorothalonil at dose levels up to 0.65 mg/kg bw/day for 21 days. However, in hens dosed 2.18 mg/kg bw/day, total residue in egg yolk levelled up to 0.05 mg/kg. In hens dosed 0.65 and 2.18 mg/kg bw/day, total residue in liver levelled up to 0.1 mg/kg.

Limitations

It should be noted that no attempts were made to characterize or identify the residue components, although total residue levels in egg yolk and liver exceeded 0.01 mg/kg in hens dosed 0.65 and 2.18 mg/kg bw/day.

B.7.2.2.2 Metabolism of SDS-3701 in hens

Previous evaluation	In DAR
RMS remark	Acceptable, however, further characterization of the TRR is recommended

Characteristics

reference	:	Zeneca, Capps, 1984	exposure	:	21 daily doses
type of study	:	distribution, metabolism and excretion	doses	:	0, 0.1, 0.3, and 1.0 mg/kg food [1]
year of execution	:	1982	vehicle	:	none, directly in capsule
test substance	:	[^{14}C]-SDS-3701 (radiochem. pur. >98 %) and SDS-3701 (pur. 99%)	GLP statement	:	no
route	:	oral	guideline	:	not applicable
species	:	laying hen			
group size	:	10/dose)			

[1] equal to 0, 0.011, 0.033, and 0.11 mg/kg bw/day

Study design

To determine whether SDS-3701 (i.e. 4-hydroxy-2,5,6-trichloroisophthalonitrile; ^{14}C-R182281,specific activity 619 Bq/µg for day 1 - 14 doses; 613 Bq/µg for day 15 - 21 doses; radiochemical purity >98%), a major metabolite of chlorothalonil, will yield detectable residues in eggs or tissue of laying hens (White Leghorn), groups of 10 laying hens where dosed orally by gelatin capsule at dose levels of 0.011, 0.033, and 0.11 mg/kg bw/day on 21 consecutive days (or 0.1, 0.3 and 1.0 mg/kg in the diet). A further group of hens were used as a control. The eggs from each test group laid on all test days were collected. The egg contents (white and yolk) were separated and pooled according to dose level and test day and quick-frozen. Following the end of the dosing regimen (day 21), the hens were sacrificed at scheduled intervals

and composite samples of muscle (adductor, cardiac, and pectoral), liver, skin, and fat were collected and quick-frozen for subsequent residue analysis by scintillation counting. The scheduled intervals for sacrifice were as follows: within 6 h after the final dose (4 hens/group), 3 days after final dose (3 hens/group), 7 days after final dose (the remaining 3 hens/group).

Samples of egg yolks, whites and tissues were homogenised, combusted and analysed by LSC. Egg yolks from day 20 of the high dose group were extracted with acetonitrile/water (3:1) and partitioned with hexane to remove oils and fats. After removal of the acetonitrile, the residual aqueous fraction was partitioned with dichloromethane. The dichloromethane fraction was then analysed by reverse phase HPLC.

Results

The distribution of ^{14}C-residues was as follows:

Table 7.2.4: Distribution of radiolabel in eggs and tissues (in mg SDS-3701 equivalents/kg)

Matrix	0.011 mg/kg bw/d	0.033 mg/kg bw/d	0.11 mg/kg bw/d
Egg white	nd	nd	nd
Egg yolks	0.03-0.040 (days 11-22)	0.05-0.12 (days 6-26)	0.06-0.42 (days 4-28)
Adductor muscle	nd	nd	nd
Pectoral muscle	nd	nd	nd
Cardiac muscle	nd	0.055 (day 21)	0.15 (days 21, 24)
Fat	nd	nd	nd
Liver	0.056 (day 21)	0.05-0.27 (days 21, 24)	0.12-0.78 (days 21, 24, 28)
Skin	nd	nd	0.37 (day 21)

[1] nd = not detectable

A plateau in egg yolk was reached in the low dose group at day 21, in the mid-dose group at day 21, and in the high dose group at day 16. The LOQ in egg white, egg yolk and tissues varied between 0.0189 and 0.0416 mg/kg.

Egg yolks from the highest dose level group collected on the 20th day of the test were subjected to further analysis. It was shown that the vast majority (84.5%) of the yolk residue partitioned into the dichloromethane fraction. Analysis of this fraction by HPLC showed that 81.5% TRR was due to unchanged R182281. The identity of R182281 was confirmed by methylation with diazomethane followed by further HPLC, GC and GC-MS analysis. In those samples with a detectable residue a broadly linear relationship is shown between the residue levels at the different dose rates.

Conclusion

The results of this study demonstrate that upon daily dosing with 0.011 mg/kg bw/day SDS-3701 for 21 days, ^{14}C-residues were close to or below the LOD in egg and tissue samples. At doses of 0.033 mg/kg bw/day, considerably higher residue levels were found in egg yolk and liver only, while at a doses of 0.11 mg/kg bw/day significant residue levels were found in egg yolk, cardiac muscle, liver, and skin. In egg yolk, plateaus of residue levels were reached after 11, 6, and 4 days in the 0.011, 0.033, and 0.11 mg/kg bw/day dose groups.

Limitations

~~In the summary of the present study as supplied by the notifier, it is stated that over 80% of the total residue in egg yolk was attributable to SDS-3701. However, no data that support this statement were incorporated in the study report. As such, it~~ It is noted that no information with respect to the nature or identity of the residue components is available while total residue levels in ~~egg yolk,~~ liver, cardiac muscle and skin considerably exceeded 0.05 mg/kg.

B.7.2.2.3 Metabolism of chlorothalonil in hens

Previous evaluation	Submitted for the purpose of renewal
RMS remark	Acceptable It must be noted that only 6 hens were used per dose, while OECD 503 requires 10 hens/dose. However, as the variation in material balance and distribution of radioactivity was low, the study is considered acceptable.

Report:	K-CA 6.2.2/01. Hardwick T (2014). [^{14}C]-Chlorothalonil - metabolism of [^{14}C]-chlorothalonil in the laying hen. Report number: 8243812. Covance Laboratories Limited, Otley Road, Harrogate, North Yorkshire, HG3 1PY, UK. Unpublished. Syngenta Task No TK0046447.Syngenta Report Number 8243812. (Syngenta File No: R044686_11082)

Guidelines

OECD Guideline for the Testing of Chemicals, 503, Metabolism in Livestock (January 2007).
EPA Residue Chemistry Test Guideline OCSPP 860.1300, Nature of the Residue in Plants, Livestock (August 1996).
Regulation (EC) No 1107/2009 of the European Parliament and of the Council of 21 October 2009 concerning the placing of plant protection products on the market.
Commission Regulation (EU) No 283/2013 of 1 March 2013 setting out the data requirements for active substances, in accordance with Regulation (EC) No 1107/2009 of the European Parliament and of the Council concerning the placing of plant protection products on the market.

GLP

The study was carried out according to the principles of Good Laboratory Practice.

EXECUTIVE SUMMARY

Six laying hens were dosed orally with [phenyl-U-^{14}C]-chlorothalonil for 14 days at a nominal rate of 15 mg/kg, based on dietary dry matter intake. The dosing occurred via gelatine capsules, which contained 2.9301 mg chlorothalonil. The actual dose rate achieved, based on measured food consumption, was approximately 24.5 mg/kg. Based on the mean body weight of the hens at the first dose administration (i.e. 1.84 kg), the dose rate has been recalculated as 1.59 mg/kg bw/d (2.9301 mg chlorothalonil / 1.84 kg). Excreta and eggs were collected daily. Eggs were separated into yolk and white. The hens were sacrificed approximately 12 hours after the administration of the final dose and tissues taken post mortem for quantification and analysis.

The radioactive residue was determined in all samples by solubilisation and LSC counting. Excreta were homogenised in water. All other samples were macerated to a homogenous consistency.

Liver, egg yolk and skin samples, which contained radioactive residues greater than 0.01 mg/kg, were subjected to further analysis to determine the metabolic profile.

Samples were extracted with organic solvents, extracted residues were further fractionated and characterised. Residues present in the principal fractions were subject to high performance liquid chromatograph and thin layer chromatography/bio-image analysis for quantification and identification/characterisation by comparison with authentic reference standards of parent chlorothalonil and its metabolites R182281, SYN546672, SYN546673, SYN546674, SYN546675, SYN546676 and SYN546677.

The mean radioactive balance for all hens was greater than 93% with the majority of the radioactivity (91%) accountable in the excreta.

Residues, determined by solubilisation, were found in liver (0.139 mg/kg), skin plus subcutaneous fat (0.100 mg/kg), perirenal fat (0.035 mg/kg) and peritoneal fat (<0.01 mg/kg). Residues in muscle and egg white were below the LOQ (<0.010 mg/kg). In egg yolk, mean residues reached a maximum of 0.087 mg/kg after 13 days dosing. Sub samples of liver, skin and egg yolk were analysed further to determine the nature of the residue. Following extraction with organic solvents 32.3 to 58.3% of the TRR was solubilised.

Chlorothalonil was not detected in any of the samples. The phenolic metabolite, R182281, was the only identified residue and was found at levels of up to 35.9% TRR and 0.050 mg/kg (see table below).

B.7.2.2.3-1: Levels of R182281 in the different animal samples

Identified Components	Egg yolk		Whole egg		Liver		Skin	
	%TRR	mg/kg	%TRR	mg/kg	%TRR	mg/kg	%TRR	mg/kg
R182281	12.5	0.011	13.0	0.003	35.9	0.050	3.2	0.004

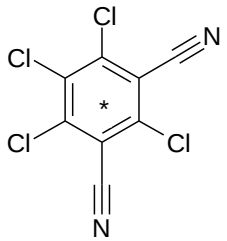
In excreta, 91% of the administered dose was recovered. The major identified component was chlorothalonil (43.2% TRR) with R182281 (2.3% TRR) the only other identified metabolite.

Unextractable residues were characterised following extraction with aqueous solvents, treatment with SDS with subsequent protein precipitation, acidic and basic hydrolysis and protease digestion. Where residue and radioactive levels were sufficient, these matrices were subjected to HPLC and TLC to

determine the nature of the residues. In the unextractable residues only R182281 was tentatively identified in eggs; in all other samples, HPLC and TLC analysis were inconclusive.
During TLC analysis, a proportion of radioactivity did not elute from the origin. This region accounted for 23.8% TRR (0.022 mg/kg) in egg yolk, 10.1% TRR (0.014 mg/kg) in liver and 4.2% TRR (0.004 mg/kg) in skin plus subcutaneous fat.
In order to demonstrate the storage stability in liver and egg yolk, chromatographic profiles obtained initially (within 6 months of sampling) were compared with profiles of the same extracts obtained on completion of the analysis.
Following 14 consecutive daily doses of [phenyl-U-^{14}C]-chlorothalonil to laying hens at a nominal rate of 15 mg chlorothalonil equivalents/kg dry matter in the feed it was concluded that:

- [^{14}C]-chlorothalonil and/or its biotransformation products are readily excreted, as ≥91% of the dosed radioactivity was accounted for in the excreta and cagewash.
- Radioactive residues reached a plateau total in eggs after 10 days.
- Radioactive residues were ≤0.207 mg/kg in eggs and tissues.
- R182281 was the only residue identified in liver, eggs and skin plus subcutaneous fat.
- In excreta, chlorothalonil was the major extractable residue with R182281 being the only other residue identified.

A1. Test Materials

Structure/Label:	[Phenyl-U-^{14}C]-Chlorothalonil
Common name	Chlorothalonil
Syngenta code	R044686
CAS Number:	1897-45-6
Batch Number:	ILA-302.1B
Specific Activity:	57.1 µCi/mg (2.113 MBq/mg)
Radiochemical Purity:	97.8%
Structure: (* marks position of radiolabel)	

A2. Test Animals

Species	Hen
Gender	Female
Weight at first dosing	1.8 to 2.0 kg
Number of animals	6
Acclimatisation Period	14 days
Diet	Measured ration of commercially available non-medicated ground concentrate and offered grit *ad libitum*
Water	Tap water, *ad libitum*
Housing	Individual metabolism cages

Environmental Conditions:	
Temperature	14-22ºC
Humidity	23-98%
Photoperiod	Alternating 16-hour light and 8 h dark cycles.

B. STUDY DESIGN AND METHODS

B1. Dosing Regime

Nominal Dose Rate:	15.0 mg/kg (dry weight)
Actual Dose Rates	18.7-29.7 mg/kg (dry weight)
Mean Food consumption on Days 1 to 14 of dosing period (kg feed as received/day):	0.118-0.161 kg
Vehicle:	Gelatin capsule
Timing:	Once daily
Duration:	14 days
Interval from last dose to sacrifice:	11-12 hours after final dose

B2. Sample Collection

Egg collection:	Twice daily, separated into yolk and white
Excreta collection:	Once daily
Samples taken post mortem:	Liver, fat, skin (including subcutaneous fat), muscle, blood; GI tract, bile, carcass were stored but not analysed

B3. Extraction and Fractionation of Residues

Tissue samples were homogenised using standard food preparation units whilst frozen on dry ice. Radioactivity in samples was quantified by combustion and subsequent LSC analysis.
Sub-samples of liver were sequentially extracted with acetonitrile: water (4:1 v/v) followed by 2% w/v sodium dodecyl sulphate solution and acetonitrile water (4:1 v/v). The extraction solids were subjected to acid hydrolysis in 1M HCl followed by base hydrolysis in 1M ammonia solution. The residue following base hydrolysis was subjected to protease digestion hydrolysis by incubation for ca 18 hours at ca 37°C.

Samples of egg yolk, from Day 14, were pooled from all animals and sequentially extracted with hexane, ethyl acetate, acetonitrile, 1% formic acid in acetonitrile, water, 1M HCl and 1M ammonia. The ethyl acetate, acetonitrile and 1% formic acid in acetonitrile extracts were pooled, concentrated and partitioned against hexane. The hexane was then partitioned against 1% formic acid in acetonitrile and acetonitrile:water (9:1 v:v). The residue from the organic and aqueous extractions was sequentially extracted with aliquots of 2% w/v sodium dodecyl sulphate solution. The extraction solids were subjected to base hydrolysis in 0.1M ammonia solution followed by acid hydrolysis in 0.1M HCl. The residue following acid hydrolysis was subjected to protease digestion hydrolysis by incubation for *ca* 18 hours at *ca* 37°C.
Composite fat with skin samples were initially homogenised in hexane then sequentially extracted with ethyl acetate, acetonitrile, 1% formic acid in acetonitrile, water, 1M HCl and 1M ammonia. The ethyl acetate, acetonitrile and 1% formic acid in acetonitrile extracts were pooled as were the water, 1M HCl and 1M ammonia extracts. The residue from the organic and aqueous extract was sequentially extracted

with aliquots of 2% w/v sodium dodecyl sulphate solution. The extraction solids were subjected to base hydrolysis in 0.1M ammonia solution followed by acid hydrolysis in 0.1M HCl. The residue following acid hydrolysis was subjected to protease digestion hydrolysis by incubation for *ca* 18 hours at *ca* 37°C. Portions of the sample extracts were analysed by HPLC and TLC to determine the metabolite profile.

II. RESULTS AND DISCUSSION

Total Radioactive Residues and Extractability

The distribution of radioactivity found in hens treated with [phenyl-U-^{14}C]-chlorothalonil is presented in Table 7.2.2.3-2. The mean radioactive balance for all hens was greater than 93% with the majority of the radioactivity (91%) accountable in the excreta.

Table 7.2.2.3-2: Distribution of radioactivity and material balance from laying hens treated with [phenyl-U-^{14}C]-chlorothalonil

Sample	% of dosed Radioactivity Recovered in Sample							
	101F	102F	103F	104F	105F	106F	Mean	SD
Excreta	94.82	90.51	89.35	87.19	90.38	93.54	90.97	2.79
Cage Wash	2.106	0.941	0.954	1.606	0.763	1.513	1.314	0.515
Egg White	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	NA
Egg Yolk	0.025	0.025	0.024	0.024	0.034	0.033	0.028	0.005
Liver	0.012	0.018	0.015	0.018	0.022	0.017	0.017	0.003
Breast Muscle	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	NA
Leg Muscle	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	NA
Peritoneal Fat	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001
Perirenal Fat	NS	<0.001	<0.001	NS	<0.001	NS	<0.001	<0.001
Skin	0.004	0.002	0.002	0.002	0.002	0.002	0.002	0.001
Gastrointestinal tract contents	0.617	1.193	0.707	0.143	1.053	2.009	0.954	0.635
Material Balance	97.59	92.69	91.05	88.98	92.25	97.11	93.28	3.408

Sample	% of dosed radioactivity recovered in sample [mean]
Excreta	90.97
Cage Wash	1.314
Egg White	<0.001
Egg Yolk	0.028
Liver	0.017
Breast Muscle	<0.001
Leg Muscle	<0.001

Peritoneal Fat	<0.001
Perirenal Fat	<0.001
Skin	0.002
Gastrointestinal tract contents	0.954
Material Balance	93.28

The mean total radioactive residues (TRRs) in egg white and yolk samples are presented in Table 7.2.2.3-3 along with calculated TRR values for whole egg. Residues in eggs white were below the LOQ (0.004 mg/kg) in all samples. Residues in egg yolk reached a plateau concentration after 10 days dosing. In whole eggs, a plateau concentration of approximately 0.024 mg/kg was reached 10 days after the start of dosing.

Table 7.2.2.3-3 a: TRR in eggs from laying hens treated with [phenyl-U- ^{14}C]-chlorothalonil

Sampling time	Mean total radioactive residue, TRR (mg/kg)		
	Egg white	Egg yolk	Whole egg[1]
Day 1	< 0.004	<0.004	<0.004
Day 2	< 0.004	0.007	0.002
Day 3	< 0.004	0.014	0.004
Day 4	< 0.004	0.029	0.008
Day 5	< 0.004	0.040	0.012
Day 6	< 0.004	0.057	0.018
Day 7	< 0.004	0.063	0.017
Day 8	< 0.004	0.074	0.015
Day 9	< 0.004	0.075	0.021
Day 10	< 0.004	0.083	0.024
Day 11	< 0.004	0.083	0.020
Day 12	< 0.004	0.086	0.026
Day 13	< 0.004	0.087	0.024
Day 14	< 0.004	0.084	0.019

[1] calculated on the basis of the weight of egg white and egg yolk.

Table 7.2.2.3-3 b: Individual results for egg white, egg yolk and whole egg, prestended in the study report:

TABLE 7 Total Radioactive Residues in Egg White from Laying Hens Dosed with [^{14}C]-Chlorothalonil

Sample Time	mg/kg							
	101F	102F	103F	104F	105F	106F	Mean	SD
Day 1 egg 1	<0.004	<0.004	<0.004	<0.004	<0.004	<0.004	<0.004	NA
Day 1 egg 2	<0.004	NA	NA	NA	NA	NA	<0.004	NA
Day 2	<0.004	<0.004	<0.004	<0.004	<0.004	<0.004	<0.004	NA
Day 3	<0.004	<0.004	<0.004	<0.004	<0.004	<0.004	<0.004	NA
Day 4	<0.004	<0.004	<0.004	<0.004	<0.004	<0.004	<0.004	NA
Day 5	<0.004	<0.004	<0.004	<0.004	<0.004	<0.004	<0.004	NA
Day 6	<0.004	<0.004	<0.004	<0.004	<0.004	<0.004	<0.004	NA
Day 7	<0.004	<0.004	<0.004	<0.004	<0.004	<0.004	<0.004	NA
Day 8 egg 1	<0.004	<0.004	<0.004	<0.004	<0.004	<0.004	<0.004	NA
Day 8 egg 2	NA	NA	<0.004	NA	NA	NA	<0.004	NA
Day 9	<0.004	<0.004	NS	<0.004	<0.004	<0.004	<0.004	NA
Day 10	<0.004	<0.004	<0.004	<0.004	<0.004	<0.004	<0.004	NA
Day 11 egg 1	<0.004	<0.004	<0.004	<0.004	<0.004	<0.004	<0.004	NA
Day 11 egg 2	NA	NA	NA	<0.004	NA	NA	<0.004	NA
Day 12	<0.004	<0.004	<0.004	<0.004	<0.004	<0.004	<0.004	NA
Day 13	<0.004	<0.004	<0.004	<0.004	<0.004	<0.004	<0.004	NA
Day 14 egg 1	<0.004	<0.004	<0.004	<0.004	<0.004	<0.004	<0.004	NA
Day 14 egg 2	NA	NA	NA	NA	NA	<0.004	<0.004	NA

NS = Not Sampled
NA – Not Applicable

TABLE 8 Total Radioactive Residues in Egg Yolk from Laying Hens Dosed with [^{14}C]-Chlorothalonil

Sample Time	mg/kg							
	101F	102F	103F	104F	105F	106F	Mean	SD
Day 1 egg 1	<0.004	<0.004	<0.004	<0.004	<0.004	<0.004	NA	NA
Day 1 egg 2	<0.004	NS	NS	NS	NS	NS	<0.004	NA
Day 2	0.011	0.010	0.008	0.004	0.007	0.004	0.007	0.003
Day 3	0.017	0.016	0.011	0.012	0.013	0.013	0.014	0.002
Day 4	0.033	0.031	0.027	0.027	0.027	0.030	0.029	0.003
Day 5	0.033	0.042	0.032	0.044	0.037	0.049	0.040	0.007
Day 6	0.050	0.063	0.052	0.050	0.057	0.069	0.057	0.008
Day 7	0.057	0.072	0.048	0.063	0.057	0.082	0.063	0.012
Day 8 egg 1	0.064	0.071	0.064	0.077	0.082	0.085	0.074	0.009
Day 8 egg 2	NS	NS	0.043	NS	NS	NS	0.043	NA
Day 9	0.065	0.076	NS	0.065	0.083	0.085	0.075	0.009
Day 10	0.072	0.076	0.083	0.088	0.091	0.090	0.083	0.008
Day 11 egg 1	0.074	0.065	0.091	0.084	0.099	0.083	0.083	0.012
Day 11 egg 2	NS	NS	NS	0.086	NS	NS	0.086	NA
Day 12	0.077	0.074	0.093	0.081	0.101	0.091	0.086	0.011
Day 13	0.078	0.065	0.099	0.082	0.100	0.100	0.087	0.015
Day 14 egg 1	0.073	0.069	0.089	0.079	0.101	0.093	0.084	0.012
Day 14 egg 2	NS	NS	NS	NS	NS	0.082	0.082	NA

NS = Not Sampled
NA – Not Applicable

TABLE 9 Mean Weight and Concentration of Radioactive Residues in Eggs from Laying Hens Dosed with [^{14}C]-Chlorothalonil

Study Day	Egg White		Egg Yolk		Whole Egg [1]	
	Sample Weight (g)	TRR (mg/kg)	Sample Weight (g)	TRR (mg/kg)	Sample Weight (g)	TRR (mg/kg)
Day 1	40.235	<0.004	15.770	<0.004	56.005	<0.004
Day 2	34.637	<0.004	12.891	0.007	47.528	0.002
Day 3	32.202	<0.004	14.869	0.014	47.071	0.004
Day 4	37.666	<0.004	14.859	0.029	52.525	0.008
Day 5	34.742	<0.004	16.057	0.040	50.798	0.012
Day 6	36.215	<0.004	13.257	0.057	49.472	0.018
Day 7	33.763	<0.004	14.738	0.063	48.501	0.017
Day 8	38.948	<0.004	16.982	0.058	55.930	0.015
Day 9	35.443	<0.004	14.419	0.075	49.862	0.021
Day 10	37.260	<0.004	13.494	0.083	50.754	0.024
Day 11	41.708	<0.004	15.664	0.084	57.372	0.020
Day 12	37.216	<0.004	13.718	0.086	50.935	0.026
Day 13	36.999	<0.004	13.484	0.087	50.482	0.024
Day 14	43.367	<0.004	16.802	0.083	60.169	0.019

TRR were 0.139 mg/kg in liver, 0.100 mg/kg in skin, 0.035 mg/kg in perirenal fat and 0.003 mg/kg in peritoneal fat. Residues in breast and leg muscle were below the LOQ. Concentrations of radioactivity in blood and plasma were 0.146 and 0.145 mg/kg, respectively. A summary of the total radioactive residues and extractability of residues is presented in Table 7.2.2.3-4.

Table 7.2.2.3-4a: Summary of total radioactive residues and extractability in tissue and egg samples from laying hens treated with ^{14}C-chlorothalonil

Sample	TRR (mg/kg) by Direct Quantification of Radioactivity (Solubilization Analysis)							TRR (mg/kg) by Summation of Extractable and Unextractable Radioactivity	TRR (mg/kg) of Composite Samples by Solubilisation Analysis
	101F	102F	103F	104F	105F	106F	Mean		
Peritoneal fat	<0.009	<0.009	<0.009	<0.009	<0.009	0.016	0.0003		
Perirenal fat[2]	NS	0.064	0.02	NS	0.018	NS	0.035		
Breast muscle	<0.009	<0.009	<0.009	<0.009	<0.009	<0.009	<0.009		
Leg/thigh muscle	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01		
Sikn plus subcuaneus fat	0.145	0.087	0.065	0.091	0.114	0.1	0.101	0.100	0.100*
Liver	0.107	0.125	0.123	0.160	0.164	0.207	0.148	0.139	0.139
Egg yolk	0.073	0.069	0.089	0.079	0.101	0.093	0.084	0.091	0.091

*104F excluded from pool as insufficient sample

[2] There was insufficient sample to extract perirenal fat further.

NA Not extracted.

Table 7.2.2.3-4b: Summary of total radioactive residues and extractability in tissue and egg samples from laying hens treated with ^{14}C-chlorothalonil (mean)

Sample	TRR [1]	Extractable Radioactivity		Non-extractable Radioactivity	
	mg/kg	%TRR	mg/kg	%TRR	mg/kg
Peritoneal fat	0.003	NA	NA	NA	NA
Perirenal fat[2]	0.035	NA	NA	NA	NA
Breast muscle	<0.009	NA	NA	NA	NA
Leg/thigh muscle	<0.010	NA	NA	NA	NA
Liver	0.139	58.3	0.081	41.7	0.058
Egg yolk	0.091	44.4	0.040	55.3	0.050
Whole egg[4]	0.025	-	-	-	-
Skin and Fat [3]	0.100	32.3	0.032	67.7	0.068

[1] mg/kg calculated directly from radioactivity extracted, radioactivity in the debris and specific activity.
[2] There was insufficient sample to extract perirenal fat further.
[3] Composite skin with fat samples (excludes peritoneal fat).
[4] Calculated based on TRR in egg white and yolk and corrected for weight of whole eggs. See Table 7.2.2.3-6.
NA Not extracted.

Table 7.2.2.3-4c: Summary of Radioactive Components Detected in Day 12 Excreta from hens dosed with ^{14}C-chlorothalonil

Assignment	ROI (min)	Peak as % in Excreta from hens dosed with chlorothalonil
Chlorothalonil	31.1	77.7
R182281 (SDS-3701)	33.6	4.2

Characterisation and Identification of Residues

The extracted radioactivity was analysed by TLC with UV and bio-imaging detection. Further analysis was conducted by HPLC. Metabolites were identified by comparison with reference standards and by HPLC-MS. Identification of radioactive residues is summarised in Table 7.2.2.3-5.

A sub-sample of excreta was sequentially extracted with ethyl acetate, acetonitrile, 1% formic acid in acetonitrile and methanol. Significant residues were extracted into the organic extracts corresponding to 55.6% TRR. Analysis by HPLC showed the presence of the parent molecule, chlorothalonil, as the major residue, accounting for 77.7% of the injected activity (43.2% TRR). Two minor metabolites were detected, one accounting for 1.3% of the injected radioactivity and the other corresponded to R182281, accounting for 4.2% of the injected radioactivity (2.3% TRR).

Table 7.2.2.3-5: Summary of the characterisation and identification of components in tissues and

eggs from laying hens treated with ^{14}C-chlorothalonil

		Liver		Egg Yolk		Skin and fat	
TRR (mg/kg)		0.139		0.091		0.100	
Extract for chromatography (% TRR)		49.1		42.5		32.3	
Origin of component	**Component**	**% TRR**	**Residue (mg/kg)**	**% TRR**	**Residue (mg/kg)**	**% TRR**	**Residue (mg/kg)**
Chromatographed [1]	R182281	35.9	0.050	12.5	0.011	3.2	0.004
	Unassigned [2]	1.3	0.002	5.0	0.004	1.1	0.001
	Baseline [3]	10.1	0.014	23.8	0.022	4.2	0.004
	Remainder [4]	0.9	<0.001	2.7	0.002	0.5	<0.001
	Other fractions[5]	-	-	-	-	13.3	0.013
	Losses on fractionation [6]	9.2	0.013	1.9	0.001	2.1	0.002
	Un-extracted [7]	41.7	0.058	55.3	0.050	67.7	0.068
	Total	99.1	0.137	101.2	0.090	92.1	0.092

[1] The components of the TRR that were derived from chromatographic analysis.
[2] Components resolved away from the origin in TLC. In all matrices this comprised at least three discrete components, none of which ≥2.3% TRR.
[3] Polar material on origin (TLC).
[4] Diffuse areas of radioactivity not assigned to discrete radioactive components.
[5] Extractable residues that were not analysed or gave no result in TLC analysis. No single fraction comprised ≥17.8% TRR (≥0.013 mg/kg).
[6] The net cumulative incremental losses during analysis. Calculation: 100 % - sum of all components.
[7] Radioactivity remaining in the debris after extraction with organic solvents.

In egg yolk, significant residues were extracted into the organic solvents corresponding to a total of 34.4% TRR (0.031 mg/kg). Only 10.0% TRR (0.009 mg/kg) was extracted by the aqueous solvents. Un-extracted radioactivity accounted for 55.3% TRR (0.050 mg/kg).

HPLC analysis of the organic egg yolk extract indicated that the largest region of radioactivity was R182281. TLC analysis with co-chromatography confirmed the presence of R182281 which accounted for 12.5% TRR (0.011 mg/kg). There were three other components each accounting for < 4.4% TRR (0.004 mg/kg).

HPLC analysis of the aqueous egg yolk extract was inconclusive, with no region above the limit of quantification. TLC analysis with co-chromatography indicated the presence of R182281. During TLC analysis, a proportion of radioactivity did not elute from the origin accounting for 15.0% TRR (0.014 mg/kg).

Base hydrolysis of the un-extracted radioactivity released 26.3% TRR (0.024 mg/kg), acid hydrolysis released 6.2% TRR (0.006 mg/kg) and enzyme hydrolysis released 37.5% TRR (0.034 mg/kg). HPLC analysis of these extracts was inconclusive. There were no regions above the LOQ. TLC analysis of the extracts indicated tentative identification of R182281.

Levels of R182281 identified in egg yolk were adjusted to take into account the whole egg based on the weight of egg white and yolk determined during the study. These data are summarised in Table 7.2.2.3-6. The residue level of R182281 in whole egg was calculated to be equivalent to 0.003 mg/kg on this basis.

Table 7.2.2.3-6: Calculation of residues in whole egg samples from laying hens treated with ^{14}C-chlorothalonil

	Egg Yolk			Egg White			Total Egg	
	TRR: 0.083 mg/kg			TRR: 0 mg/kg			TRR: 0.023 mg/kg	
	1.395 µg equiv			0.000 µg equiv			1.395 µg equiv	
Radiocomponent	%TRR	mg/kg	µg equiv	%TRR	mg/kg	µg equiv	µg equiv	mg/kg
R182281	12.5	0.011	0.174	0	0.000	0.000	0.174	0.003

Average weight of **yolks** that were combined for nature of residue analysis	16.802 g
Average weight of **whites** that were combined for nature of residue analysis	43.367 g
Average weight of **whole egg** (calculated from above values)	60.169 g

In liver, 58.3% TRR (0.081 mg/kg) was extracted in aqueous acetonitrile. HPLC analysis of the organic extract indicated that the major region (34.7% TRR, 0.048 mg/kg) was R182281. There was one other component present which accounted for <7% TRR. TLC analysis with co-chromatography supported the presence of R182281 (35.9%TRR, 0.050 mg/kg). Three other regions were separated, but each accounted for <1.0% TRR. During TLC analysis, a proportion of radioactivity did not elute from the origin accounting for 10.1% TRR (0.014 mg/kg). Confirmation of the identity of R182281 was achieved by LC-MS.

Initial levels of un-extracted radioactivity accounted for 41.7% TRR (0.058 mg/kg). Following repeated extraction with 2% SDS solution, 2.9% (0.004 mg/kg) was released. The remaining residue associated to protein released further radioactivity when hydrolysed with acid (3.4% TRR, 0.005 mg/kg), base (7.4% TRR, 0.010 mg/kg) and enzyme (17.1% TRR, 0.024 mg/kg).

The base hydrolysate and the protease digest were taken for chromatographic analysis. No regions above the limit of quantification were observed following HPLC analysis. TLC analysis yielded no regions that could be quantified with the majority of the radioactivity remaining at the origin.

In fat with skin, total of 11.1% TRR (0.011 mg/kg) was extracted into the organic solvents (ethyl acetate, acetonitrile, 1% formic acid in acetonitrile) and 13.3% TRR (0.013 mg/kg) was extracted into the aqueous solvents (water, 1M HCl and 1M ammonia solution). Un-extracted radioactivity accounted for 67.7% TRR (0.068 mg/kg).

HPLC analysis of the organic extract indicated that the major region (4.8% TRR, 0.005 mg/kg) was R182281. A second minor component at was also found (0.5% TRR, <0.001 mg/kg). TLC analysis with co-chromatography confirmed the presence of R182281 (3.2% TRR, 0.003 mg/kg) in addition to two unassigned regions each accounting for <1.0% TRR. During TLC analysis, a proportion of radioactivity did not elute from the origin accounting for 4.2% TRR (0.004 mg/kg).

SDS extracts were pooled and subjected to protein precipitation with diethyl ether/ethanol followed by basic, acidic and enzyme hydrolysis. The diethyl ether/ethanol extracted 4.8% TRR (0.005 mg/kg). Basic hydrolysis of the protein precipitate contained 1.3% TRR (0.001 mg/kg). Acidic hydrolysis of the protein precipitate contained 0.5% TRR (0.001 mg/kg). Enzyme hydrolysis of the protein precipitate contained 7.5% TRR (0.008 mg/kg).

Basic, acid and enzyme hydrolysis of the un-extracted residues released a further 2.2% TRR (0.002 mg/kg), 1.2% TRR (0.001 mg/kg) and 9.2% TRR (0.009 mg/kg) respectively. These extracts were not analysed further due to the low levels found.

Storage Stability Analysis

Initial analysis of the original extracts (egg yolk and liver organic fractions) took place 107 days after egg yolk collection or 85 days after necropsy. The original extracts were then re-analysed after 305 days of storage. Comparison of the initial and final radio-profiles obtained showed that no significant change in the profiles had occurred during the period of storage.

III. CONCLUSION

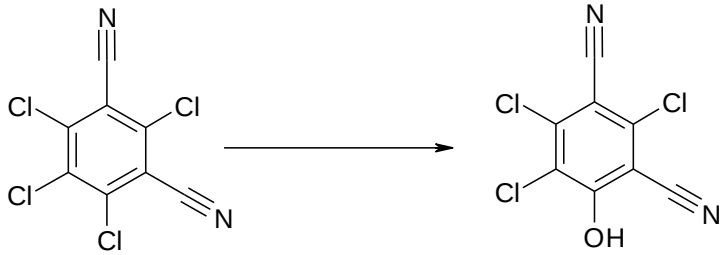
Following 14 consecutive daily doses of [phenyl-U-^{14}C]-chlorothalonil to laying hens at a nominal rate of 15 mg chlorothalonil equivalents/kg dry matter in the feed it was concluded:

- [^{14}C]-chlorothalonil and/or its biotransformation products are readily excreted, as ≥91% of the dosed radioactivity was accounted for in the excreta and cagewash.
- Radioactive residues reached a plateau concentration in eggs after 10 days
- Radioactive residues per individual were ≤0.207 mg/kg in eggs and tissues
- R182281 was the only residue identified in liver, eggs and skin plus subcutaneous fat.
- In excreta, chlorothalonil was the major extractable residue with R182281 being the only other residue identified.

Proposed Metabolic Pathway

A metabolic pathway for chlorothalonil in the laying hen is proposed.

Figure 7.2.2.3-1: Proposed metabolic pathway for chlorothalonil in laying hens



Chlorothalonil R182281

B.7.2.3 Lactating ruminants

For the initial peer review, the metabolism of chlorothalonil and SDS-3701 has been studied in lactating ruminants using ^{14}C-chlorothalonil and ^{14}C-SDS-3701 labelled uniformly in the phenyl ring. No additional studies have been submitted within the framework of the renewal of chlorothalonil.

B.7.2.3.1 Metabolism of chlorothalonil in goats

Previous evaluation	In DAR
RMS remark	Acceptable

Characteristics

reference	:	Zeneca, Duane, 1990	exposure	:	8 daily doses
type of study	:	distribution, metabolism and excretion	doses	:	0, 6 and 60 mg [2]
year of execution	:	1986 [1]	vehicle	:	0.75% methyl cellulose in water
test substance	:	[^{14}C]-chlorothalonil (radiochem. pur. 99.2 %) and chlorothalonil (pur. 99.8%)	GLP statement	:	yes
route	:	oral	guideline	:	not applicable
species	:	goat (lactating)			
group size	:	2/dose and 1 control			

[1] study initiation 1986; completion of laboratory work 1990
[2] equal to 0, 0.115 and 1.15 mg/kg bw/day

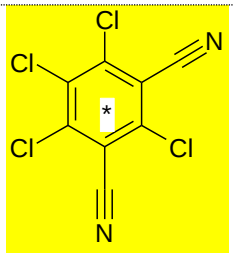
Study design

Lactating goats (50-60 kg; 2/dose) were administered 8 daily doses of 6 or 60 mg [^{14}C]-chlorothalonil in a capsule with a balling gun starting on day 0 (equivalent to 3.0 mg/kg or 30 mg/kg based on a 2 kg diet) ~~(estimated dose rates 4 N and 38 N, respectively)~~. One control animal received placebo capsules for 8 consecutive days. The animals were dosed immediately after the a.m. feeding. Milk samples were collected twice daily (a.m. and p.m.) from day -1 (before the first administration) until the day of the last application. Urine and faeces were sampled throughout the study starting at day -1. Blood was sampled prior to the last dose, during the p.m. milking on day 7, and just prior to sacrifice on day 8. Kidneys, liver, muscle, and fat samples were collected at sacrifice within 8 - 10 hours of the final dose. All samples were kept frozen until analysis. Milk and tissue samples were derivatised, extracted by several solvents, and analysed by HPLC, GC-MS, and GPC. Radiolabel was determined by combustion analysis and LSC.

AMATERIALS

A1Test Materials:

Company Code:	R044686
Use	Fungicide
IUPAC name:	tetrachloroisophthalonitrile
CAS Name:	2,4,5,6-tetrachloro-1,3-benzenedicarbonitrile
CAS Number:	1897-45-6
Molecular Formula:	$C_8Cl_4N_2$
Molecular Weight:	265.9 (unlabelled material)

Structure/Label:	[Phenyl-U-^{14}C]-Chlorothalonil
Specific Activity:	3.088 mCi/mmol
Radiochemical Purity:	99.2%
Structure: (* marks position of radiolabel)	

A2Test Animals:

Species	Goat
Gender	Female
Age	*ca* 2 years at the start of acclimatisation
Weight at dosing	50-60 kg
Number of animals	1
Acclimatisation Period	12 days
Diet	Per kg feed - 0.4 kg grain/0.6 kg roughage
Water	water, *ad libitum*
Housing	Metabolism stalls

BSTUDY DESIGN AND METHODS

B1Dosing Regime:

Nominal Dose Rate:	3.0 and 30 mg/kg (dry weight)
Actual Dose Rates	3.1-3.2 and 30-31 mg/kg (dry weight)
Mean Food consumption on Days 1 to 8 of dosing period (kg feed as received/day):	1.94 – 2.0 kg
Vehicle:	Gelatin capsule
Timing:	Once daily
Duration:	8 days
Interval from last dose to sacrifice:	10 hours after final dose

B2Sample Collection:

Milk collection:	Twice daily
Urine collection:	Once daily
Faeces collection	Once daily
Samples taken (and TRRs determined)	Milk, liver, kidney, fat, muscle, urine, faeces, gall bladder contents, heart, blood, cage wash
Samples analysed	Milk, liver, kidney, muscle, fat, urine, blood, heart

Tissue samples were homogenised whilst frozen. Radioactivity in samples was quantified by combustion and subsequent LSC analysis.

B3 Extraction and Fractionation of Residues

Standard chlorothalonil (SDS-3701) and milk extracts were methylated with diazomethane.

Milk was extracted as follows. Milk (80 g) was diluted with 75 mL of 95% ethanol. The mixture was acidified with sulphuric acid and agitated. Extraction was performed as: 1) 75/25 (v/v) diethyl ether (DEE)/hexane, break emulsion with 20 mL hexane; 2) 80/20 (v/v) DEE/hexane, break emulsion with 20 mL hexane; 3) repeat step 2; 4) 100% diethyl ether/hexane, break emulsion with 10 mL hexane; 5) 100% diethyl ether/hexane, break emulsion with 10 mL ether.

The organic phases were pooled and evaporated under a stream of clean air. The residue was dissolved in hexane. The hexane residue was extracted with 2 x 50 mL acetonitrile and these layers washed with hexane (HEX). The hexane and acetonitrile portions were concentrated under stream of clean air.

The tissues were extracted with H2SO4/acetone (1:40). Following centrifugation to remove the acetone, the residual solids were suspended in water. The acetone free supernatant was rinsed with water and partitioned as follows 1) DEE/HEX 70:30 (v/v); 2) repeat 1; 3) DEE/HEX 80:20 (v/v); 4) DEE. The organic phases were then pooled.

The organic fraction OR1 was portioned against sodium chloride. This was flash evaporated to remove DEE/HEX and the residue partitioned between acetonitrile and HEX.

A neutral pH buffer extraction method was used for kidney. Following thawing kidney was diluted on ice with buffered physiological saline (0.05 M potassium phosphate in 0.9% sodium chloride) and centrifuged.

After radioactivity measurements some supernatants were analysed by G-75 Sephadex chromatography to show molecular size distribution and HPLC to characterise the 14C distribution. HPLC was also applied to the small molecular weight fraction obtained by G-75 chromatography. Portions of supernatant and pellet were subjected to acid hydrolysis in 6N HCl (24 h, 90°C)

Results

The distribution of ^{14}C-residues was as follows:

Table 7.2.1-1: Distribution of ^{14}C-residues (in mg chlorothalonil eq/kg and % of total dose) [1]

Matrix	Low dose (6 mg)		High dose (60 mg)	
	mg eq. / kg	% of total dose	mg eq. / kg	% of total dose
Faeces	--	61	--	63
Urine	--	6.6	--	6.9
Muscle	0.005	0.1	0.03	0.1
Fat	0.004	0.1	0.03	0.1
Liver	0.08	0.2	0.71	0. 2
Kidney	0.22	0.1	2.2	0. 1

Matrix	Low dose (6 mg)		High dose (60 mg)	
	mg eq. / kg	% of total dose	mg eq. / kg	% of total dose
Milk	0.005-0.015	0.2	0.03-0.19	0.3

[1] Levels in % of total dose were cumulative levels for faeces, urine, and milk; levels at sacrifice for edible tissues.

Total residue levels in milk were 0.005-0.015 and 0.03-0.19 mg equivalents/kg in the low and high dose group, respectively, throughout the study. Highest total residue levels were detected in kidneys, (0.22 and 2.2 mg equivalents/kg in the low and high dose groups, respectively), followed by liver (0.08 and 0.7 mg equivalents/kg) and muscle and fat (<0.01 and 0.03 mg equivalents/kg, respectively, in both dose groups).

The cumulative 7-day excretion was around 62% of the total dose in faeces, somewhat below 7% in urine, and about 0.2 - 0.3% in milk for both dose levels. At sacrifice, not more than 0.1-0.2% of the dose was recovered from each edible organ. Total radioactivity excreted in the 7-day period as well as the total recovery of the radiolabel were around 70% of the dose for both dose levels.

Table 7.2.1-2: Recovery of Administered Dose

Sample	Low dose % recovered (mean)	High dose % recovered (mean)
Faeces	60 - 61 (61)	60 - 65 (63)
Urine	6.1 - 7.1 (6.6)	6.2 - 7.5 (6.9)
Muscle	0.091 - 0.10 (0.10)	0.080 (0.08)
Fat	0.063 - 0.10 (0.08)	0.048 - 0.062 (0.06)
Liver	0.16 - 0.20 (0.18)	0.14 - 0.17 (0.16)
Kidney	0.089 - 0.081 (0.09)	0.072 - 0.075 (0.07)
Milk	0.15 - 0.18 (0.17)	0.11 - 0.39 (0.25)
Total	68.0 (68.0)	67.0 - 73.4 (70.2)

Parent compound was not detected in milk and edible tissue samples (<0.01 mg/kg; detection limit of 0.0004 mg/kg in milk and 0.003 to 0.005 mg/kg in liver and kidney). SDS-3701 was the only identified metabolite of chlorothalonil in goat milk and tissue samples. The identification of SDS-3701 in milk was confirmed by methylation using diazomethane followed by GC-MS analysis. In the low dose group, SDS-3701 levels were <0.01 mg/kg in milk and in the edible organs expressing the highest total residue levels (liver and kidney). In the high dose group, SDS-3701 levels were <0.01-0.05 in milk (9 - 58% TRR - average 30%), 0.03-0.04 in liver (3 - 6% TRR), and 0.05-0.07 mg/kg in kidneys (2 - 3% TRR), and accounted for approx. 3-6% of the radiolabel in liver and kidneys.
Despite several attempts to characterise more of the residue in milk, no other compounds were identified. The residues in the non-extractable fraction (<0.01 and 0.01-0.07 mg/kg in milk from low and high dose goats, respectively) were suggested to be covalent adducts to low or moderate molecular weight proteins as determined by GPC. The residue was solubilised in ammonium carbonate/ammonium hydroxide and

characterised by gel permeation chromatography (Sephadex G-10), which indicated that the majority of components had a molecular weight of >700. The unidentified residues in milk from low and high dose goats were respectively <0.01 and 0.1 mg/kg in the organosoluble fraction, and <0.01 mg/kg for both dose levels in the watersoluble fraction.

In liver, between 17 and 37% of the residue was organosoluble and 20-30% of this fraction consisted of multiple non-polar residues. Between 21 and 31% of the total liver residue was watersoluble, presumably representing mono-, di-, and triglutathione conjugates (gel permeation chromatography (Sephadex G75) of the aqueous fraction from high dose livers, indicated molecular weights in the range of 400 to 700 for the majority of the radioactivity), since they were not converted to organosoluble components with aqueous hydrochloric acid at 90oC. Treating with hydrochloric acid in butanol produced organosoluble components, but TLC analysis indicated a very complex mixture of components. Between 30 and 45% of the liver residue remained not extracted. The levels of unidentified residues in liver from low and high dose goats were <0.01 and <0.1 mg/kg in the organosoluble fraction, 0.02 and 0.2 mg/kg in the watersoluble fraction, and 0.03 and 0.3 mg/kg in the not extracted fraction.

In kidneys, about 10-15% of the total residue was organosoluble (low and high dose group), and 19-26% and 30-48% of the radiolabel was watersoluble in the low and high dose group, respectively. The watersoluble residues mainly consisted of protein bound and smaller conjugated residue compounds. The level of non-extractable radiolabel in solids was 43-44% (0.1 mg/kg) and 35-38% (0.8 mg/kg) in low dose and high dose goats, respectively, and was not further analysed. The levels of unidentified residues in low and high dose goats were approx. 0.02 and 0.15 mg/kg in the organosoluble fraction and 0.07 and 0.7 mg/kg in the watersoluble fraction, respectively.

Residues in muscle and fat were not further characterised or identified.

More details on the analysis of the different commodities is presented below:

Milk

Milk samples were analysed using the acid treatment/extraction sequence to stabilise hydrolysis of chlorothalonil. In order to completely extract chlorothalonil and its metabolites from milk, the samples were acidified and extracted with ethyl ether and hexane. The portion of radioactivity that was not extractable into the organic phases is given as the percentage non-extractable as >95% of the 14C in the aqueous phase after extraction was associated with solids. The amount of non extractable radioactivity and that in the hexane phases is shown in Table 7.2.1-3.

Following analysis the only metabolite observed corresponding to a known analytical standard was 4-hydroxy-chlorothalonil (SDS-3701). The limit of detection of the acetonitrile fraction was 0.0004 mg/kg.

Table 7.2.1-3: Total Radioactive Residues in Goat Milk (% TRR and mg/kg)

Dose	3 mg/kg	30 mg/kg
	%TRR (mg/kg)	
Total (mg/kg)	0.005-0.015	0.03-0.19
Non Extractable	36-56 (0.002-0.006)	28-48 (0.01-0.07)

Organosoluble		
SDS-3701	30-45 (<0.001-0.007)	9-58 (<0.01-0.05)
Acetonitrile other	0-30 (<0.001-0.002)	0-23 (<0.001-0.04)
Hexane	8-16 (0.001-0.002)	7-28 (0.003-0.05)

* mg/kg total expressed as the sum of residues in the four fractions/total 14C in corresponding whole milk samples

The acetonitrile extract of the milk was analysed by non-buffered reversed phase HPLC and one minute fractions collected and counted by LSC. These were analysed against SDS-2787, -3701, -19221, -47524, -47525, -48651. Only SDS-3701 had a corresponding peak. An unknown peak in day 0 and day 1 samples comprised up to 33% of the acetonitrile fraction, at 0.018 mg/kg on day 1 pm and 0.007 mg/kg for day 6 pm. HPLC analysis following methylation suggested either multiple component or possible disproportionation reactions as both more and less polar species were observed

Florosil column chromatography separated the hexane soluble fraction into fractions A to G. The only sizable peak was E1 with 20-60% of radioactivity recovered. However, no definitive identification was achieved only characterisation, due to the amount, with similar retention time to SDS-05080. The non-extractable fraction of milk refers to the radioactivity remaining in the aqueous phase diluted with ethyl alcohol, acidified with sulphuric acid and extracted with ethyl ether/hexane (3x) and ethyl ether (2X). The precipitate would be expected for proteins following acid treatment. The precipitate also exhibited the solubility characteristics expected from protein since the majority of the material was solubilised in ammonium carbonate solutions. Therefore, suggesting compounds that are chemically bound to proteins. This was supported by the Sephadex chromatography.

Liver

Table 7.2.1-4: Total Radioactive Residues in Goat Liver (% TRR and mg/kg)

	3 mg/kg	30 mg/kg
	%TRR (mg/kg)	
Total (mg/kg)	(0.08)	(0.7)
Extractable	68.9-71.8	49.4-59.1
Organosoluble	17-31 (0.01– 0.02)	18-21 (0.13– 0.14)
SDS-3701	(0.004)	3-6 (0.03-0.04)
Acetonitrile other	(0.01)	(0.1)
Hexane	(0.004)	(0.03)
Aqueous soluble	21-31 (0.02)	25-28 (0.2)
Non Extractable	31-36 (0.03)	30-34 (0.2-0.3)
Total Conjugate	(0.02)	(0.2)

The organoextractable fraction was 17-31% of total residue and partitioning resulted in 11-15% of total radioactivity in the acetonitrile fraction. From the 30 mg/kg dose group 29-44% of the radioactivity in the acetonitrile fraction was SDS-3701. No other peaks corresponded to authentic reference standards. The aqueous extractable fraction was 20-31% of total 14C in liver. Gel permeation chromatography indicated

molecular weights in the range of 400-700 for the majority of the 14C. These were stable to HCl at 90°C.

Kidney

Table 7.2.1-5: Total Radioactive Residues in Goat Kidney (% TRR and mg/kg)

	3 mg/kg	30 mg/kg
	%TRR (mg/kg)	
Total (mg/kg)	0.2-0.24	2.1-2.3
Non Extractable [1]	43-44 (0.09-0.10)	35-38 (0.7-0.8)
Organosoluble [2]	12-15 (0.01– 0.02)	10-12 (0.20– 0.27)
SDS-3701	(0.007)	2.4-3.2 (0.05-0.07) [3]
Acetonitrile other [4]	(0.01)	4.2-5.9(0.09-0.12)
Hexane	0.6-4.4 (0.001-0.01)	2.8-4 (0.06-0.1)
Aqueous soluble [4]	19-25 (0.04-0.06)	29-48 (0.6-1.1)
Protein Bound 14C [5]	0.04*	17% of total supernatant (0.36)
		15-17 (0.34-0.36)
Total conjugate [6]	0.04*	~ (0.1- 0.15)
Di and tri glutathione conjugate of	0.01*	
chlorothalonil (tentative ID) [6]		~ (0.05)
Unknown polar [6]	<0.01*	

1 Non-extractable 14C remaining after acetone/sulphuric acid extraction/treatment

2 Ether soluble fraction extracted after removal of acetone from acetone/sulphuric acid extract

3 SDS-3701 by HPLC analysis of neutral extract supernatant

4 Calculated by subtracting the amount of SDS-3701 found from total acetonitrile fraction. Consists of multiple components shown by broad bands of radioactivity that eluted

5 From analysis of neutral extract by HPLC and Sephadex

6 From fractions detected by G-10 and G-75 Sephadex

* Estimated by extrapolation from amount in high dose

Table 7.2.1-6: Comparison of Sephadex Chromatography results for 30 mg/kg Goat Kidney Extracts as % TRR

G-10 Sephadex Aqueous fraction from acetone-sulphuric acid extraction			G-75 Sephadex Supernatant A from neutral buffer extraction	
Fraction of kidney ^{14}C (mg/kg)		48% (1.08)	Fraction of kidney ^{14}C (mg/kg)	48% (1.08)
Peak 1:	High molecular weight gives expected protein absorbance ratio	9.6% (0.20)	High molecular weight fraction (ca 53000 - protein bound ^{14}C)	17% (0.36)
Peak 2	Multiple components with retention characteristics of conjugates (standards)	15.5% (0.38)	Low molecular weight fraction conjugates 330-360	17% (0.36)
Peak 3	Very broad band; eluted after solvent pH change (thus contained strongly absorbing functional groups*)	11.5% (0.24)		

* eluted radioactivity insufficient for further analysis

Conclusion

Upon feeding of radiolabelled chlorothalonil, the majority of label was excreted via urine and faeces, while only around 1% or less of the label was recovered from milk and edible tissues. Highest residue levels were detected in kidneys and liver and SDS-3701 was the only compound identified. Chlorothalonil was not detected in any of the samples. Unidentified label presumably was partly attributable to glutathione conjugates and protein bound residue. Total radiolabel levels in milk, liver, and kidneys mounted up to 0.015, 0.08, and 0.22 mg equivalents/kg, respectively, for low dose goats, and 0.19, 0.7, and 2.2 mg equivalents/kg, respectively, for high dose goats.

Limitations

Considerable levels of unidentified residues were detected in milk, liver, and kidneys. However, reasonable efforts were undertaken to characterize and identify these residues. These residues were shown to comprise multiple components, whereby many of these are expected not to significantly exceed 0.01 mg/kg at low dose administration. It should be noted however that some of them, presumably also glutathione conjugates, are expected to exceed 0.01 mg/kg at high dose administration. Furthermore, the level of not extracted residue in kidneys of low dose animals was 43% (0.1 mg eq/kg). This study is regarded suitable for the overall evaluation.

B.7.2.3.2 Metabolism of SDS-3701 in goats

Previous evaluation	In DAR
RMS remark	Acceptable. However, some limitiation have been identified by the expert Peer Review Meeting (#164). The paragraph “limitations” has been updated accordingly.

Characteristics

reference	:	Zeneca, Ku, 1990	exposure	:	9 daily doses
type of study	:	distribution, metabolism and excretion	doses	:	0, 0.4 and 4.0 mg [1]
year of execution	:	1988	vehicle	:	0.75% methyl cellulose in water
test substance	:	[^{14}C]-SDS-3701 (radiochem. pur. 97 %) and SDS-3701 (pur. 97.0%)	GLP statement	:	yes
route	:	oral	guideline	:	not applicable
species	:	goat (lactating)			
group size	:	2/dose and 1 control			

[1] equal to 0, 0.0068, and 0.075 mg/kg bw/day

Study design

Lactating goats (50-60 kg) were administered 9 daily doses (0.4 and 4 mg) of [^{14}C]-4-hydroxy-2,5,6-trichloroisophthalonitrile ([^{14}C]-SDS-3701), the major metabolite of chlorothalonil, in a capsule with a balling gun. A control animal received placebo capsules for 9 consecutive days (day 0 - day 8). Animals were dosed immediately after the a.m. feeding. Milk samples were collected twice daily (a.m. and p.m.) beginning at the day before the first administration (day -1) up to and including day 8. Urine and faeces were sampled daily during this period. Blood samples were taken prior to the last dose, before the p.m.

milking on day 8, and just prior to sacrifice on day 8. Samples of edible tissues were sampled at sacrifice within 8 hours of the final dose and kept frozen until analysis. Urine, milk, and tissue samples were derivatised and/or extracted by several solvents and analysed by HPLC, GS-MS, GPC. Radiolabel was determined by combustion analysis and LSC.

Milk was treated with acetonitrile and the soluble fraction separated from solids. After washing with hexane and evaporation of most of the acetonitrile, the residual aqueous solution was acidified (concentrated hydrochloric acid) and partitioned with dichloromethane. Similar solvent extraction and partitioning procedures were used for other tissues although the initial extraction step was conducted using acidified solvent (acetone containing 2% hydrochloric acid for all tissues except fat; acetonitrile containing 2% hydrochloric acid for fat). The dichloromethane extracts of all tissues and milk, derived from the final partition step, were analysed by HPLC to determine the nature of the residue.

Results

The distribution of ^{14}C-residues in SDS-3701 dosed goats was as follows:

Table 7.2.2: Distribution of ^{14}C-residues in goats dosed with ^{14}C-SDS-3701

Matrix	low dose (0.4 mg)		high dose (4.0 mg)	
	mg eq. / kg	% of dose	mg eq. / kg	% of dose
Urine	--	6.4-6.5	--	8.7-9.8
Faeces	--	16.9-17.5	--	16.6-18.7
Milk [1]	0.09-0.15	13.0-18.0	0.95-1.0	15.1-22.6
Liver	0.07	2.1-2.2	0.57-0.77	1.6-2.1
Kidneys	0.17-0.26	0.9-1.1	0.82-1.35	0.4-0.7
Muscle	0.01-0.02	4.7-6.0	0.11-0.13	3.7-4.4
Fat	0.01-0.02	3.5	0.08	1.8-1.9

[1]Range of mg eq. / kg values after plateau was reached (days 6-9)

Radiolabel excreted in urine accounted for 6 to 10% of total label and the amount excreted via faeces for 17 to 19%. Between 13 and 23% of the total label was excreted via milk. At sacrifice, 6% or less or the total label was recovered from each individual edible tissue. Total radioactivity excretion was around 40-50% and total recovery around 50-60%.

Total residue levels in milk reached plateau levels in 5 to 7 days and mounted up to 0.15 and about 1.0 mg equivalents/kg for the low and high dose group, respectively. Highest total residue levels were detected in kidneys (0.17-0.26 and 0.82-1.33 mg equivalents/kg for the low and high dose group,

respectively), followed by liver (0.07 and 0.57-0.77 mg equivalents/kg, respectively) and muscle and fat (0.01-0.02 and 0.07-0.14 mg equivalents/kg in the low and high dose group, respectively).

Over 90% of the total residue in each milk and tissue sample was organosoluble and over 90% of this fraction was attributable to the parent compound SDS-3701. Confirmation of the identity of this compound in milk was achieved by methylation using diazomethane followed by GC-MS analysis. No other identifiable ^{14}C-residue was detected in the milk or tissue samples. In urine, the metabolites SDS-47524 and SDS-47525 were identified and each accounted for less then 5% of total label present in urine (<0.014 mg/kg). One unidentified urinary metabolite accounted for about 5-20% of the urinary label but was not detected in any other matrix. In total, over 90% of the total residues in milk and tissue samples was identified and less than 4% remained not extracted.

Conclusion

In all milk and tissue samples the major residue component was the administered compound SDS-3701, accounting for over 90% of the total label.

It should be noted that the supposed formation of SDS-47524 (as found in urine) from chlorothalonil is a rather uncommon biotransformation step. It is therefore doubted whether the removal of a hydroxyl group from a phenyl ring (of SDS-3701) is actually taking place. Furthermore, as name and structure of SDS-47524 as presented in this report do not match, possibly SDS-47524 has been mixed up with SDS-47523 as presented in a tomato metabolism study by the same notifier. However, also formation of SDS-47523 from SDS-3701 in goat would be very unlikely. Therefore, none of these metabolites (SDS-47523/SDS-47524) is included as identified in goat.

Limitations

~~No limitations.~~

During expert Peer Review Meeting, it was concluded that the reported data is not complete and not presented according to OECD recommednations. The only information is that SDS-3701 accounted for over 90% of the total label in milk and tissues. No detailed information on identification and characterisation steps for metabolites in all the fractions has been reported and it is still relevant.

B.7.2.3.3 Metabolism of chlorothalonil in goats

Previous evaluation	In DAR
RMS remark	Supportive

Characteristics

reference	:	Vischim, Shaw, 1997	exposure	:	5 daily doses
type of study	:	distribution, metabolism and excretion	doses	:	20 mg [1]
year of execution	:	1986 [1]	vehicle	:	D-(+)-glucose in a capsule
test substance	:	[ring-^{14}C]-chlorothalonil	GLP statement	:	yes

		(radiochem. pur. 98 %) and chlorothalonil (pur. 99%)			
route	:	oral	guideline	:	not applicable
species	:	lactating goat (British Saanen)			
group size	:	1			

[1] Stated by the authors of the study to be approx. equivalent to 10 mg as/kg feed. As calculated by the reviewer, equivalent to 0.42-0.46 mg as/kg bw/day (see Guidelines and limitations).

Study design

A lactating goat (47.5-43.5 kg; see Guidelines and limitations) was administered five consecutive daily doses of 20 mg ^{14}C- chlorothalonil (equivalent to approx. 10 mg/kg feed as stated by the authors, and equivalent to 0.42-0.46 mg as/kg bw/day ~~(estimated dose rate 15 N in comparison to the preliminarily calculated TMDI on the basis of the Zeneca dossier)~~; see Guidelines and limitations) in a capsule with a balling gun after the morning milking. Milk samples were collected twice daily from immediately prior to dosing until 23 h after the last dose. Urine and faeces were sampled daily throughout the study starting 24 h preceding the first dose, up to 23 h after the last dose. Blood samples were taken 1 h before and 2 h after each daily dose and then at 1,2,3,4,6, 8 and 12 h after the last dose. Liver, kidneys, bile, rumen, reticulum and contents, omasum, abomasum and contents, intestines and contents, samples of muscle, fat and blood were collected at sacrifice and kept frozen until analysis. Radioactivity in excreta and tissues was quantified by (combustion) LSC. Radioactivity from milk and tissue samples was extracted by several solvents (acetone, acetonitrile, acetonitrile/water) and released by treatment with protease, dilute acid and base and determined by combustion analysis and LSC. In addition, for liver and kidney samples radioactivity was released by acid hydrolysis. Extracted residue components were identified by co-chromatography (TLC, HPLC), GC-MS and LC-MS.

Results

The distribution of ^{14}C-residues were as follows:

Table 7.2.5: Distribution of radiolabel (in mg chlorothalonil equivalents/kg and % of total dose)[1]

Matrix	mg eq. / kg	% of total dose
Faeces	--	12
Urine	--	39
Muscle	0.02	
Fat	0.03	
Liver	0.23	
Kidney	1.1	
Milk	0.09	

[1] Levels in % of total dose were cumulative levels for faeces, urine, and milk; levels at sacrifice for edible tissues.

At the time of sacrifice, 51% of the total administered radioactive dose had been excreted via urine and faeces and 0.3% via milk. Unchanged chlorothalonil was not present in either urine or faeces. A further 23% was recovered from the GIT. At sacrifice, not more than 0.06-0.2% of the total dose was recovered from each individual edible organ. The overall recovery of the total dose from the animal at sacrifice was 75%. Concentrations in plasma and milk rose throughout the dosing period. Steady state concentrations were not yet reached after five days. Highest total residues were detected in milk (0.09 mg equivalents/kg), liver (0.23 mg equivalents/kg) and kidneys (1.1 mg equivalents/kg). Levels in muscle and fat remained <0.05 mg equivalents/kg (0.018 and 0.028 mg equivalents/kg, respectively).

In pooled milk (0.060 mg equivalents/kg), 0.039 mg equivalents/kg (65% TRR) was extracted and further split in an organosoluble fraction (0.022 mg equivalents/kg; 37% TRR) and an aqueous extract (0.017 mg equivalents/kg; 28%TRR). The not extracted debris contained 0.021 mg equivalents/kg (36% TRR) and was further characterized by various treatments (protease, 0.1 M acid, 0.1 M base) resulting in various fractions ≤0.011 mg equivalents/kg (≤19% TRR). It should be noted that plateau levels were not yet reached during the 5 day dosing period.

In liver (0.23 mg equivalents/kg), 0.12 mg equivalents/kg (52% TRR) was extracted and further split in an organosoluble fraction (0.072 mg equivalents/kg; 31% TRR) and an aqueous extract (0.048 mg equivalents/kg; 20%TRR). The not extracted fraction contained 0.11 mg equivalents/kg (49% TRR) and was further characterized by various treatments (protease, 0.1 M acid, 0.1 M base) resulting in a release of fractions varying from 0.012-0.037 mg equivalents/kg (5.2%-16% TRR).

In kidneys (1.1 mg equivalents/kg), 0.059 mg equivalents/kg (54% TRR) was extracted and further split in an organosoluble fraction (0.11 mg equivalents/kg; 9.7% TRR) and an aqueous extract (0.48 mg equivalents/kg; 44%TRR). The not extracted fraction contained 0.51 mg equivalents/kg (46% TRR) and was further characterized by various treatments (protease, 0.1 M acid, 0.1 M base). This resulted in a release of fractions varying from 0.067-0.19 mg equivalents/kg (6.1%-17.5% TRR) including a remaining not extracted fraction (0.058 mg equivalents/kg; 5.3% TRR). According to the authors of the study the released residue is composed of covalent adducts to tissue macromolecules. The released fractions, of which some contained >0.05 mg equivalents/kg, were not identified.

Metabolite identification was preceded by extraction procedures that were not identical to the ones used for characterization and included HPLC analysis. Parent compound was not detected in any of the excreta and tissues examined. Only two metabolites were identified in milk, liver, fat and muscle, i.e. 4-hydroxychlorothalonil (that was coded SDS-3701 by Zeneca) and 2,5,6-trichloro-1,3-dicyanobenzene (R613801). Metabolite profiles (including remaining non identified metabolites) were similar. The highest level of 4-hydroxychlorothalonil (SDS-3701) was about 0.025 mg equivalents/kg and found in milk. Only in liver and muscle, the level of 2,5,6-trichloro-1,3-dicyanobenzene (R613801) outreached that of 4-hydroxychlorothalonil (SDS-3701). In liver, levels were 0.030 and 0.024 mg eq/kg, respectively. In

muscle, they were 0.006 and 0.002 mg eq/kg, respectively. The metabolite R613801 has been characterized but not identified due to the lack of a specific analytical standard.

Other metabolites remained unidentified and were each found at levels <0.05 mg equivalents/kg. Two metabolites were identified in urine and presumably also in kidneys. These were the mono- and diglutathione conjugates of chlorothalonil. In urine, the latter represented 3.2% of the total dose. In kidneys, one major compound (at a level of 0.074 mg equivalents/kg) was not identified.
The freezer storage stability of R182281 and of R613801, was investigated during the study for seven months after sacrifice. Residues in milk were similar on each occasion. In liver, fat and muscle, the situation was different. The concentration of R613801 decrease after seven months of storage, on the contrary the concentrations of R182281 increased during the storage period. These results may suggest that R613801 is a precursor of R182281.

Conclusion

Upon feeding of radiolabelled chlorothalonil, about 50% of the label was excreted through urine and faeces. By the time of sacrifice, 23% was found in the GIT. Highest residue levels were detected in milk, kidneys and liver and mounted up to 0.09, 0.23 and 1.1 mg equivalents/kg, respectively. 4-Hydroxychlorothalonil (SDS-3701) and 2,5,6-trichloro-1,3-dicyanobenzene (R613801) were the only metabolites identified in edible tissues. Both were found in various commodities, the highest levels of 4-hydroxy-chlorothalonil (SDS-3701) were found in liver and milk (approx. 0.025 mg/kg in both commodities). The highest level of 2,5,6-trichloro-1,3-dicyanobenzene (R613801) was found in liver (0.030 mg eq/kg). Glutathione conjugates were found in urine and tentatively in kidneys, albeit at low levels. Chlorothalonil was not identified in any of the samples.

Limitations

The health status of the goat is very questionable as it lost 10 kg during the 11-day pre-dosing period and another 4 kg during the 5-day dosing period, which means a total decrease in body weight of approx. 24%. Furthermore, because of this significant weight loss of the goat during the experiment, calculation of the doses expressed as mg as/kg bw/d is only indicative.

It should be noted also that a major part of TRR in the kidneys (41%), released by the various treatments of the not extracted residue was not identified although the residue level of the released fractions mounted up to 0.076-0-19 mg equivalents/kg. These fractions could still contain bioavailable compounds as some of these fractions could be released by 0.1 M acid treatment for a short time.

Considering these aspects, this study is regarded not suitable for the overall evaluation. The results can be used merely indicative. As glutathione conjugates are very common metabolites of chlorinated benzene derivatives, they were included as identified metabolites.

B.7.2.4 Pigs

The metabolism of chlorothalonil in ruminants was similar to that seen in the rat and therefore a metabolism study in pigs is not required.

Previously, there were discussions regarding the requirement of metabolism studies in pigs, since the formation of SDS-3701 was not observed in rat, while it was a major metabolite in ruminants. However, a repeat rat biotransformation study has now demonstrated that SDS-3701 is the most abundant component circulating in rat plasma, accounting for up to 38% of the total circulating radioactivity exposure. Therefore, the metabolic patterns of chlorothalonil are considered not different between the monogastric rat and ruminants.

B.7.2.5 Fish

For the time being there are no agreed test guidelines for the estimation of the dietary burden of pesticide residues for fish or for the design and conduct of fish metabolism studies. Therefore, no fish metabolism studies have been conducted (see also SANCO/10181/2013 rev 2.1). Furthermore, regarding the representative uses, potato, wheat and barley grain are considered potential ingredients for fish feed. Since the low residue levels in these commodities, a significant exposure to fish is unlikely.

If the working document SANCO/11187/2013 is used, it suggests that the guidance shall be applied to all active substances that are fat soluble, i.e. substances with log Pow ≥ 3. The log Pow of chlorothalonil is 2.94 at 25°C, pH = ca. 7. In addition, the log Pow for SDS-3701 is 0.6-1.0, showing also low fat solubility. Therefore, chlorothalonil does not meet the criteria for a fish metabolism study. Furthermore, from the animal metabolism studies it can be observed that residues in fat tissue, if any, are very low compared to residues in other tissues, confirming low fat solubility of residues and the conclusion that no fish metabolism study is required.

In addition, on a metabolism basis, studies have been conducted in rat, dog, monkey, hen and goat. The results of these studies demonstrate that the metabolism of chlorothalonil is uncomplicated with the primary metabolite SDS-3701 (4-hydroxychlorothalonil) observed in all species. Additional metabolism is possible via the glutathione pathway, hydrolysis of the nitrile to the amide to carboxylic acid and potentially reductive dehalogenation. However, these pathways generally result in low residues. In the two livestock species used, the main metabolite considered is SDS-3701. As the basic metabolic step of hydroxylation of the reactive chlorine in the 4 position is likely to be conserved in fish, via P450 enzymes. There is a strong weight of evidence that the primary and possibly only detectable metabolite in fish would also be SDS-3701.

B.7.3 Magnitude of residue trials in plants

The defended uses for the renewal of chlorothalonil are on wheat, barley, tomatoes and potatoes. For the initial peer review, the crops which were at the start of the peer review requested, also included these crops (in addition to several other crops). At the end of the initial peer review process, wheat was the only crop which remained as defended use. Furthermore, often the GAPs during the initial peer review were more critical than the GAPs applied for within the framework of the renewal. Therefore, new trials are available for all defended uses for the renewal.

B.7.3.1 Tomatoes

Chlorothalonil is proposed for use on tomato in NEU and SEU according to the following GAP:
1x 1000 g/ha, PHI 3 days.

This GAP is less critical than the GAP for tomatoes during the initial peer review, in which the number of application was 3 or 4 in the corresponding trials. Therefore, the evaluation of these trials is not copied from the old DAR into this RAR.

For the renewal of chlorothalonil, 3 additional reports containing supervised residue trials have been submitted. The residue reports supporting the proposed EU critical GAP for chlorothalonil on tomato are referenced in Table 7.3.1-1 and the data are presented in Table 7.3.1-4.

Previous evaluation	Submitted for the purpose of renewal
RMS remark	Acceptable It should be noted that conjugates of chlorothalonil and metabolite SDS-3701 (included in the RD for risk assessment) have not been measured in the field trials.

Table 7.3.1-1: Report references for trials supporting the proposed EU critical GAP for chlorothalonil on tomato

Annex Pt.	Number.	Author/s	Issue Year	Report Title
K-CA 6.3.1/01	(1 of 3)	D Schulz, C Trumper	2016	Chlorothalonil - Residue study on Field Tomatoes in Southern France, Spain and Italy in 2014 Syngenta File No. A7867A_11391, Syngenta Report No. S14-02773
K-CA 6.3.1/02	(2 of 3)	D Schultz C Trumper	2015	Chlorothalonil - Residue study on Field Tomatoes in Northern France, Poland and Hungary in 2014 Syngenta File No. A7867A_11386, Report No. S14-02774
K-CA 6.3.1/03	(3 of 3)	D Schultz C Trumper	2016a	Chlorothalonil – Residue study on Field Tomato in Northern France and Germany in 2015 Syngenta File No. A7867A_11403, Report No. S15-02003

Guidelines

The studies meet the requirements of the Commission of the European Communities: General Recommendations for the Design, Preparation and Realization of Residue Trials (7029/V1/95 rev. 5, 22/7/1997), and are designed to comply with Regulation (EC) 1107/2009.

GLP

All trials (field and analytical phases) were carried out in compliance with the principles of Good Laboratory Practice.

Materials and Methods

Sixteen supervised residue trials were conducted on field grown tomato in 2014 and 2015, in northern or southern Europe. A summary of the trials conducted is presented in Table 7.3.1-2.

Table 7.3.1-2: Summary of chlorothalonil residue trials on tomato

Country	2014	2015
Northern Europe		
France (north)	1 Decline	1 Decline; 1 Harvest
Germany	-	1 Decline; 1 Harvest
Hungary	1 Harvest	-
Poland	1 Decline; 1 Harvest	
Southern Europe		
France (south)	1 Harvest	-
Spain	2 Decline; 3 Harvest	-
Italy	2 Decline	-

Decline trials are those with three or more sampling times.

Tomatoes are a major crop in northern and southern Europe and therefore generally require eight trials in each residue region.

Treatments with chlorothalonil were conducted as post emergence (BBCH 61-89) spray applications utilising the formulation as detailed in Table 7.3.1-3 at a nominal application rate of 1000 g a.s./ha (actual rates 959-1086 g a.s./ha).

Table 7.3.1-3: Summary of chlorothalonil formulations used in the presented trials

Product code	Formulation type	Composition	
		2014	2015
A7867A	SC	498 g/L chlorothalonil	510 g/L chlorothalonil

Samples of whole fruits were taken and analysed for residues of parent chlorothalonil and the metabolite R182281 using analytical method GRM005.01A with an LOQ of 0.01 mg/kg for both compounds. The method involved extraction with acetone/5M sulphuric acid (95:5 v/v), dilution with water followed by SPE clean up for chlorothalonil or taking up in acetonitrile:water (50/50 v/v) for R182281. Subsequently, analysis was performed by gas chromatography with mass selective detection (GC-MSD) for chlorothalonil and LC-MS/MS for R182281. Full method descriptions and validation data are presented in B.5.2.1 (K-CA 4.2/01-K-CA 4.2/07). Procedural recovery data are presented with the results of the residue trials in Table 7.3.1-4. Linearity was acceptable with an R^2 >0.99.

Samples were stored up to a maximum of 9 months from sampling to extraction. Samples were homogenised in the presence of acid before freezing. Residues of chlorothalonil and R18221 are stable in acidified homogenised tomatoes for at least 24 months (see section B.7.1) and therefore no degradation will have occurred between sampling and analysis.

No residues of Chlorothalonil and R182281 at or above the LOQ (0.01 mg/kg) were found in the untreated ~~potato tuber~~ tomato samples.

All trials have been conducted according to the cGAP of the defended use. Therefore, in total, for NEU 8 acceptable trials are available, and for SEU also 8 acceptable trials are available.

The results of the residue trials for chlorothalonil and R182281 are presented in Table 7.3.1-4.

Table 7.3.1-4: Summary of residue data supporting the EU critical GAP for chlorothalonil on tomato

GLP and Trial Details	Crop (Variety)	Country (Region)	Application Rate Product Code	Growth Stage at application	PHI (days)	Crop Part (Growth stage at sampling)	Residue Found (Uncorrected)		Recovery Data
							chlorothalonil mg/kg	R182281 (mg/kg)	
Northern Europe									
Report: S14-02774 Study: : S14-02774 Trial: S14-02774-01 - Study to GLP - Study carried out in 2014	Tomato (Topkapi)	FRANCE (Europe North) Varennes Sur Loire Pays de la Loire Maine et Loire	1039 g a.s./ha A7867A	BBCH 83 09/09/2014	0	Fruit (BBCH 83)	0.69	< 0.01	Chlorothalonil Whole fruit: mean = 84% RSD = 7.9% (n = 3 in 0.01 – 2.0 mg/kg spiking range) R182281 Whole fruit: mean = 105% RSD = NA (n = 2 in 0.01 – 0.10 mg/kg spiking range) Chlorothalonil (Fruit) GRM005.01A R182281 (Fruit) GRM005.01A
					1	Fruit (BBCH 83)	0.27	< 0.01	
					3	Fruit (BBCH 83)	0.63	< 0.01	
Report: S14-02774 Study: : S14-02774 Trial: S14-02774-03 - Study to GLP - Study carried out in 2014	Tomato (Galilea)	POLAND (Europe North) Gaj Maly Wielkopolska Szamotulski 64-520	987 g a.s./ha A7867A	BBCH 84-85 19/08/2014	0	Fruit (BBCH 84-85)	0.46	< 0.01	
					1	Fruit (BBCH 84-85)	0.34	< 0.01	
					3	Fruit (BBCH 85-86)	0.15	<.0.01	
Report: S14-02774 Study: : S14-02774 Trial: S14-02774-06 - Study to GLP - Study carried out in 2014	Tomato (Benito)	POLAND (Europe North) Chrzypsko Wielkie Wielkopolskie Miedzychodzki 64-412	1038 g a.s./ha A7867A	BBCH 84-85 22/08/2014	3	Fruit (BBCH 85-86)	0.44	< 0.01	
Report: S14-02774 Study: : S14-02774 Trial: S14-02774-07 - Study to GLP - Study carried out in 2014	Tomato (Alfréd F1)	HUNGARY (Europe North) Szatymaz Dél-Alföd Csongrád 6763	1049 g a.s./ha A7867A	BBCH 85 09/09/2014	3	Fruit (BBCH 87)	0.26	< 0.01	
Report: S15-02003 Study: : S15-02003 Trial: S15-02003-01 - Study to GLP - Study carried out in	Tomato (Petula)	FRANCE (Europe North) Varennes Sur Loire Pays de la Loire	1013 g a.s./ha A7867A	BBCH 85-86 28/08/2015	0	Fruit (BBCH 85-86)	0.49	< 0.01	Chlorothalonil Whole fruit: mean = 94% RSD = 8.4% (n = 4 in 0.01 – 1.0 mg/kg spiking range)
					1	Fruit (BBCH 85-86)	0.43	< 0.01	

GLP and Trial Details	Crop (Variety)	Country (Region)	Application Rate Product Code	Growth Stage at application	PHI (days)	Crop Part (Growth stage at sampling)	Residue Found (Uncorrected) chlorothalonil mg/kg	 R182281 (mg/kg)	Recovery Data
2015		Maine-et-Loire 49730			3	Fruit (BBCH 85-86)	0.13	< 0.01	R182281 Whole fruit: mean = 101% RSD = NA (n = 2 in 0.01 – 0.10 mg/kg spiking range) Chlorothalonil (Fruit) GRM005.01A R182281 (Fruit) GRM005.01A
Report: S15-02003 Study: : S15-02003 Trial: S15-02003-02 - Study to GLP - Study carried out in 2015	Tomato (Phantasia)	GERMANY (Europe North) Kirchheim Baden-Württemberg Heidelberg, Stadtkreis 69124	988 g a.s./ha A7867A	BBCH 61-83 27/07/2015	0	Fruit (BBCH 61-89)	0.43	< 0.01	
					1	Fruit (BBCH 61-89)	0.56	< 0.01	
					3	Fruit (BBCH 61-89)	0.36	< 0.01	
Report: S15-02003 Study: : S15-02003 Trial: S15-02003-03 - Study to GLP - Study carried out in 2015	Tomato (Monfavet)	FRANCE (Europe North) St Hilaire St Mesmin Loiret 45160	986 g a.s./ha A7867A	BBCH 72-81 03/08/2015	3	Fruit (BBCH 81-85)	0.25	< 0.01	
Report: S15-02003 Study: : S15-02003 Trial: S15-02003-04 - Study to GLP - Study carried out in 2015	Tomato (Pannovy)	GERMANY (Europe North) Markgröningen Baden-Württemberg Ludwigsburg 71706	977 g a.s./ha A7867A	BBCH 74-82 18/08/2015	3	Fruit (BBCH 75-85)	0.83	< 0.01	
Southern Europe									
Report: S14-02773 Study: S14-02773 Trial: S14-02773-02 - Study to GLP unchecked - Study carried out in 2014 (Field)	Tomato (H-9036)	SPAIN (Europe South) Remolinos Zaragoza Aragon 50635	959 g ai/ha (A7867A) (-)	BBCH 85 - 87 25/08/2014	0	Fruit (BBCH 87-89)	0.80	< 0.01	Chlorothalonil Fruit Mean = 93% RSD = 7% (n = 5 in 0.01 - 2 mg/kg spiking range) R182281 Fruit Mean = 103% RSD = 14% (n = 4 in 0.01 - 0.1 mg/kg spiking range)
					1	Fruit (BBCH 87-89)	0.39	< 0.01	
					3	Fruit (BBCH 87-89)	0.36	< 0.01	
Report: S14-02773 Study: S14-02773 Trial: S14-02773-03 - Study to GLP unchecked - Study carried out in 2014 (Field)	Tomato (Gamlex)	ITALY (Europe South) Lagosanto Emilia Romagna Ferrara 44023	1086g ai/ha (A7867A) (-)	BBCH 87 18/08/2014	0	Fruit (BBCH 87-89)	1.64	< 0.01	Chlorothalonil (Fruit) GRM005.01A R182281 (Fruit) GRM005.01A
					1	Fruit (BBCH 87-89)	0.87	< 0.01	
					3	Fruit	0.58	< 0.01	

GLP and Trial Details	Crop (Variety)	Country (Region)	Application Rate Product Code	Growth Stage at application	PHI (days)	Crop Part (Growth stage at sampling)	Residue Found (Uncorrected) chlorothalonil mg/kg	 R182281 (mg/kg)	Recovery Data
						(BBCH 87-89)			
Report: S14-02773 Study: S14-02773 Trial: S14-02773-04 - Study to GLP unchecked - Study carried out in 2014 (Field)	Tomato (Littano)	ITALY (Europe South) Crevalcore Emilia Romagna Bologna 40014	1003 g ai/ha (A7867A) (-)	BBCH 85 – 87 01/08/2014	0	Fruit (BBCH 87-89)	1.02	0.02	
					1	Fruit (BBCH 87-89)	0.86	< 0.01	
					3	Fruit (BBCH 87-89)	0.59	< 0.01	
Report: S14-02773 Study: S14-02773 Trial: S14-02773-05 - Study to GLP unchecked - Study carried out in 2014 (Field)	Tomato (Hector)	FRANCE (Europe South) (-) Saint Laurent de la Salanque Languedoc-Roussillon Pyrénées-Orientales 66250	1016 g ai/ha (A7867A) (-)	BBCH 87 04/08/2014	3	Fruit (BBCH 89)	0.06	< 0.01	
Report: S14-02773 Study: S14-02773 Trial: S14-02773-06 - Study to GLP unchecked - Study carried out in 2014 (Field)	Tomato (H-9036)	SPAIN (Europe South) Ribaforada Comunidad Foral de Navarra Navarra 31550	1085 g ai/ha (A7867A) (-)	BBCH 85 – 87 07/10/2014	3	Fruit (BBCH 87-89)	0.58	< 0.01	
Report: S14-02773 Study: S14-02773 Trial: S14-02773-07 - Study to GLP unchecked - Study carried out in 2014 (Field)	Tomato (Albatros)	SPAIN (Europe South) Las Marismillas (Lebrija) Andalucia Sevilla 41740	983 g ai/ha (A7867A) (-)	BBCH 87 – 89 15/07/2014	3	Fruit (BBCH 88-89)	0.12	< 0.01	
Report: S14-02773 Study: S14-02773 Trial: S14-02773-09 - Study to GLP unchecked - Study carried out in 2014	Tomato (Matias)	SPAIN (Europe South) Conil de la Frontera Andalucia Cádiz	960 g ai/ha (A7867A) (-)	BBCH 81 – 82 15/09/2014	0	Fruit (BBCH 82-83)	0.37	< 0.01	
					1	Fruit (BBCH 82-83)	0.36	< 0.01	

GLP and Trial Details	Crop (Variety)	Country (Region)	Application Rate Product Code	Growth Stage at application	PHI (days)	Crop Part (Growth stage at sampling)	Residue Found (Uncorrected) chlorothalonil mg/kg	 R182281 (mg/kg)	Recovery Data
(Field)					3	Fruit (BBCH 82-83)	0.17	< 0.01	
Report: S14-02773 Study: S14-02773 Trial: S14-02773-10 - Study to GLP unchecked - Study carried out in 2014 (Field)	Tomato (Bosca)	SPAIN (Europe South) La Palma del Condado Andalucia Huelva	1026 g ai/ha (A7867A) (-)	BBCH 88 – 89 15/09/2014	3	Fruit (BBCH 88-89)	0.46	< 0.01	
Unless otherwise stated residues of chlorothalonil and R182281 in untreated samples were less than the LOQ.									

B.7.3.2 Barley

Chlorothalonil is proposed for use on barley in NEU and SEU according to the following GAP:

2x 750 g/ha, interval 14 days, BBCH 30-59 (no PHI).

The requested GAPs for barley during the initial peer review were more critical regarding dose rate, and either comparable regarding application timing (BBCH 59) or more critical (BBCH 75) than the GAP applied for within the framework of the renewal. Therefore, none of the trials have been copied into this RAR.

For the renewal of chlorothalonil, additional reports containing supervised residue trials have been submitted. The residue reports supporting the proposed EU critical GAP for chlorothalonil on ~~tomato~~ barley are referenced in Table 7.3.2-1 and the data are presented in Table 7.3.2-4.

Previous evaluation	Submitted for the purpose of renewal
RMS remark	Acceptable, pending the submission of storage stability data for chlorothalonil in cereal grain and straw and for metabolite SDS-3701 in cereal grain. It should be noted that conjugates of chlorothalonil and metabolite SDS-3701 (included in the RD for risk assessment) have not been measured in the field trials.

Table 7.3.2-1: Report references for trials supporting the proposed EU critical GAP for chlorothalonil on barley

Annex Pt.	Number.	Author/s	Issue Year	Report Title
K-CA 6.3.2/01	(1 of 9)	T White	2013	Chlorothalonil – residue study on barley in northern France and the United Kingdom in 2011. Eurofins Agroscience Services Ltd, Slade Lane, Wilson, Melbourne, Derbyshire, UK, Syngenta File No. A14111B_10905, Report No. S11-00522
K-CA 6.3.2/02	(2 of 9)	T White	2014	Chlorothalonil – residue study on barley in Germany, Poland and the United Kingdom in 2012. Eurofins Agroscience Services Ltd, Slade Lane, Wilson, Melbourne, Derbyshire, UK, Syngenta File No. A14111B_10908, Report No. S11-01274
K-CA 6.3.2/03	(3 of 9)	T White	2014a	Chlorothalonil – residue study on barley in Spain, Italy and southern France in 2011. Eurofins Agroscience Services Ltd, Slade Lane, Wilson, Melbourne, Derbyshire, UK, Syngenta File No. A14111B_11144, Report No. S11-00523
K-CA 6.3.2/04	(4 of 9)	T White	2014b	Chlorothalonil – residue study on barley in southern France, Italy and Spain in 2012, Final Report Amendment 1. Eurofins Agroscience Services Ltd, Slade Lane, Wilson, Melbourne, Derbyshire, UK, Syngenta File No. A14111B_10899, Report No. S12-01275
K-CA 6.3.2/05	(5 of 9)	T White	2013a	Chlorothalonil – residue study on barley in southern France in 2013 . Eurofins Agroscience Services Ltd, Slade Lane, Wilson, Melbourne, Derbyshire, UK,Syngenta File No. A14111B_10861, Report No. S13-01041

Annex Pt.	Number.	Author/s	Issue Year	Report Title
K-CA 6.3.2/06	(6 of 9)	A Sala	2014	Determination of chlorothalonil and its metabolite SDS3701 residues in raw agricultural commodity barley (grain, straw), following two applications of chlorothalonil 500 SC, 2 trials, northern Europe, year 2013. Research Centre Biospheres, Via Vittorio Veneto, 81, 26857 Salerano sul Lambro, Italy, Syngenta File No R044686_11190, Report No. RAU-020-13
K-CA 6.3.2/07	(7 of 9)	A Sala	2014a	Determination of chlorothalonil and its metabolite SDS3701 residues in raw agricultural commodity barley (grain, straw), following two applications of chlorothalonil 500 SC, 2 trials, southern Europe, year 2013. Research Centre Biospheres, Via Vittorio Veneto, 81, 26857 Salerano sul Lambro, Italy, Syngenta File No. R044686_11181, Report No. RAU-018-13
K-CA 6.3.2/08	(8 of 9)	F Mazzi	2014	Determination of chlorothalonil and its metabolite SDS3701 residues in raw agricultural commodity barley (plant, silage, hay, grain, straw) following two applications of Clortosip 500 SC (northern Europe – 6 trials year 2014). Research Centre Biospheres, Via Vittorio Veneto, 81, 26857 Salerano sul Lambro, Italy, Syngenta File No. R044686_11180, Report No. BIU-017-14
K-CA 6.3.2/09	(9 of 9)	F Mazzi	2014a	Determination of chlorothalonil and its metabolite SDS3701 residues in raw agricultural commodity barley (plant, silage, hay, grain, straw) following two applications of Clortosip 500 SC (south Europe – 6 trials year 2014). Research Centre Biospheres, Via Vittorio Veneto, 81, 26857 Salerano sul Lambro, Italy, Syngenta File No. R044686_11182, Report No. BIU-016-14

Guidelines

The studies meet the requirements of the Commission of the European Communities: General Recommendations for the Design, Preparation and Realization of Residue Trials (7029/V1/95 rev. 5, 22/7/1997), and are designed to comply with Regulation (EC) 1107/2009.

GLP

All trials (field and analytical phases) were carried out in compliance with the principles of Good Laboratory Practice.

Materials and Methods

Thirty-two supervised residue trials were conducted on barley in 2011, 2012, 2013 and 2014 in northern or southern Europe. A summary of the trials conducted is presented in Table 7.3.2-2.

Table 7.3.2-2: Summary of chlorothalonil residue trials on barley

Country	2011	2012	2013	2014
Northern Europe				
United Kingdom	2 Harvest	1 Harvest	-	-
France (north)	1 Harvest	2 Harvest	1 Harvest	3 Harvest
Germany	1 Harvest	-	-	-
Poland	-	1 Harvest	-	3 Harvest
Belgium	-	-	1 Harvest	-
Southern Europe				
France (south)	-	2 Harvest	2 Harvest	1 Harvest
Spain	1 Harvest	1 Harvest	-	-
Italy	2 Harvest	1 Harvest	1 Harvest	5 Harvest

Barley is a major crop in northern and southern Europe and therefore generally requires eight trials in each residue region.

Treatments with chlorothalonil were conducted as two post emergence (BBCH 30-32 [up to BBCH 63 for some trials] and BBCH 59 [up to BBCH 69 for some trials]) spray applications utilising the formulation as detailed in Table 7.3.2-3 at a nominal application rate of 750 g a.s./ha (actual rates 682-806 g a.s./ha). The water volumes during application ranged from 94 to 413 L/ha.

Table 7.3.2-3: Summary of chlorothalonil formulations used in the presented trials

Product code	Formulation type	Composition			
		2011	2012	2013	2014
A14111B	SC	385 g/L chlorothalonil 78.4 g/L azoxystrobin	384 g/L chlorothalonil 74.7 g/L azoxystrobin	384 g/L chlorothalonil 74.7 g/L azoxystrobin	-
Chlorothalonil 500 SC	SC	-	-	502 g/L chlorothalonil (batch O232)	507 g/L chlorothalonil (batch PN1911)

Samples of various parts of mature and immature barley plants were taken and analysed for residues of chlorothalonil and R182281 using either analytical method GRM005.01A with an LOQ of 0.01 mg/kg for both compounds in all commodities analysed, or by analytical methods described in study BIU-016-14, with an LOQ of 0.01 mg/kg in grain and 0.05 mg/kg for other commodities for chlorothalonil and 0.02 mg/kg for all commodities for R182281. Method GRM005.01A involved extraction with acetone/5M sulphuric acid (95:5 v/v), dilution with water followed by SPE clean up for chlorothalonil or taking up in acetonitrile:water (50/50 v/v) for R182281. Subsequently, analysis was performed by gas chromatography with mass selective detection (GC-MSD) for chlorothalonil using two ion masses and LC-MS/MS for R182281 using 2 mass transitions. Study 6.3.2/01 to 6.3.2/05 included minor modifications. The analytical method (sometimes indicated by RAU076-01) used in study RAU-020-13, RAU-018-13, BIU-017-14 and BIA-016-14 involved extraction with acidified ethyl acetate followed by analysis using a gas chromatograph equipped with µ-ECD detector for chlorothalonil. For R182281 the analytical method consisted of extraction with methanol and analysis with LC-MS/MS. Full method descriptions and validation data are presented in B.5.2.1 (K-CA 4.2/01-K-CA 4.2/07) and B.5.1.2.5 (K-CA 4.1.2/32). Procedural recovery data are presented with the results of the residues trials in Table 7.3.2-4.

Samples were stored up to a maximum of 12 months from sampling to extraction. Samples of whole plant only were homogenised in the presence of acid before freezing. Grain samples were mixed thoroughly and then sub-sampled before freezing. Samples of straw were broken down with a cutting mill before freezing.

Storage stability of chlorothalonil has been discussed during the expert Peer Review Meeting (#164). It was concluded that based on the available data storage stability for chlorothalonil has been demonstrated in cereal straw up to 9 months and cereal grain 62 days. For metabolite SDS-3701 no storage stability data in cereal grain in available and the meeting concluded that extrapolation from other high starch matrix commodities is not acceptable. Hence a data gap is set. In cereal straw, SDS-3701 is stable up to 12 months.

~~Residues of chlorothalonil and R18221 are stable in acidified homogenised high water crops for at least 24 months, in samples of cereal grains for up to 24 months, and in straw for up to 12 months (chlorothalonil) and 27 months (R182281) (see CA 6.1); residues of R182281 are stable for 24 months in high water content commodities and high starch content commodities (covering cereal grains), and for 12 months in cereal straw, and, therefore, no degradation will have occurred between sampling and analysis.~~

Hence, ~~A~~all trials are considered acceptable only for residues of SDS-3701 in cereal straw. For chlorothalonil in cereal grain and straw and SDS-3701 in grain, storage stability still should be demonstarted.

In most trials, the second application took place at the exact growth stage of the cGAP (BBCH 59). In some trials, the second application took place at a somewhat later growth stage than the growth stage of the cGAP applied for. Therefore, these trials could be considered more critical. However, the formation of the edible part for cereals starts from stage BBCH 51 onwards. Furthermore, a maximum of 25% deviation of the growth stage is allowed, but this rule is difficult to apply on growth stages. Since the PHIs of the trials in which the second application was later than BBCH 59 are in the same range as the PHIs of the trials in which were the second application was exactly at BBCH 59, these trials are considered acceptable. Furthermore, 9 out of the 16 NEU trials and 8 out of 16 trials SEU have the second application somewhat later than BBCH 59. If the data sets are being compared (i.e. NEU trials with the second application at BBCH 59 versus NEU trials with the second application later than BBCH 59; and similar for the SEU trials), it can be observed that the data belong to similar populations, i.e. the MRL for chlorothalonil based on only NEU trials with the second application at BBCH 59 would be 0.07 mg/kg versus 0.08 mg/kg for the NEU trials with the second application somewhat later than BBCH 59; for SEU, the MRL would even be higher when only the trials with the second application at BBCH 59 would be taken into account (0.4 mg/kg versus 0.03 mg/kg for the trials with a somewhat later application). For SDS-3701 similar conclusions can be drawn: in grain, residues of SDS-3701 are anyway mostly below LOQ, independent of the timing of the second application. For straw, residues of chlorothalonil and SDS-3701 in some cases are higher when the second application is somewhat later than BBCH 59, while in other cases residues are lower. ~~Therefore, it can be concluded that the growth stage at the second~~

~~application does not seriously impact the residue levels, and all trials should be considered for MRL-setting. Importantly, taking the complete data set into account leads to a bigger data set and a more robust MRL.~~

During the expert Peer Review Meeting (#164) it was agreed that residue trials with second application at latest BBCH 61 (63) can be considered acceptable, while since the ears are already present at BBCG 69, second application on this growing stage is not acceptable.

In addition, the interval between the applications differs often significantly from the interval of the cGAP. ~~However, probably the interval has only minimal influence on the residue values, since the pre-harvest interval is large in comparison to the interval between applications.~~

In the expert Peer Review Meeting (# 164), it was concluded that a proposed interval is an important parameter and it should be considered as much comparable to the proposed cGAP. In the residue trials on barley intervals were reported from 7 to 35 days. It was discussed that the reported difference is significantly high and the acceptable interval should be up to ± 17 days and not much longer. The results will be selected accordingly (2nd application up to BBCH 61 and the interval between the applications up to ± 17 days).

The results of the residue trials for chlorothalonil and R182281 are presented in Table 7.3.2-4.

Where two or more values are available from duplicate analysis for the same trial following applications according to the GAP, the mean has been used. Where two or more values are available from duplicate sampling for the same trial following applications according to the GAP, the mean has been used.

It should be noted that the acceptability of the residue trials is pending the susbmission of the storage stability data of chlorothalonil and SDS-3701 in cereal grain. Moreover, in the trials, conjugates of chlorothalonil and SDS-3701 were not measured.

Table 7.3.2-4: Summary of residue data supporting the EU critical GAP for chlorothalonil on barley

GLP and Trial Details	Crop (Variety)	Country (Region)	Application Rate Product Code	Growth Stage and date at application	PHI (days)	Crop Part (Growth stage at sampling)	Residue Found (Uncorrected)		Recovery Data
							chlorothalonil mg/kg	R182281 (mg/kg)	
Northern Europe									
Report: S11-00522 Study: S11-00522 Trial: S11-00522-01 - Study to GLP - Study carried out in 2011	Barley (Waggon)	UNITED KINGDOM (Europe North) Arnold, Nottinghamshire	741 g a.s./ha 758 g a.s./ha (21-d interval) A14111B	BBCH 32; 24-05-2011 BBCH 59-63; 14-06-2011, interval 20 day	0 DAA1	Whole plant (BBCH 32)	30	0.42	**Chlorothalonil** Whole plant (immature): mean = 104% RSD = 10.9% (n = 6 in 0.01 – 50.0 mg/kg spiking range) Whole plant (silage): mean =100% RSD = 10.5% (n = 6 in 0.01 – 5.00 mg/kg spiking range) Whole plant (hay): mean = 104% RSD = 3.5% (n = 4 in 0.01 – 10.0 mg/kg spiking range) Grain: mean = 98% RSD = 4.7% (n = 6 in 0.01 -0.10 mg/kg spiking range) Straw: mean = 101% RSD = 8.4% (n = 6 in 0.01 –10.0 mg/kg spiking range) **R182281** Whole plant (immature): mean = 98% RSD = 13.1% (n = 4 in 0.01 – 1.00 mg/kg spiking range) Whole plant (silage): mean = 97% RSD = 14.1% (n = 4 in 0.01 – 1.00 mg/kg spiking range) Whole plant (hay): mean = 101% RSD = 13.0% (n = 6 in 0.01 – 5.00 mg/kg spiking range) Grain: mean = 100% RSD = 5.0% (n = 6 in 0.01- 0.10 mg/kg spiking range) Straw: mean = 87% RSD = 11.3% (n = 6 in 0.01 – 10.0 mg/kg spiking range)
					28/31†	Whole plant/ silage (BBCH 77-83)	5.0	0.08	
					28/37†	Whole plant/ hay (BBCH 77-83)	4.5	0.11	
					62	Grain (BBCH 89)	<0.01	<0.01	
					62	Straw (BBCH 89)	5.7	0.30	
Report: S11-00522 Study: S11-00522 Trial: S11-00522-02 - Study to GLP - Study carried out in 2011	Barley (Waggon)	UNITED KINGDOM (Europe North) Ibstock, Leicestershire	759 g a.s./ha 745 g a.s./ha (28-d interval) A14111B	BBCH 32; 13-05-2011 BBCH 59; 10-06-2011, interval > 17 day	0 DAA1	Whole plant (BBCH 32)	21	0.27	
					33/41†	Whole plant/silage (BBCH 77-83)	2.1	0.08	
					33/46†	Whole plant/ hay (BBCH 77-83)	1.5	0.06	
					73	Grain (BBCH 89)	0.02, 0.02, 0.01 Mean = 0.02	<0.01	
					73	Grain (BBCH 89)	0.01, 0.04 Mean = 0.03 Overall mean = 0.02	<0.01	
					73	Straw (BBCH 89)	2.2	0.09	
Report: S11-00522 Study: S11-00522	Barley (Highlight)	GERMANY (Europe North)	755 g a.s./ha 753 g a.s./ha	BBCH 31-32;	0 DAA1	Whole plant (BBCH 31-32)	45	0.47	

GLP and Trial Details	Crop (Variety)	Country (Region)	Application Rate Product Code	Growth Stage and date at application	PHI (days)	Crop Part (Growth stage at sampling)	Residue Found (Uncorrected)		Recovery Data
							chlorothalonil mg/kg	R182281 (mg/kg)	
Trial: S11-00522-03 - Study to GLP - Study carried out in 2011		Blumberg, Brandenburg	(29-d interval) A14111B	20-04-2011 BBCH 59; 19-05-2011, interval > 17 days	22/28†	Whole plant/silage (BBCH 81-83)	1.7	0.07	
					22/40†	Whole plant/ hay (BBCH 81-83)	0.83	0.06	
					49	Grain (BBCH 89)	<0.01	<0.01	
					49	Straw (BBCH 89)	1.7	0.09	
Report: S11-00522 Study: S11-00522 Trial: S11-00522-04 - Study to GLP - Study carried out in 2011	Barley (Sunshine)	FRANCE (Europe North) Les Rosiers sur Loire, Pays de la Loire	757 g a.s./ha 733 g a.s./ha (33-d interval) A14111B	BBCH 31-32; 04-05-2011 BBCH 59; 06-06-2011, interval > 17 days	0 DAA1	Whole plant (BBCH 31-32)	27	0.48	
					21/21†	Whole plant/silage (BBCH 75-77)	1.8	0.01	
					21/22†	Whole plant/ hay (BBCH 75-77)	2.3	0.03	
					49	Grain (BBCH 89)	<0.01	<0.01	
					49	Straw (BBCH 89)	0.72	0.03	
Report: S12-01274 Study: S12-01274 Trial: S12-01274-01 - Study to GLP - Study carried out in 2012	Barley (Westminster)	UNITED KINGDOM (Europe North) Nottinghamshire	750 g a.s./ha 727 g a.s./ha (29-d interval) A14111B	BBCH 30-32; 04-06-2012 BBCH 59; 03-07-2012, interval > 17 days	0 DAA1	Whole plant (BBCH 30-32)	33	0.43	**Chlorothalonil** Whole plant (immature): mean = 98% RSD = 7.7% (n = 5 in 0.01 – 50.0 mg/kg spiking range) Whole plant (silage): mean = 92% RSD = 6.5% (n = 4 in 0.01 – 5.00 mg/kg spiking range) Whole plant (hay): mean = 94% RSD = 3.4% (n = 4 in 0.01 – 5.00 mg/kg spiking range) Grain: mean = 86% RSD = 7.8% (n = 6 in 0.01 - 0.10 mg/kg spiking range)
					35/38†	Whole plant/silage (BBCH 85)	0.80	0.03	
					35/43†	Whole plant/ hay (BBCH 85)	0.83	0.06	
					64	Grain (BBCH 89)	0.04	<0.01	
					64	Straw (BBCH 89)	0.44	0.03	

GLP and Trial Details	Crop (Variety)	Country (Region)	Application Rate Product Code	Growth Stage and date at application	PHI (days)	Crop Part (Growth stage at sampling)	Residue Found (Uncorrected)		Recovery Data
							chlorothalonil mg/kg	R182281 (mg/kg)	
									Straw: mean = 92% RSD = 2.3% (n = 4 in 0.01 – 5.00 mg/kg spiking range) **R182281** Whole plant (immature): mean = 100% RSD = 2.9% (n = 4 in 0.01 – 1.00 mg/kg spiking range) Whole plant (silage): mean = 103% RSD = 1.7% (n = 4 in 0.01 – 1.00 mg/kg spiking range) Whole plant (hay): mean = 101% RSD = 11.4% (n = 4 in 0.01 – 1.00 mg/kg spiking range) Grain: mean = 107% RSD = 5.4% (n = 6 in 0.01- 0.10 mg/kg spiking range) Straw: mean = 100% RSD = 4.7% (n = 4 in 0.01 – 1.00 mg/kg spiking range)
Report: S12-01274 Study: S12-01274 Trial: S12-01274-02 - Study to GLP - Study carried out in 2012	Barley (Quench)	GERMANY (Europe North) Baden Württemberg	730 g a.s./ha 771 g a.s./ha (35-d interval) A14111B	BBCH 30-31; 17-05-2012 BBCH 59; 21-06-2012, interval > 17 days	0 DAA1	Whole plant (BBCH 30-31)	17	0.09	
					20/22†	Whole plant/silage (BBCH 79)	0.46	0.02	
					20/25†	Whole plant/hay (BBCH 79)	0.59	0.03	
					40	Grain (BBCH 89)	0.01	<0.01	
					40	Straw (BBCH 89)	0.68	0.10	
Report: S12-01274 Study: S12-01274 Trial: S12-01274-03 - Study to GLP - Study carried out in 2012	Barley (Lomerit)	GERMANY (Europe North) Brandenburg	808 g a.s./ha 778 g a.s./ha (15-d interval) A14111B	BBCH 32; 25-04-2012 BBCH 59; 10-05-2012, interval 15 days	0 DAA1	Whole plant (BBCH 32)	7.9	0.10	
					29/34†	Whole plant/silage (BBCH 77-83)	4.5	0.03	
					29/48†	Whole plant/hay (BBCH 77-83)	6.3	0.13	
					62	Grain (BBCH 89)	0.01	<0.01	
					62	Straw (BBCH 89)	2.8	0.11	
Report: S12-01274 Study: S12-01274 Trial: S12-01274-04 - Study to GLP - Study carried out in 2012	Barley (Frontier)	POLAND (Europe North) woj. Wielkopolskie	753 g a.s./ha 723 g a.s./ha (18-d interval) A14111B	BBCH 30-32; 28-05-2012 BBCH 59; 15-06-2012, interval 17 days	0 DAA1	Whole plant (BBCH 30-32)	23	0.40	
					17/18†	Whole plant/silage (BBCH 75-77)	1.3	<0.01	
					17/27†	Whole plant/ hay (BBCH 75-77)	1.4	0.04	
					47	Grain	<0.01	<0.01	

GLP and Trial Details	Crop (Variety)	Country (Region)	Application Rate Product Code	Growth Stage and date at application	PHI (days)	Crop Part (Growth stage at sampling)	Residue Found (Uncorrected) chlorothalonil mg/kg	 R182281 (mg/kg)	Recovery Data
						(BBCH 89)			
					47	Straw (BBCH 89)	0.45	0.02	
Report: RAU-020-13 Study: RAU-020-13 Trial: F/CH13/BA03 - Study to GLP - Study carried out in 2013	Barley (Cervoise)	FRANCE (Europe North) Blanzy la Salonnaise	722 g a.s./ha 722 g a.s./ha (27-d interval) 500 g/L SC	BBCH 32; 30-04-2013 BBCH 61; 27-05-2013, interval > 17 days	52 52	Grain (BBCH 89)	0.03	<0.02	**Chlorothalonil** Grain: mean = 97% RSD = NA (n = 2 in 0.01 -0.10 mg/kg spiking range) Straw: mean = 100% RSD = 4.7% (n = 4 in 0.05 –2.0 mg/kg spiking range) **R182281** Grain: mean = 98% RSD = 3.4% (n = 3 in 0.02- 0.20 mg/kg spiking range) Straw: mean = 105% RSD = NA (n = 2 in 0.02– 0.20 mg/kg spiking range)
						Straw (BBCH 89)	0.74	0.06	
Report: RAU-020-13 Study: RAU-020-13 Trial: B/CH13/BA04 - Study to GLP - Study carried out in 2013	Barley (Meridian)	BELGIUM (Europe North) Sombreffe	743 g a.s./ha 758 g a.s./ha (35-d interval) 500 g/L SC	BBCH 31; 29-04-2013 BBCH 61; 03-06-2013, interval > 17 days	44 44	Grain (BBCH 89)	0.04	<0.02	
						Straw (BBCH 89)	1.0	<0.02	
Report: BIU-017-14 Study: BIU-017-14 Trial: F/CH14/BA07 - Study to GLP - Study carried out in 2014	Barley (Cervoise)	FRANCE (Europe North) Menil Lepinois	791 g a.s./ha 804 g a.s./ha (20-d interval) 500 g/L SC	BBCH 63; 05-05-2014 BBCH 69; 25-05-2014, interval 19 days	0	Whole plant (BBCH 69)	18	0.49	**Chlorothalonil** Grain: mean = 91% RSD = 7.6% (n = 4 in 0.01 -0.10 mg/kg spiking range) Hay: mean = 94% RSD = NA (n = 2 in 0.05 –20 mg/kg spiking range) Straw: mean = 101% RSD = 8.4%
					18/26†	Whole plant/silage (BBCH 77)	0.38	0.36	
					18/33†	Whole plant/hay (BBCH 77)	5.1	0.50	
					42	Grain (BBCH 89)	0.02	<0.02	
					42	Straw	2.3	0.61	

GLP and Trial Details	Crop (Variety)	Country (Region)	Application Rate Product Code	Growth Stage and date at application	PHI (days)	Crop Part (Growth stage at sampling)	Residue Found (Uncorrected)		Recovery Data
							chlorothalonil mg/kg	R182281 (mg/kg)	
						(BBCH 89)			(n = 4 in 0.05 –20 mg/kg spiking range) **R182281** Grain: mean = 87% RSD = NA (n = 2 in 0.02- 0.20 mg/kg spiking range) Whole plant : mean = 88% RSD = NA (n = 2 in 0.02 –1.0 mg/kg spiking range) Straw: mean = 101% RSD = NA (n = 2 in 0.02– 2.0 mg/kg spiking range)
Report: BIU-017-14 Study: BIU-017-14 Trial: F/CH14/BA08 - Study to GLP - Study carried out in 2014	Barley (Esterel)	FRANCE (Europe North) Blanzy la Salonnaise	797 g a.s./ha 797 g a.s./ha (10-d interval) 500 g/L SC	BBCH 61; 30-04-2014 BBCH 69; 10-05-2014, interval 9 days	47	Grain (BBCH 89)	0.04	<0.02	
					47	Straw (BBCH 89)	1.2	0.38	
Report: BIU-017-14 Study: BIU-017-14 Trial: F/CH14/BA09 - Study to GLP - Study carried out in 2014	Barley (Esterel)	FRANCE (Europe North) Inchy en Artois	773 g a.s./ha 794 g a.s./ha (9-d interval) 500 g/L SC	BBCH 57; 05-05-2014 BBCH 69; 14-05-2014, interval 8 days	49	Grain (BBCH 89)	0.04	<0.02	
					49	Straw (BBCH 89)	1.3	0.07	
Report: BIU-017-14 Study: BIU-017-14 Trial: P/CH14/BA10 - Study to GLP - Study carried out in 2014	Barley (Lomerit)	POLAND (Europe North) Michalow	777 g a.s./ha 769 g a.s./ha (7-d interval) 500 g/L SC	BBCH 55; 05-05-2014 BBCH 61; 12-05-2014, interval 6 days	53	Grain (BBCH 89)	<0.01	<0.02	
					53	Straw (BBCH 89)	4.9	1.1	
Report: BIU-017-14 Study: BIU-017-14 Trial: P/CH14/BA11 - Study to GLP - Study carried out in 2014	Barley (Maybrit)	POLAND (Europe North) Popowo Koscielne	752 g a.s./ha 760 g a.s./ha (8-d interval) 500 g/L SC	BBCH 55; 05-05-2014 BBCH 61; 13-05-2014, interval 7 days	58	Grain (BBCH 89)	0.03	<0.02	
					58	Straw (BBCH 89)	2.0	0.19	
Report: BIU-017-14 Study: BIU-017-14 Trial: P/CH14/BA12 - Study to GLP - Study carried out in 2014	Barley (Souleika)	POLAND (Europe North) Zduny	750 g a.s./ha 741 g a.s./ha (14-d interval) 500 g/L SC	BBCH 39; 30-04-2014 BBCH 61; 14-05-2014, interval 13 days	54	Grain (BBCH 89)	<0.01	<0.02	
					54	Straw (BBCH 89)	2.3	0.23	

GLP and Trial Details	Crop (Variety)	Country (Region)	Application Rate Product Code	Growth Stage and date at application	PHI (days)	Crop Part (Growth stage at sampling)	Residue Found (Uncorrected) chlorothalonil mg/kg	 R182281 (mg/kg)	Recovery Data
Southern Europe									
Report: S11-00523 Study: S11-00523 Trial: S11-00523-02 - Study to GLP - Study carried out in 2011	Barley (Prestige)	SPAIN (Europe South) Fonfria, Teruel, Aragon	695 g a.s./ha 742 g a.s./ha (24-d interval) A14111B	BBCH 30-32; 06-05-2011 BBCH 59; 30-05-2011, interval > 17 days	0 DAA1	Whole plant# (BBCH 30-32)	32	0.51	**Chlorothalonil** Whole plant (immature): mean = 98% RSD = 13% (n = 5 in 0.01 – 50 mg/kg spiking range) Whole plant (silage): mean = 105% RSD = 9.6% (n = 5 in 0.01 – 10 mg/kg spiking range) Whole plant (hay): mean = 97% RSD = 9.0% (n = 4 in 0.01 – 5.0 mg/kg spiking range) Grain: mean = 95% RSD = 3.6% (n =4 in 0.01 -0.10 mg/kg spiking range) Straw: mean = 101% RSD = 12% (n = 7 in 0.01 – 50 mg/kg spiking range) **R182281** Whole plant (immature): mean = 93% RSD = 6.5% (n = 4 in 0.01 – 1.0 mg/kg spiking range) Whole plant (silage): mean = 103% RSD = 5.1% (n = 4 in 0.01 – 1.0 mg/kg spiking range) Whole plant (hay): mean = 97% RSD = 1.7% (n = 4 in 0.01 – 1.0 mg/kg spiking range) Grain: mean = 107% RSD = 5.3% (n = 4 in 0.01- 0.10 mg/kg spiking range) Straw: mean = 99% RSD = 7.7% (n = 7 in 0.01 – 10 mg/kg spiking range)
					22/25†	Whole plant/silage# (BBCH 75-85)	0.58	<0.01	
					22/28†	Whole plant/hay# (BBCH 75-85)	0.44	0.01	
					58	Grain# (BBCH 89)	<0.01	<0.01	
					58	Straw# (BBCH 89)	0.20	<0.01	
Report: S11-00523 Study: S11-00523 Trial: S11-00523-03 - Study to GLP - Study carried out in 2011	Barley (Amorosa)	ITALY (Europe South) Idice, Bologna	758 g a.s./ha 751 g a.s./ha (27-d interval) A14111B	BBCH 32; 13-04-2011 BBCH 59; 10-05-2011, interval > 17 days	0 DAA1	Whole plant# (BBCH 32)	22	0.31	
					27/36†	Whole plant/silage# (BBCH 77)	0.07	<0.01	
					27/39†	Whole plant/hay# (BBCH 77)	0.04	<0.01	
					41	Grain# (BBCH 89)	<0.01	<0.01	
					41	Straw# (BBCH 89)	0.15	0.02	
Report: S11-00523 Study: S11-00523 Trial: S11-00523-04 - Study to GLP - Study carried out in 2011	Barley (Atomo)	ITALY (Europe South) Conselice, Ravenna	727 g a.s./ha 806 g a.s./ha (20-d interval) A14111B	BBCH 30-32; 19-04-2011 BBCH 59; 09-05-2011, interval > 17 days	0 DAA1	Whole plant# (BBCH 30-32)	18	0.27	
					23/30†	Whole plant/silage# (BBCH 77-83)	7.3	0.10	
					23/36†	Whole plant/hay# (BBCH 77-83)	4.8	0.15	
					40	Grain# (BBCH 89)	<0.01	<0.01	

GLP and Trial Details	Crop (Variety)	Country (Region)	Application Rate Product Code	Growth Stage and date at application	PHI (days)	Crop Part (Growth stage at sampling)	Residue Found (Uncorrected)		Recovery Data
							chlorothalonil mg/kg	R182281 (mg/kg)	
					40	Straw[#] (BBCH 89)	0.64	0.12	
Report: S12-01275 Study: S12-01275 Trial: S12-01275-01 - Study to GLP - Study carried out in 2012	Barley (Azurel)	FRANCE (Europe South) Pyrénées-Orientales	764 g a.s./ha 725 g a.s./ha (29-d interval) A14111B	BBCH 32; 10-04-2012 BBCH 59; 09-05-2012, interval > 17 days	0 DAA1	Whole plant (BBCH 32)	18	0.19	**Chlorothalonil** Whole plant (immature): mean = 93% RSD = 8.5% (n = 7 in 0.01 – 50 mg/kg spiking range) Whole plant (silage): mean = 92% RSD = 5.8% (n = 4 in 0.01 – 5.0 mg/kg spiking range) Whole plant (hay): mean = 95% RSD = 5.2% (n = 4 in 0.01 – 10 mg/kg spiking range) Grain: mean = 92% RSD = 10.3% (n = 6 in 0.01 - 5.0 mg/kg spiking range) Straw: mean = 90% RSD = 5.3% (n = 4 in 0.01 – 5.0 mg/kg spiking range) **R182281** Whole plant (immature): mean = 103% RSD = 5.6% (n = 3 in 0.01 – 1.0 mg/kg spiking range) Whole plant (silage): mean = 93% RSD = 5.7% (n = 4 in 0.01 – 5.0 mg/kg spiking range) Whole plant (hay): mean = 95% RSD = 16.5% (n = 5 in 0.01 – 10 mg/kg spiking range) Grain: mean = 100% RSD = 5.1% (n = 6 in 0.01- 1.0 mg/kg spiking range) Straw: mean = 102% RSD = 5.2% (n = 4 in 0.01 – 1.0 mg/kg spiking range)
					22/22†	Whole plant/silage (BBCH 75-85)	2.0	0.02	
					22/30†	Whole plant/hay (BBCH 75-85)	3.9	0.11	
					37	Grain (BBCH 89)	0.14, 0.22, 0.20 Mean = 0.19	<0.01, <0.01, 0.01 Mean = 0.01	
					37	Straw (BBCH 89)	3.1	0.27	
Report: S12-01275 Study: S12-01275 Trial: S12-01275-02 - Study to GLP - Study carried out in 2012	Barley (Campagnill)	FRANCE (Europe South) Ain	704 g a.s./ha 750 g a.s./ha (34-d interval) A14111B	BBCH 31-32; 06-04-2012 BBCH 59; 10-05-2012, interval > 17 days	0 DAA1	Whole plant (BBCH 31-32)	18	0.17	
					20/21†	Whole plant/silage (BBCH 75-85)	3.6	0.04	
					20/28†	Whole plant/hay (BBCH 75-85)	2.1	0.37	
					46	Grain (BBCH 89)	<0.01	<0.01	
					46	Straw (BBCH 89)	0.45	0.03	
Report: S12-01275 Study: S12-01275 Trial: S12-01275-03 - Study to GLP - Study carried out in 2012	Barley (Atomo)	ITALY (Europe South) Bologna	764 g a.s./ha 725 g a.s./ha (28-d interval) A14111B	BBCH 30-32; 02-04-2012 BBCH 59; 30-04-2012, interval > 17	0 DAA1	Whole plant (BBCH 30-32)	16	0.24	
					25/30†	Whole plant/silage (BBCH 75-85)	4.2	0.05	
					25/32†	Whole plant/hay (BBCH 75-85)	5.2	0.07	

GLP and Trial Details	Crop (Variety)	Country (Region)	Application Rate Product Code	Growth Stage and date at application	PHI (days)	Crop Part (Growth stage at sampling)	Residue Found (Uncorrected)		Recovery Data
							chlorothalonil mg/kg	R182281 (mg/kg)	
				days	45	Grain (BBCH 89)	0.12, 0.16, 0.26 Mean = 0.18	<0.01, <0.01, <0.01 Mean = <0.01	
					45	Straw (BBCH 89)	1.8	0.06	
Report: S12-01275 Study: S12-01275 Trial: S12-01275-04 - Study to GLP - Study carried out in 2012	Barley (Volley)	SPAIN (Europe South) Zaragoza	682 g a.s./ha 742 g a.s./ha (18-d interval) A14111B	BBCH 30-32; 07-05-2012 BBCH 59; 25-05-2012, interval 17 days	0 DAA1	Whole plant (BBCH 30-32)	24	0.39	
					18/19†	Whole plant/silage (BBCH 75-85)	1.7	0.05	
					18/24†	Whole plant/hay (BBCH 75-85)	1.5	0.06	
					34	Grain (BBCH 89)	<0.01	<0.01	
					34	Straw (BBCH 89)	0.67	0.07	
Report: S13-01041 Study: S13-01041 Trial: S13-01041-01 - Study to GLP - Study carried out in 2013	Barley (Prestige)	FRANCE (Europe South) Midi-Pyrénées	745 g a.s./ha 756 g a.s./ha (28-d interval) A14111B	BBCH 32; 07-05-2013 BBCH 59; 04-06-2013, interval > 17 days	0 DAA1	Whole plant (BBCH 32)	26	0.07	**Chlorothalonil** Whole plant (immature): mean = 87% RSD = 2.4% (n = 3 in 0.01 – 50 mg/kg spiking range) Grain: mean = 87% RSD = NA (n =2 in 0.01 -0.1 mg/kg spiking range) Straw: mean = 82% RSD = NA (n = 2 in 0.01 – 1.0 mg/kg spiking range) **R182281** Whole plant (immature): mean = 84% RSD = NA (n = 2 in 0.01 – 0.1 mg/kg spiking range) Grain: mean = 104% RSD = NA (n = 2 in 0.01- 0.1 mg/kg spiking range) Straw: mean = 98% RSD = NA (n = 2
					50	Grain (BBCH 89)	<0.01	<0.01	
					50	Straw (BBCH 89)	0.30	0.19	

GLP and Trial Details	Crop (Variety)	Country (Region)	Application Rate Product Code	Growth Stage and date at application	PHI (days)	Crop Part (Growth stage at sampling)	Residue Found (Uncorrected)		Recovery Data
							chlorothalonil mg/kg	R182281 (mg/kg)	
									in 0.01 – 0.1 mg/kg spiking range)
Report: RAU-018-13 Study: RAU-018-13 Trial: I/CH13/BA01 - Study to GLP - Study carried out in 2013	Barley (Arda)	ITALY (Europe South) Monticelli Pavese	766 g a.s./ha 757 g a.s./ha (20-d interval) 500 g/L SC	BBCH 33; 17-04-2013 BBCH 61-65; 07-05-2013, interval 19 days	50	Grain (BBCH 89)	<0.01	<0.02	**Chlorothalonil** Grain: mean = 95% RSD = NA (n = 2 in 0.01 -0.10 mg/kg spiking range) Straw: mean = 101% RSD = NA (n = 2 in 0.05 –0.5 mg/kg spiking range) **R182281** Grain: mean = 90% RSD =NA (n = 2 in 0.02- 0.20 mg/kg spiking range) Straw: mean = 98% RSD = NA (n = 2 in 0.02– 0.20 mg/kg spiking range)
					50	Straw (BBCH 89)	0.25	0.05	
Report: RAU-018-13 Study: RAU-018-13 Trial: F/CH13/BA02 - Study to GLP - Study carried out in 2013	Barley (Sebastian)	FRANCE (Europe South) Grenade sur Garonne	753 g a.s./ha 773 g a.s./ha (34-d interval) 500 g/L SC	BBCH 31; 02-05-2013 BBCH 61; 05-06-2013, interval > 17 days	39	Grain (BBCH 89)	<0.01	<0.02	
					39	Straw (BBCH 89)	0.34	0.05	
Report: BIU-016-14 Study: BIU-016-14 Trial: I/CH14/BA01 - Study to GLP - Study carried out in 2014	Barley (Arda)	ITALY (Europe South) Monticelli Pavese	750 g a.s./ha 741 g a.s./ha (21-d interval) 500 g/L SC	BBCH 47-49; 17-04-2014 BBCH 65-69; 08-05-2014, interval 20 days	42	Grain (BBCH 89)	<0.01	<0.02	**Chlorothalonil** Grain: mean = 100% RSD = 20% (n = 6 in 0.01 -0.10 mg/kg spiking range) Whole plant: mean = 100% RSD = 12% (n = 6 in 0.05 –50 mg/kg spiking range) Straw: mean = 87% RSD = 7.1%
					42	Straw (BBCH 89)	0.15	0.10	
Report: BIU-016-14 Study: BIU-016-14	Barley (Etincel)	ITALY (Europe South)	756 g a.s./ha 753 g a.s./ha	BBCH 33; 18-04-2014	45	Grain (BBCH 89)	<0.01	<0.02	

GLP and Trial Details	Crop (Variety)	Country (Region)	Application Rate Product Code	Growth Stage and date at application	PHI (days)	Crop Part (Growth stage at sampling)	Residue Found (Uncorrected)		Recovery Data
							chlorothalonil mg/kg	R182281 (mg/kg)	
Trial: I/CH14/BA02 - Study to GLP - Study carried out in 2014		Isola Sant' Antonio	(20-d interval) 500 g/L SC	BBCH 61; 08-05-2014, interval 19 days	45	Straw (BBCH 89)	0.06	0.03	(n = 6 in 0.05 –1.0 mg/kg spiking range) **R182281** Grain: mean = 82% RSD = 11% (n = 6 in 0.02- 0.20 mg/kg spiking range) Whole plant : mean = 102% RSD = 8.9% (n = 6 in 0.02 –1.0 mg/kg spiking range) Straw: mean = 95% RSD = 11% (n = 6 in 0.02– 2.0 mg/kg spiking range)
Report: BIU-016-14 Study: BIU-016-14 Trial: I/CH14/BA03 - Study to GLP - Study carried out in 2014	Barley (Cometa)	ITALY (Europe South) Soresina	745 g a.s./ha 755 g a.s./ha (8-d interval) 500 g/L SC	BBCH 54; 15-04-2014 BBCH 61; 23-04-2014, interval 8 days	49	Grain (BBCH 89)	<0.01	<0.02	
					49	Straw (BBCH 89)	0.06	0.06	
Report: BIU-016-14 Study: BIU-016-14 Trial: I/CH14/BA04 - Study to GLP - Study carried out in 2014	Barley (Tuareg)	ITALY (Europe South) San Bassano	734 g a.s./ha 775 g a.s./ha (8-d interval) 500 g/L SC	BBCH 56; 15-04-2014 BBCH 61; 23-04-2015, interval 8 days	0	Whole plant (BBCH 61)	27	0.54	
					16	Whole plant/silage (BBCH 75)	4.2	0.10	
					17	Whole plant/hay (BBCH 75)	4.7	0.14	
					49	Grain (BBCH 89)	<0.01	<0.02	
					49	Straw (BBCH 89)	0.43	0.11	
Report: BIU-016-14 Study: BIU-016-14 Trial: I/CH14/BA05 - Study to GLP - Study carried out in 2014	Barley (Cometa)	ITALY (Europe South) Cappella Cantone	755 g a.s./ha 755 g a.s./ha (8-d interval) 500 g/L SC	BBCH 54; 15-04-2014 BBCH 61; 23-04-2014, interval 8 days	49	Grain (BBCH 89)	<0.01	<0.02	
					49	Straw (BBCH 89)	0.48	0.13	
Report: BIU-016-14 Study: BIU-016-14 Trial: F/CH14/BA06 - Study to GLP - Study carried out in 2014	Barley (Sebastian)	FRANCE (Europe South) Grenade sur Garonne	725 g a.s./ha 750 g a.s./ha (8-d interval) 500 g/L SC	BBCH 59; 30-04-2014 BBCH 61; 08-05-2014, interval 7	47	Grain (BBCH 89)	0.02	<0.02	
					47	Straw (BBCH 89)	0.66	0.13	

GLP and Trial Details	Crop (Variety)	Country (Region)	Application Rate Product Code	Growth Stage and date at application	PHI (days)	Crop Part (Growth stage at sampling)	Residue Found (Uncorrected)		Recovery Data
							chlorothalonil mg/kg	R182281 (mg/kg)	
				days					
Residues in all untreated samples were less than the LOQ. DAA1 = days after first application. All other PHI values are days after the second application. †Silage and hay samples were cut at various growth stages between BBCH 75 and 85 then left in the field to dry until the moisture content reached the typical moisture content of silage and hay respectively. For these crops there are therefore two PHI values reported: the PHI when the crop was cut and the PHI when the sample was taken from the field (i.e. including drying). NA = not applicable # No storage stability study available to cover extract storage for 56 days, however, levels in whole plant are not significantly lower, but comparable to other studies with shorter extract storage.									

B.7.3.3 Wheat

Chlorothalonil is proposed for use on wheat in NEU and SEU according to the following GAP:

2x 750 g/ha, interval 14 days, BBCH 30-69 (no PHI).

The representative use finally supported in the original peer review was an outdoor foliar application on wheat. The cGAP contained 2 applications at 1.0 kg/ha with last application at BBCH 51. This GAP for wheat during the initial peer review was more critical regarding dose rate, and less critical regarding application timing (BBCH 59) than the GAP applied for within the framework of the renewal. Therefore, none of the trials have been copied into this RAR.

For the renewal of chlorothalonil, additional reports containing supervised residue trials have been submitted. The residue reports supporting the proposed EU critical GAP for chlorothalonil on wheat are referenced in Table 7.3.3-1 and the data are presented in Table 7.3.3-4.

Previous evaluation	Submitted for the purpose of renewal
RMS remark	~~Out of the 32 trials in total, for NEU 16 acceptable residue levels are available for grain and 6 residue levels for straw, while for SEU 16 acceptable residue levels are available for grain and 8 residue levels for straw~~ Acceptablity of the trial is pending submission of storage stability data covering the storage time of chlorothalonil and metabolite SDS-3701 in cereal grain. It should be noted that conjugated of chlorothalonil and SDS-3701 were not measured in the trials.

Table 7.3.3-1: Report references for trials supporting the proposed EU critical GAP for chlorothalonil on wheat

Annex Pt.	Number.	Author/s	Issue Year	Report Title
K-CA 6.3.3/01	(1 of 6)	S Lakaschus A Gizler	~~2014~~ 2017	Chlorothalonil – residue study on wheat in northern France, Germany, Poland and the United Kingdom in 2012. Eurofins Agroscience Services Chem GmbH, Grossmoorbogen 25, D21079 Hamburg. Syngenta File No. A14111B_11147, Report No. S12-01272[a]
K-CA 6.3.3/02	(2 of 6)	S Lakaschus A Gizler	~~2014a~~ 2017	Chlorothalonil – residue study on wheat in southern France, Italy and Spain in 2012. Eurofins Agroscience Services Chem GmbH, Grossmoorbogen 25, D21079 Hamburg. Syngenta File No. A14111B_11149, Report No. S12-01273[a]
K-CA 6.3.3/03	(3 of 6)	A Sala	2014b	Determination of chlorothalonil and its metabolite SDS3701 residues in raw agricultural commodity winter wheat (grain, straw) following two applications of chlorothalonil 500 SC, 4 trials, northern Europe, year 2013. Research Centre Biospheres, Via Vittorio Veneto 81, 26857 Salerano sul Lambro, Italy. Syngenta File No R044686_11186, Report No. RAU-019-13
K-CA 6.3.3/04	(4 of 6)	A Sala	2014c	Determination of chlorothalonil and its metabolite SDS3701 residues in raw agricultural commodity winter wheat (grain, straw) following two applications of chlorothalonil 500 SC, 4 trials, southern Europe, year 2013. Research Centre Biospheres, Via Vittorio Veneto 81, 26857 Salerano sul Lambro, Italy. Syngenta File No R044686_11188. Report No. RAU-017-13

Annex Pt.	Number.	Author/s	Issue Year	Report Title
K-CA 6.3.3/05	(5 of 6)	F Mazzi	2014b	Determination of chlorothalonil and its metabolite SDS3701 residues in raw agricultural commodity winter wheat (plant, silage, hay, grain, straw) following two applications of Clortosip 500 SC (northern Europe – 4 trials year 2014). . Research Centre Biospheres, Via Vittorio Veneto 81, 26857 Salerano sul Lambro, Italy. Syngenta File No. R044686_11187, Report No. BIU-015-14
K-CA 6.3.3/06	(6 of 6)	F Mazzi	2014c	Determination of chlorothalonil and its metabolite SDS3701 residues in raw agricultural commodity winter wheat (plant, silage, hay, grain, straw) following two applications of chlorothalonil 500 SC (south Europe – 4 trials year 2014). Research Centre Biospheres, Via Vittorio Veneto 81, 26857 Salerano sul Lambro, Italy. Syngenta File No. R044686_11185, Report No. BIU-014-14

a The final reports for studies S12-01272 and S12-01273 have been amended to include the storage periods for each of the wheat samples.

Guidelines

The studies meet the requirements of the Commission of the European Communities: General Recommendations for the Design, Preparation and Realization of Residue Trials (7029/V1/95 rev. 5, 22/7/1997), and are designed to comply with Regulation (EC) 1107/2009.

GLP

All trials (field and analytical phases) were carried out in compliance with the principles of Good Laboratory Practice.

Materials and Methods

Thirty-two supervised residue trials were conducted on wheat in 2012, 2013 and 2014, in northern or southern Europe. A summary of the trials conducted is presented in Table 7.3.3-2.

Table 7.3.3-2: Summary of chlorothalonil residue trials on wheat

Country	2012	2013	2014
Northern Europe			
France (north)	2 Harvest	1 Harvest	3 Harvest
Germany	3 Harvest	2 Harvest	-
Poland	1 Harvest	-	1 Harvest
United Kingdom	2 Harvest	-	-
Belgium	-	1 Harvest	-
Southern Europe			
France (south)	3 Harvest	1 Harvest	1 Harvest
Spain	3 Harvest	-	-
Italy	2 Harvest	3 Harvest	3 Harvest

Wheat is a major crop in northern and southern Europe and therefore generally requires eight trials in each residue region.

Treatments with chlorothalonil were conducted as post emergence (BBCH 30-32 [up to BBCH 41 in three trials] and BBCH 69 [up to 70/73 in two trials]) spray applications utilising the formulations as detailed in Table 7.3.3-3 at a nominal application rate of 750 g a.s./ha (actual rates 681-791 g a.s./ha). The water volumes during application ranged from 100 to 420 L/ha. Trials were widespread across the northern and southern EU regions.

Table 7.3.3-3: Summary of chlorothalonil formulation used in the presented trials

Product code	Formulation type	Composition		
		2012	2013	2014
A14111B	SC	384 g/L chlorothalonil 74.7 g/L azoxystrobin	-	-
Chlorothalonil 500 SC	SC	-	502 g/L chlorothalonil (batch O232)	507 g/L chlorothalonil (batch PN1911)

Samples of various parts of mature and immature wheat plants were taken and analysed for residues of chlorothalonil and R182281 using either analytical method GRM005.01A with an LOQ of 0.01 mg/kg for both compounds in all commodities analysed, or by analytical methods described in study BIU-016-14, with an LOQ of 0.01 mg/kg in grain and 0.05 mg/kg for other commodities for chlorothalonil and 0.02 mg/kg for all commodities for R182281. Method GRM005.01A involved extraction with acetone/5M sulphuric acid (95:5 v/v), dilution with water followed by SPE clean up for chlorothalonil or taking up in acetonitrile:water (50/50 v/v) for R182281. Subsequently, analysis was performed by gas chromatography with mass selective detection (GC-MSD) for chlorothalonil using two ion masses and LC-MS/MS for R182281 using 2 mass transitions. Matrix-matched standards were used in study 6.3.3/01 and 6.3.3./02. The analytical method (sometimes indicated by RAU-076-00 or RAU-076-01) used in study RAU-019-13, RAU-017-13, BIU-015-14 and BIU-014-14 involved extraction with acidified ethyl acetate followed by analysis using a gas chromatograph equipped with µ-ECD detector for chlorothalonil. For R182281 the analytical method consisted of extraction with methanol and analysis with LC-MS/MS. Full method descriptions and validation data are presented in B.5.2.1 (K-CA 4.2/01-K-CA 4.2/07), B.5.1.2.5 (K-CA 4.1.2/32) and B.5.1.2.5 (K-CA 4.1.2/33). Procedural recovery data are presented with the results of the residues trials in Table 7.3.3-4.

Samples were stored up to a maximum of 16 months in report S12-01272 and S12-01273, while in the other reports the maximum storage period was 6 months from sampling to extraction. Samples of whole plant only were homogenised in the presence of acid before freezing. Grain samples were mixed thoroughly and then sub-sampled before freezing. Samples of straw were broken down with a cutting mill before freezing. ~~Residues of chlorothalonil and R182281 are stable in acidified homogenised high water content crops for at least 24 months, in samples of high starch content commodities (covering cereal grains) for up to 24 months, and in straw for up to 12 months (chlorothalonil) and 27 months (R182281) (see CA 6.1). Therefore, no degradation will have occurred between sampling and analysis, except for chlorothalonil in the straw samples. The demonstrated stability for chlorothalonil and R182281 in straw is 12 months, while in report S12-01272 and S12-01273 the straw samples were stored up to a maximum of~~

~~16 months~~. ~~Results of these samples are, therefore, not acceptable.~~ However, during the peer review process, the applicant provided specific storage period data for these straw samples (the final reports for studies S12-01272 and S12-01273 have been amended). In study report S12-01272, the longest storage period for the straw samples was 7 months, while in study report S12-01273 the longest storage period for the straw samples was close to 10 months (295 days). Therefore, all results of these straw samples are now considered acceptable.
During the expert Peer Review Meeting (# 164) storage stability of chlorothalonil and metabolite SDS-3701 has been discussed. The stability in cereal grain has been demonstrated up to 62 days for chlorothalonil and no data is available for SDS-3701. In cereal straw chlorothalonil was stable up to 9 months and SDS-3701 up to 12 months.

No weather data are reported for G/CH13/WW07 and B/CH13/WW08. The applicant is requested to provide these data. Since the residue data of these trials are in the same range as the data from the other trials, these trials are included in the evaluation.

All trials are acceptable regarding application rate and timing, except for trial F/CH14/WW07 in which the second application was at BBCH 62, which is not according to the cGAP. This trial could be considered less critical. However, the formation of the edible part for cereals starts from stage BBCH 51 onwards. Furthermore, a maximum of 25% deviation of the growth stage is allowed, but this rule is difficult to apply on growth stages. Since the PHI of this trial is in the same range as the PHI of the trials in which were the second application was exactly at BBCH 69, this trial is considered acceptable. Much longer intervals between applications have been used than the interval of the cGAP. ~~However, it is expected that the interval has only minimal influence on the residue values as the pre-harvest interval is large in comparison to the interval between applications. Therefore, trials with an interval that deviates are considered acceptable.~~ In the expert Peer Review Meeting (# 164), it was concluded that the proposed interval is an important parameter and it should be considered as much comparable to the proposed cGAP. In the residue trials on barley intervals were reported from 19 to 57 days. It was discussed that the reported difference is significantly high and the acceptable interval should be up to ± 17 days and not much longer. The results will be selected accordingly.
~~In total, for NEU 16 acceptable residue levels are available for grain and 14 6 residue levels for straw, while in SEU 16 acceptable residue levels are available for grain and 16 8 residue levels for straw.~~
In total two trials were selected with interval 18-19 days, which could be still considered acceptable from the Southern Europe. No trials in Northern Europe were performed with the interval between 14-17 days. However, the acceptability of the trial is pending the susbmission of the storage stability data of chlorothalonil and SDS-3701 in cereal grain. Moreover, in the trials, conjugates of chlorothalonil and SDS-3701 were not measured.

The results of the residue trials for chlorothalonil and R182281 are presented in Table 7.3.3-4.

Table 7.3.3-4: Summary of residue data supporting the EU critical GAP for chlorothalonil on wheat

GLP and Trial Details	Crop (Variety)	Country (Region)	Application Rate Product Code	Growth Stage at application	PHI (days)	Crop Part (Growth stage at sampling)	Residue Found (Uncorrected)		Recovery Data
							chlorothalonil mg/kg	R182281 (mg/kg)	
Northern Europe									
Report: S12-01272 Study: S12-01272 Trial: S12-01272-01 - Study to GLP - Study carried out in 2012	Wheat (Granary)	UNITED KINGDOM (Europe North) Yorkshire	750 g a.s./ha 738 g a.s./ha (46-d interval) A14111B	BBCH 30-32; 28-05-2012 BBCH 69; 13-07-2012, interval >17 days	0 DAA1	Whole plant (BBCH 30-32)	19	0.38	**Chlorothalonil** Whole plant (immature): mean = 83% RSD = 17% (n = 8 in 0.01 – 60 mg/kg spiking range) Whole plant (silage): mean = 108% RSD = 6.0% (n = 7 in 0.01 – 10 mg/kg spiking range) Whole plant (hay): mean = 105% RSD = 7.8% (n = 7 in 0.01 – 10 mg/kg spiking range) Grain: mean = 89% RSD = 8.7% (n =13 in 0.01 - 0.1 mg/kg spiking range) Straw: mean = 93% RSD = 8.5% (n = 12 in 0.01 – 10 mg/kg spiking range) **R182281** Whole plant (immature): mean = 99% RSD = 9.3% (n = 8 in 0.01 – 0.5 mg/kg spiking range) Whole plant (silage): mean = 108% RSD = 13% (n = 6 in 0.01 – 0.1 mg/kg spiking range) Whole plant (hay): mean = 109% RSD = 5.5% (n = 6 in 0.01 – 0.1 mg/kg spiking range) Grain: mean = 112% RSD = 5.6% (n = 12 in 0.01- 0.1 mg/kg spiking range) Straw: mean = 107% RSD = 11% (n = 12 in 0.01 – 0.1 mg/kg spiking range)
					49/52†	Whole plant/silage (BBCH 83)	13	0.05	
					49/56†	Whole plant/hay (BBCH 83)	0.56	0.05	
					64	Grain (BBCH 89-92)	<0.01	<0.01	
					64	Straw# (BBCH 89-92)	0.18	0.03	
Report: S12-01272 Study: S12-01272 Trial: S12-01272-02 - Study to GLP - Study carried out in 2012	Wheat (Granary)	UNITED KINGDOM (Europe North) Gloucestershire	769 g a.s./ha 734 g a.s./ha (43-d interval) A14111B	BBCH 37-39; 12-06-2012 BBCH 69; 25-07-2012, interval > 7 days	0 DAA1	Whole plant[1] (BBCH 37-39)	12	0.10	
					22/26†	Whole plant/silage (BBCH 75-79)	12	0.12	
					22/30†	Whole plant/hay (BBCH 75-79)	12	0.13	
					72	Grain (BBCH 89)	<0.01	<0.01	
					72	Straw# (BBCH 89)	1.2	0.07	
Report: S12-01272 Study: S12-01272 Trial: S12-01272-03 - Study to GLP - Study carried out in	Wheat (Tabasco)	GERMANY (Europe North) Niedersachsen	756 g a.s./ha 729 g a.s./ha (48-d interval)	BBCH 31-32; 02-05-2012 BBCH 69; 19-06-2012,	0 DAA1	Whole plant (BBCH 31-32)	40	0.66	
					23/24†	Whole plant/silage (BBCH 81-83)	2.5	0.01	

GLP and Trial Details	Crop (Variety)	Country (Region)	Application Rate Product Code	Growth Stage at application	PHI (days)	Crop Part (Growth stage at sampling)	Residue Found (Uncorrected)		Recovery Data
							chlorothalonil mg/kg	R182281 (mg/kg)	
2012			A14111B	interval > 17 days	23/36†	Whole plant/hay (BBCH 81-83)	4.7	0.09	
					56	Grain (BBCH 89)	<0.01	<0.01	
					56	Straw# (BBCH 89)	0.79	0.06	
Report: S12-01272 Study: S12-01272 Trial: S12-01272-04 - Study to GLP - Study carried out in 2012	Wheat (Asano)	GERMANY (Europe North) Baden Württemberg	733 g a.s./ha 750 g a.s./ha (40-d interval) A14111B	BBCH 30-32; 26-04-2012 BBCH 67-69; 05-06-2012, interval > 17 days	0 DAA1	Whole plant (BBCH 30-32)	23	0.27	
					29/31†	Whole plant/silage (BBCH 75-85)	0.94	0.02	
					29/35†	Whole plant/hay (BBCH 75-85)	1.4	0.04	
					52	Grain (BBCH 89)	<0.01	<0.01	
					52	Straw# (BBCH 89)	1.0	0.09	
Report: S12-01272 Study: S12-01272 Trial: S12-01272-05 - Study to GLP - Study carried out in 2012	Wheat (Magister)	GERMANY (Europe North) Brandenburg	738 g a.s./ha 744 g a.s./ha (43-d interval) A14111B	BBCH 31-32; 02-05-2012 BBCH 69; 14-06-2012, interval > 17 days	0 DAA1	Whole plant (BBCH 31-32)	29	0.61	
					15/22†	Whole plant/silage (BBCH 79-83)	8.0	0.07	
					15/33†	Whole plant/hay[2] (BBCH 79-83)	8.7	0.18	
					49	Grain (BBCH 89)	<0.01	<0.01	
					49	Straw# (BBCH 89)	0.88	0.03	

GLP and Trial Details	Crop (Variety)	Country (Region)	Application Rate Product Code	Growth Stage at application	PHI (days)	Crop Part (Growth stage at sampling)	Residue Found (Uncorrected)		Recovery Data
							chlorothalonil mg/kg	R182281 (mg/kg)	
Report: S12-01272 Study: S12-01272 Trial: S12-01272-06 - Study to GLP - Study carried out in 2012	Wheat (Arrezzo)	FRANCE (Europe North) Alsace	742 g a.s./ha 763 g a.s./ha (56-d interval) A14111B	BBCH 31-32; 13-04-2012 BBCH 69-73; 08-06-2012, interval > 17 days	0 DAA1	Whole plant (BBCH 31-32)	38	0.52	
					18/19†	Whole plant/silage (BBCH 75-77)	3.1	0.03	
					18/21†	Whole plant/hay (BBCH 75-77)	4.5	0.14	
					40	Grain (BBCH 89)	0.01	<0.01	
					40	Straw# (BBCH 89)	1.1	0.04	
Report: S12-01272 Study: S12-01272 Trial: S12-01272-07 - Study to GLP - Study carried out in 2012	Wheat (Campero)	FRANCE (Europe North) Loiret	725 g a.s./ha 770 g a.s./ha (57-d interval) A14111B	BBCH 32; 05-04-2012 BBCH 69; 01-06-2012, interval > 17 days	0 DAA1	Whole plant (BBCH 32)	33	0.22	
					31/31†	Whole plant/silage (BBCH 77)	1.4	0.02	
					31/47†	Whole plant/hay (BCH 77)	2.1	0.05	
					53	Grain (BBCH 89)	<0.01	<0.01	
					53	Straw# (BBCH 89)	0.58	0.02	
Report: S12-01272 Study: S12-01272 Trial: S12-01272-08 - Study to GLP - Study carried out in 2012	Wheat (Tybalt)	POLAND (Europe North) Woj. Wielkopolskie	752 g a.s./ha 711 g a.s./ha (31-d interval) A14111B	BBCH 30-32; 25-05-2012 BBCH 69; 25-06-2012, interval > 17 days	0 DAA1	Whole plant (BBCH 30-32)	21	0.34	
					15/15†	Whole plant/silage (BBCH 75)	1.4	0.01	
					15/29†	Whole plant/hay (BBCH 75)	3.6	0.08	

GLP and Trial Details	Crop (Variety)	Country (Region)	Application Rate Product Code	Growth Stage at application	PHI (days)	Crop Part (Growth stage at sampling)	Residue Found (Uncorrected)		Recovery Data
							chlorothalonil mg/kg	R182281 (mg/kg)	
					60	Grain (BBCH 89)	<0.01	<0.01	
					60	Straw# (BBCH 89)	0.07	0.02	
Report: RAU-019-13 Study: RAU-019-13 Trial: F/CH13/WW05 - Study to GLP - Study carried out in 2013	Wheat (Pakito)	FRANCE (Europe North) Donnelay	742 g a.s./ha 742 g a.s./ha (23-d interval) 500 g/L SC	BBCH 31; 25-04-2013 BBCH 69; 17-05-2013, interval > 17 days	68	Grain (BBCH 89)	<0.01	<0.02	**Chlorothalonil** Grain: mean = 98% RSD = 4.7% (n = 4 in 0.01 -0.10 mg/kg spiking range) Straw: mean = 96% RSD = 3.6% (n = 6 in 0.05 –5.0 mg/kg spiking range) **R182281** Grain: mean = 103% RSD = 11% (n = 4 in 0.02- 0.20 mg/kg spiking range) Straw: mean = 102% RSD = 5.9% (n = 4 in 0.02– 0.20 mg/kg spiking range)
					68	Straw (BBCH 89)	0.85	0.10	
Report: RAU-019-13 Study: RAU-019-13 Trial: G/CH13/WW06 - Study to GLP - Study carried out in 2013	Wheat (Genius)	GERMANY (Europe North) Steinfield	764 g a.s./ha 727 g a.s./ha (49-d interval) 500 g/L SC	BBCH 31; 25-04-2013 BBCH 69; 13-06-2013, interval > 17 days	43	Grain (BBCH 89)	<0.01	<0.02	
					43	Straw (BBCH 89)	0.77	0.06	
Report: RAU-019-13 Study: RAU-019-13 Trial: G/CH13/WW07 - Study to GLP - Study carried out in 2013	Wheat (Cubus)	GERMANY (Europe North) Friesenheim	727 g a.s./ha 791 g a.s./ha (51-d interval) 500 g/L SC	BBCH 31; 29-04-2013 BBCH 69; 19-06-2013, interval > 17 days	33	Grain (BBCH 89)	<0.01	<0.02	
					33	Straw (BBCH 89)	1.9	0.07	
Report: RAU-019-13 Study: RAU-019-13 Trial: B/CH13/WW08 - Study to GLP - Study carried out in 2013	Wheat (Matrix)	BELGIUM (Europe North) Villers-Perwin	748 g a.s./ha 723 g a.s./ha (49-d interval) 500 g/L SC	BBCH 31; 06-05-2013 BBCH 69; 24-06-2013, interval > 17 days	43	Grain (BBCH 89)	<0.01	<0.02	
					43	Straw (BBCH 89)	0.44	0.07	
Report: BIU-015-14	Wheat	FRANCE	729 g a.s./ha	BBCH 37;	42	Grain	<0.01	<0.02	**Chlorothalonil**

GLP and Trial Details	Crop (Variety)	Country (Region)	Application Rate Product Code	Growth Stage at application	PHI (days)	Crop Part (Growth stage at sampling)	Residue Found (Uncorrected)		Recovery Data
							chlorothalonil mg/kg	R182281 (mg/kg)	
Study: BIU-015-14 Trial: F/CH14/WW05 - Study to GLP - Study carried out in 2014	(Lear)	(Europe North) Houdilcourt	774 g a.s./ha (32-d interval) 500 g/L SC	05-05-2014 BBCH 69; 06-06-2014, interval > 17 days		(BBCH 89)			Grain: mean = 82% RSD = 2.3% (n = 4 in 0.01 -0.10 mg/kg spiking range) Whole plant: mean = 89% RSD = 5.0% (n = 4 in 0.05 –20 mg/kg spiking range) Straw: mean = 86% RSD = 16% (n = 4 in 0.05 –12 mg/kg spiking range) **R182281** Grain: mean = 85% RSD = NA (n = 2 in 0.02- 0.20 mg/kg spiking range) Whole plant: mean = 100% RSD = NA (n = 2 in 0.02 –1.0 mg/kg spiking range) Straw: mean = 91% RSD = NA (n = 2 in 0.02– 0.2 mg/kg spiking range)
					42	Straw (BBCH 89)	1.1	0.07	
Report: BIU-015-14 Study: BIU-015-14 Trial: F/CH14/WW06 - Study to GLP - Study carried out in 2014	Wheat (Rubisko)	FRANCE (Europe North) Saint Fergeux	764 g a.s./ha 771 g a.s./ha (32-d interval) 500 g/L SC	BBCH 41; 05-05-2014 BBCH 69; 06-06-2014, interval > 17 days	0	Whole plant (BBCH 69)	12	0.16	
					11/19†	Whole plant/silage (BBCH 75-77)	1.6	0.11	
					11/28†	Whole plant/hay (BBCH 75-77)	2.7	0.15	
					41	Grain (BBCH 89)	<0.01	<0.02	
					41	Straw (BBCH 89)	1.1	0.04	
Report: BIU-015-14 Study: BIU-015-14 Trial: F/CH14/WW07 - Study to GLP - Study carried out in 2014	Wheat (JB Diego)	FRANCE (Europe North) Inchy en Artois	744 g a.s./ha 735 g a.s./ha (38-d interval) 500 g/L SC	BBCH 32; 05-05-2014 BBCH 62; 12-06-2014, interval > 17 days	53	Grain (BBCH 89)	<0.01	<0.02	
					53	Straw (BBCH 89)	0.50	0.03	
Report: BIU-015-14 Study: BIU-015-14 Trial: P/CH14/WW08 - Study to GLP - Study carried out in 2014	Wheat (Jantarka)	POLAND (Europe North) Michalow	756 g a.s./ha 763 g a.s./ha (31-d interval) 500 g/L SC	BBCH 37; 05-05-2014 BBCH 69; 05-06-2014, interval > 17 days	0	Whole plant (BBCH 69)	13	0.20	
					13/14†	Whole plant/silage (BBCH 77)	12	0.19	
					13/22†	Whole plant/hay (BBCH 77)	11	0.15	
					55	Grain (BBCH 89)	<0.01	<0.02	

GLP and Trial Details	Crop (Variety)	Country (Region)	Application Rate Product Code	Growth Stage at application	PHI (days)	Crop Part (Growth stage at sampling)	Residue Found (Uncorrected)		Recovery Data
							chlorothalonil mg/kg	R182281 (mg/kg)	
					55	Straw[4] (BBCH 89)	0.06	0.01	
Southern Europe									
Report: S12-01273 Study: S12-01273 Trial: S12-01273-01 - Study to GLP - Study carried out in 2012	Wheat (Ingenio)	FRANCE (Europe South) Tarn et Garonne	755 g a.s./ha 727 g a.s./ha (47-d interval) A14111B	BBCH 31-32; 06-04-2012 BBCH 69; 23-05-2012, interval> 17 days	0 DAA1	Whole plant (BBCH 31-32)	20	0.19	**Chlorothalonil** Whole plant (immature): mean = 96% RSD = 14% (n = 7 in 0.01 – 60 mg/kg spiking range) Whole plant (silage): mean = 106% RSD = 6.0% (n = 7 in 0.01 – 10 mg/kg spiking range) Whole plant (hay): mean = 104% RSD = 15% (n = 7 in 0.01 – 10 mg/kg spiking range) Grain: mean = 87% RSD = 8.2% (n =11 in 0.01 - 0.1 mg/kg spiking range) Straw: mean = 99% RSD = 10% (n = 11 in 0.01 – 10 mg/kg spiking range) **R182281** Whole plant (immature): mean = 99% RSD = 4.0% (n = 6 in 0.01 – 0.1 mg/kg spiking range) Whole plant (silage): mean = 106% RSD = 12% (n = 6 in 0.01 – 0.1 mg/kg spiking range) Whole plant (hay): mean = 111% RSD = 9.3% (n = 6 in 0.01 – 0.1 mg/kg spiking range) Grain: mean = 107% RSD = 7.3% (n = 11 in 0.01- 0.1 mg/kg spiking range) Straw: mean = 101% RSD = 5.2% (n =
					29/29†	Whole plant/silage (BBCH 83-85)	1.5	0.02	
					29/33†	Whole plant/hay (BBCH 83-85)	1.7	0.08	
					41	Grain (BBCH 89)	<0.01	<0.01	
					41	Straw# (BBCH 89)	0.92	0.07	
Report: S12-01273 Study: S12-01273 Trial: S12-01273-02 - Study to GLP - Study carried out in 2012	Wheat (Sirtaki)	FRANCE (Europe South) Aude	784 g a.s./ha 750 g a.s./ha (54-d interval) A14111B	BBCH 32; 30-03-2012 BBCH 69; 23-05-2012, interval > 17 days	0 DAA1	Whole plant (BBCH 32)	20	0.24	
					26/27†	Whole plant/silage[3] (BBCH 75)	1.4	0.03	
					26/30†	Whole plant/hay[3] (BBCH 75)	0.44	0.02	
					47	Grain (BBCH 89)	<0.01	<0.01	
					47	Straw# (BBCH 89)	0.62	0.04	

GLP and Trial Details	Crop (Variety)	Country (Region)	Application Rate Product Code	Growth Stage at application	PHI (days)	Crop Part (Growth stage at sampling)	Residue Found (Uncorrected)		Recovery Data
							chlorothalonil mg/kg	R182281 (mg/kg)	
Report: S12-01273 Study: S12-01273 Trial: S12-01273-03 - Study to GLP - Study carried out in 2012	Wheat (Arezzo)	FRANCE (Europe South) Ain	716 g a.s./ha 749 g a.s./ha (48-d interval) A14111B	BBCH 32; 13-04-2012 BBCH 69; 31-05-2012, interval > 17 days	0 DAA1	Whole plant (BBCH 32)	18	0.17	
					13/14†	Whole plant/silage (BBCH 75-85)	3.1	0.05	
					13/18†	Whole plant/hay (BBCH 75-85)	0.70	0.22	
					40	Grain (BBCH 89)	<0.01	<0.01	
					40	Straw# (BBCH 89)	0.40	0.04	
Report: S12-01273 Study: S12-01273 Trial: S12-01273-04 - Study to GLP - Study carried out in 2012	Wheat (Dylan)	ITALY (Europe South) Emilia-Romagna, Granarolo dell' Emilia	780 g a.s./ha 727 g a.s./ha (37-d interval) A14111B	BBCH 30-32; 03-04-2012 BBCH 69; 10-05-2012, interval > 17 days	0 DAA1	Whole plant (BBCH 30-32)	38	0.16	
					33/36†	Whole plant/silage (BBCH 79-83)	2.7	0.04	
					33/40†	Whole plant/hay (BBCH 79-83)	1.0	0.08	
					46	Grain (BBCH 89)	<0.01	<0.01	
					46	Straw# (BBCH 89)	0.99	0.08	
Report: S12-01273 Study: S12-01273 Trial: S12-01273-05 - Study to GLP - Study carried out in 2012	Wheat (Simeto)	ITALY (Europe South) Emilia-Romagna, Conselice	750 g a.s./ha 755 g a.s./ha (27-d interval) A14111B	BBCH 30-32; 12-04-2012 BBCH 69; 09-05-2012, interval > 17 days	0 DAA1	Whole plant (BBCH 30-32)	35	0.55	
					27/30†	Whole plant/silage (BBCH 79-83)	2.0	0.12	
					27/33†	Whole plant/hay (BBCH 79-83)	0.68	0.15	
					43	Grain	<0.01	<0.01	

GLP and Trial Details	Crop (Variety)	Country (Region)	Application Rate Product Code	Growth Stage at application	PHI (days)	Crop Part (Growth stage at sampling)	Residue Found (Uncorrected)		Recovery Data
							chlorothalonil mg/kg	R182281 (mg/kg)	
						(BBCH 89)			
					43	Straw# (BBCH 89)	1.1	0.32	
Report: S12-01273 Study: S12-01273 Trial: S12-01273-06 - Study to GLP - Study carried out in 2012	Wheat (Marius)	SPAIN (Europe South) Aragon, Fonfria	784 g a.s./ha 733 g a.s./ha (29-d interval) A14111B	BBCH 30-32; 10-05-2012 BBCH 69; 08-06-2012, interval > 17 days	0 DAA1	Whole plant (BBCH 30-32)	29	0.43	
					13/14†	Whole plant/silage (BBCH 75-85)	9.4	0.04	
					13/18†	Whole plant/hay (BBCH 75-85)	4.9	0.15	
					48	Grain (BBCH 89)	<0.01	<0.01	
					48	Straw# (BBCH 89)	9.9	0.43	
Report: S12-01273 Study: S12-01273 Trial: S12-01273-07 - Study to GLP - Study carried out in 2012	Wheat (Bastide)	SPAIN (Europe South) Aragon, Lechon	773 g a.s./ha 781 g a.s./ha (35-d interval) A14111B	BBCH 30-32; 11-05-2012 BBCH 68-70; 15-06-2012, interval > 17 days	0 DAA1	Whole plant (BBCH 30-32)	34	0.46	
					11/12†	Whole plant/silage (BBCH 75-85)	6.6	0.40	
					11/14†	Whole plant/hay (BBCH 75-85)	2.9	0.37	
					26	Grain (BBCH 89)	0.02	<0.01	
					26	Straw# (BBCH 89)	4.1	0.58	
Report: S12-01273 Study: S12-01273 Trial: S12-01273-08 - Study to GLP - Study carried out in	Wheat (Marius)	SPAIN (Europe South) Aragon, Lagueruela	681 g a.s./ha 776 g a.s./ha (33-d interval)	BBCH 30-32; 09-05-2012 BBCH 69; 11-06-2012,	0 DAA1	Whole plant (BBCH 30-32)	34	0.28	
					14/16†	Whole plant/silage (BBCH 75-85)	5.6	0.51	

GLP and Trial Details	Crop (Variety)	Country (Region)	Application Rate Product Code	Growth Stage at application	PHI (days)	Crop Part (Growth stage at sampling)	Residue Found (Uncorrected)		Recovery Data
							chlorothalonil mg/kg	R182281 (mg/kg)	
2012			A14111B	interval > 17 days	14/18†	Whole plant/hay (BBCH 75-85)	2.6	0.30	
					31	Grain (BBCH 89)	0.01	<0.01	
					31	Straw# (BBCH 89)	6.8	0.38	
Report: RAU-017-13 Study: RAU-017-13 Trial: I/CH13/WW01 - Study to GLP - Study carried out in 2013	Wheat (Levante)	ITALY (Europe South) Castelnuovo Scrivia	786 g a.s./ha 791 g a.s./ha (31-d interval) 500 g/L SC	BBCH 31; 17-04-2013 BBCH 69; 18-05-2013, interval > 17 days	58	Grain (BBCH 89)	<0.01	<0.02	**Chlorothalonil** Grain: mean = 97% RSD = 1.5% (n = 6 in 0.01 -0.10 mg/kg spiking range) Straw: mean = 100% RSD = 2.6% (n = 8 in 0.05 –5.0 mg/kg spiking range) **R182281** Grain: mean = 95% RSD =4.6% (n = 4 in 0.02- 0.20 mg/kg spiking range) Straw: mean = 87% RSD = 5.8% (n = 4 in 0.02– 0.50 mg/kg spiking range)
					58	Straw (BBCH 89)	0.22	0.08	
Report: RAU-017-13 Study: RAU-017-13 Trial: I/CH13/WW02 - Study to GLP - Study carried out in 2013	Wheat (PR 058)	ITALY (Europe South) Roccabianca	745 g a.s./ha 745 g a.s./ha (34-d interval) 500 g/L SC	BBCH 31; 18-04-2013 BBCH 69; 22-05-2013, interval > 17 days	42	Grain (BBCH 89)	<0.01	<0.02	
					42	Straw (BBCH 89)	0.08	0.03	
Report: RAU-017-13 Study: RAU-017-13 Trial: I/CH13/WW03 - Study to GLP - Study carried out in 2013	Wheat (Arrocco)	ITALY (Europe South) Busseto	761 g a.s./ha 740 g a.s./ha (34-d interval) 500 g/L SC	BBCH 31; 18-04-2013 BBCH 69; 22-05-2013, interval > 17 days	40	Grain (BBCH 89)	<0.01	<0.02	
					40	Straw (BBCH 89)	0.07	0.04	
Report: RAU-017-13 Study: RAU-017-13 Trial: F/CH13/WW04 - Study to GLP - Study carried out in 2013	Wheat (Quality)	FRANCE (Europe South) Castelnau-d'Estretefonds	779 g a.s./ha 764 g a.s./ha (19-d interval) 500 g/L SC	BBCH 41; 24-04-2013 BBCH 69; 13-05-2013, interval 19 days	63	Grain (BBCH 89)	<0.01	<0.02	
					63	Straw (BBCH 89)	2.1	0.12	
Report: BIU-014-14 Study: BIU-014-14	Wheat (Salgemma)	ITALY (Europe South)	759 g a.s./ha 763 g a.s./ha	BBCH 39; 17-04-2014	58	Grain (BBCH 89)	<0.01	<0.02	**Chlorothalonil**

GLP and Trial Details	Crop (Variety)	Country (Region)	Application Rate Product Code	Growth Stage at application	PHI (days)	Crop Part (Growth stage at sampling)	Residue Found (Uncorrected)		Recovery Data
							chlorothalonil mg/kg	R182281 (mg/kg)	
Trial: I/CH14/WW01 - Study to GLP - Study carried out in 2014		Monticelli Pavese	(27-d interval) 500 g/L SC	BBCH 69; 14-05-2014, interval > 17 days	58	Straw (BBCH 89)	0.08	<0.02	Grain: mean = 100% RSD = 15% (n = 4 in 0.01 -0.10 mg/kg spiking range) Silage: mean = 97% RSD = 5.6% (n = 9 in 0.05 –28 mg/kg spiking range) Straw: mean = 90% RSD = NA (n = 2 in 0.05 –2.0 mg/kg spiking range) Hay: mean = 92% RSD = NA (n = 2 at 11 mg/kg) **R182281** Grain: mean = 78% RSD = 12% (n = 10 in 0.02- 0.20 mg/kg spiking range) Silage : mean = 105% RSD = 8.5% (n = 9 in 0.02 –1.0 mg/kg spiking range) Straw: mean = 98% RSD = 6.2% (n = 6 in 0.02– 0.2 mg/kg spiking range)
Report: BIU-014-14 Study: BIU-014-14 Trial: I/CH14/WW02 - Study to GLP - Study carried out in 2014	Wheat (Levante)	ITALY (Europe South); Castelnuovo Scrivia	746 g a.s./ha 756 g a.s./ha (27-d interval) 500 g/L SC	BBCH 33; 18-04-2014 BBCH 69; 15-05-2014, interval > 17 days	0	Whole plant (BBCH 69)	21	0.74	
					12	Whole plant/silage (BBCH 75)	5.8	0.06	
					13	Whole plant/hay (BBCH 75)	6.8	<0.02	
					48	Grain (BBCH 89)	<0.01	<0.02	
					48	Straw (BBCH 89)	0.08	<0.02	
Report: BIU-014-14 Study: BIU-014-14 Trial: I/CH14/WW03 - Study to GLP - Study carried out in 2014	Wheat (Aubusson)	ITALY (Europe South); Roccabianca	739 g a.s./ha 756 g a.s./ha (27-d interval) 500 g/L SC	BBCH 32; 17-04-2014 BBCH 69; 14-05-2014, interval > 17 days	0	Whole plant (BBCH 69)	20	0.36	
					21	Whole plant/silage (BBCH 77)	2.5	0.02	
					22	Whole plant/hay (BBCH 77)	2.7	<0.02	
					40	Grain (BBCH 89)	<0.01	<0.02	
					40	Straw (BBCH 89)	0.33	<0.02	
Report: BIU-014-14	Wheat	FRANCE	764 g a.s./ha	BBCH 61;	46	Grain	<0.01	<0.02	

GLP and Trial Details	Crop (Variety)	Country (Region)	Application Rate Product Code	Growth Stage at application	PHI (days)	Crop Part (Growth stage at sampling)	Residue Found (Uncorrected)		Recovery Data
							chlorothalonil mg/kg	R182281 (mg/kg)	
Study: BIU-014-14 Trial: F/CH14/WW04 - Study to GLP - Study carried out in 2014	(Solveig)	(Europe South) Langon	782 g a.s./ha (19-d interval) 500 g/L SC	07-05-2014 BBCH 69; 26-05-2014, interval 18 days		(BBCH 89)			
					46	Straw (BBCH 89)	0.69	0.03	

Residues in all untreated samples were < 0.01 mg/kg for chlorothalonil and <0.02 mg/kg for R182281 with the following exceptions:

- [1]Trial S12-01272-02 – residues of chlorothalonil were found in untreated samples of whole plant (0.03 mg/kg).
- [2]Trial S12-01272-05 – residues of chlorothalonil were found in untreated samples of hay (0.02 mg/kg).
- [3]Trial S12-01273-02 – residues of chlorothalonil were found in untreated samples of whole plant (0.27 mg/kg), silage (0.17 mg/kg) and hay (0.15 mg/kg).
- [4]Trial P/CH14/WW08 (Poland) – residues of chlorothalonil and R182281 were found in untreated samples of straw (0.20-0.23 and 0.065-0.092 mg/kg respectively).

DAA1 = days after first application. All other PHI values are days after the second application

†For silage and hay samples were cut at various growth stages between BBCH 75 and 85 then left in the field to dry until the moisture content reached the typical moisture content of silage and hay respectively. For these crops there are therefore two PHI values reported: the PHI when the crop was cut and the PHI when the sample was taken from the field (i.e. including drying).

#~~The demonstrated storage stability of chlorothalonil in straw is 12 months, while these samples have been stored up to maximally 16 months. Results of chlorothalonil in straw are not acceptable.~~

B.7.3.4 Potato

Chlorothalonil is proposed for use on potato in NEU and SEU according to the following GAP:

1x 750 g/ha, PHI 28 days.

This GAP is less critical than the GAP for potatoes during the initial peer review, in which the number of application was 5 to 13 in the corresponding trials. Therefore, the evaluation of these trials is not copied from the old DAR into this RAR.

For the renewal of chlorothalonil, additional reports containing supervised residue trials have been submitted. The residue reports supporting the proposed EU critical GAP for chlorothalonil on potato are referenced in Table 7.3.4-1 and the data are presented in Table 7.3.4-4.

Previous evaluation	Submitted for the purpose of renewal
RMS remark	Acceptable

Table 7.3.4-1: Report references for trials supporting the proposed EU critical GAP for chlorothalonil on potato

Annex Pt.	Number.	Author/s	Issue Year	Report Title
K-CA 6.3.4/01	(1 of 4)	A Sala	2014d	Determination of chlorothalonil and its metabolite SDS3701 residues in raw agricultural commodity potato following three applications of chlorothalonil 500SC, 2 trials, northern Europe, year 2013 . Research Centre Biospheres, Via Vittorio Veneto 81, 26857 Salerano sul Lambro, Italy. Syngenta File No. R044636_11232, Report No. RAU-022-13
K-CA 6.3.4/02	(2 of 4)	A Sala	2014e	Determination of chlorothalonil and its metabolite SDS3701 residues in raw agricultural commodity potato following three applications of chlorothalonil 500SC (2 trials, northern Europe, year 2014) . Research Centre Biospheres, Via Vittorio Veneto 81, 26857 Salerano sul Lambro, Italy. Syngenta File No. R044636_11234, Report No. RAU-011-14
K-CA 6.3.4/03	(3 of 4)	A Sala	2014f	Determination of chlorothalonil and its metabolite SDS3701 residues in raw agricultural commodity potato following three applications of chlorothalonil 500SC, 2 trials, southern Europe, year 2013. Research Centre Biospheres, Via Vittorio Veneto 81, 26857 Salerano sul Lambro, Italy. Syngenta File No. R044636_11231, Report No. RAU-021-13
K-CA 6.3.4/04	(4 of 4)	A Sala	2014g	Determination of chlorothalonil and its metabolite SDS3701 residues in raw agricultural commodity potato following three applications of chlorothalonil 500SC (2 trials, southern Europe, year 2014). Research Centre Biospheres, Via Vittorio Veneto 81, 26857 Salerano sul Lambro, Italy. Syngenta File No. R044636_11233, Report No. RAU-022-14

Guidelines

The studies meet the requirements of the Commission of the European Communities: General Recommendations for the Design, Preparation and Realization of Residue Trials (7029/V1/95 rev. 5, 22/7/1997), and are designed to comply with Regulation (EC) 1107/2009.

GLP

All trials (field and analytical phases) were carried out in compliance with the principles of Good Laboratory Practice.

Materials and Methods

Eight supervised residue trials were conducted on field grown potato in 2013 and 2014, in northern or southern Europe. A summary of the trials conducted is presented in Table 7.3.4-2.

Table 7.3.4-2: Summary of chlorothalonil residue trials on potato

Country	2013	2014
Northern Europe		
France (north)	1 Harvest	2 Harvest
Belgium	1 Harvest	-
Southern Europe		
France (south)	1 Harvest	1 Harvest
Italy	1 Harvest	1 Harvest

Potatoes are a major crop in northern and southern Europe and therefore generally require eight trials in each residue region.

Treatments with chlorothalonil were conducted as post emergence (BBCH 39-47 based on growth stages of the tuber) spray applications utilising the formulation as detailed in Table 7.3.4-3 at a nominal application rate of 750 g a.s./ha (actual rates 697-812 g a.s./ha) with an interval of 7 days between applications. The water volumes during application ranged from 346 to 510 L/ha.

Table 7.3.4-3: Summary of chlorothalonil formulations used in the presented trials

Product code	Formulation type	Composition	
		2013	2014
Chlorothalonil 500 SC	SC	502 g/L chlorothalonil (batch O232)	507 g/L chlorothalonil (batch PN1911)

Samples of whole tubers were taken and analysed for residues of parent chlorothalonil and the metabolite R182281 (SDS3701) by analytical methods described in study RAU-022-14. The LOQ is 0.01 mg/kg for chlorothalonil and 0.02 mg/kg for R182281. The analytical method (indicated by RAU-076-00 or RAU-076-01) used involved extraction with acidified ethyl acetate followed by analysis using a gas chromatograph equipped with µ-ECD detector for chlorothalonil. For R182281 the analytical method consisted of extraction with methanol and analysis with LC-MS/MS. Full method descriptions and validation data are presented in B.5.1.2.5 (K-CA 4.1.2/31). Procedural recovery data are presented with the results of the residues trials in Table 7.3.4-4.

Samples were stored up to a maximum of 5 months from sampling to extraction. Samples were not homogenised before freezing. Residues of chlorothalonil and R18221 are stable in high starch

commodities for at least 24 months ~~(see section CA 6.1)~~ and, therefore, no degradation will have occurred between sampling and analysis.

The proposed GAP is for one application; however, all the trials were conducted with three applications. Residues of both chlorothalonil and R182281 were below the LOQ in tubers in all the trials, indicating that the number of applications did not impact on the residue levels at harvest. Since the overdosed trials still do not result in detectable residues, these trials are considered acceptable. Furthermore, the expected 'zero' residue situation is also supported by the primary metabolism study on carrot, in which a very limited translocation of total residues from foliage to roots was observed (see B.7.2.1.3).
Therefore, in total 4 acceptable supervised residue trials are available in NEU and also 4 acceptable trial in SEU. Although generally a minimum of 8 trials are required in each region, the residues of both chlorothalonil and R182281 were below the LOQ in all trials, therefore, a reduced data set of 4 trials for each region is acceptable.

The applicant states that the latest modelling results (MCP section 9) have shown that the use on potato can only be supported with 1 application. Nevertheless trials to address one application on potatoes are ongoing and can be presented in the course of the EU-evaluation.

The results of the residue trials for chlorothalonil and R182281 are presented in Table 7.3.4-4.

Table 7.3.4-4: Summary of residue data supporting the EU critical GAP for chlorothalonil on potato

GLP and Trial Details	Crop (Variety)	Country (Region)	Application Rate Product Code	Growth Stage at application	PHI (days)	Crop Part (Growth stage at sampling)	Residue Found (Uncorrected) chlorothalonil mg/kg	 R182281 (mg/kg)	Recovery Data
Northern Europe									
Report: RAU-022-13 Study : RAU-022-13 Trial: F/CH13/PO03 Study to GLP - Study carried out in 2013	Potato (Marabel)	FRANCE (Europe North) Handschuheim	715 g a.s./ha 778 g a.s./ha 766 g a.s./ha (7-d interval) 500 g/L SC	BBCH 39-40; 19-06-2013 BBCH 40; 26-06-2013 BBCH 40-43; 02-07-2013	28	Tuber (BBCH 49)	<0.01	<0.02	Chlorothalonil Whole tubers: mean = 96% RSD = NA (n = 2 in 0.01 – 0.1 mg/kg spiking range) R182281 (SDS3701) Whole tubers: mean = 101% RSD = NA (n = 2 in 0.02 – 0.20 mg/kg spiking range)
Report: RAU-022-13 Study : RAU-022-13 Trial: B/CH13/PO04 Study to GLP - Study carried out in 2013	Potato (Bintje)	BELGIUM (Europe North) Marbais	759 g a.s./ha 747 g a.s./ha 697 g a.s./ha (7-d interval) 500 g/L SC	BBCH 45†; 01-08-2013 BBCH 47-48†; 09-08-2013 BBCH 49†; 16-08-2013	28	Tuber (BBCH 49)	<0.01	<0.02	
Report: RAU-011-14 Study : RAU-011-14 Trial: F/CH14/PO03 Study to GLP - Study carried out in 2014	Potato (Samba)	FRANCE (Europe North) Dame Marie les Bois	786 g a.s./ha 786 g a.s./ha 812 g a.s./ha (7-d interval) 500 g/L SC	BBCH 43; 31-07-2014 BBCH 43; 07-08-2014 BBCH 45; 14-08-2014	28	Tuber (BBCH 49)	<0.01	<0.02	Chlorothalonil Whole tubers: mean = 97% RSD = NA (n = 2 in 0.01 – 0.1 mg/kg spiking range) R182281 (SDS3701) Whole tubers: mean = 90% RSD = NA (n = 2 in 0.02 – 0.20 mg/kg spiking range)
Report: RAU-011-14 Study : RAU-011-14 Trial: F/CH14/PO04 Study to GLP - Study carried out in 2014	Potato (Bintje)	FRANCE (Europe North) Inchy en Artois	751 g a.s./ha 766 g a.s./ha 766 g a.s./ha (7-d interval) 500 g/L SC	BBCH 45; 23-07-2014 BBCH 45; 30-07-2014 BBCH 46; 07-08-2014	28	Tuber (BBCH 49)	<0.01	<0.02	

GLP and Trial Details	Crop (Variety)	Country (Region)	Application Rate Product Code	Growth Stage at application	PHI (days)	Crop Part (Growth stage at sampling)	Residue Found (Uncorrected) chlorothalonil mg/kg	 R182281 (mg/kg)	Recovery Data
Southern Europe									
Report: RAU-021-13 Study : RAU-021-13 Trial: F/CH13/PO01 Study to GLP - Study carried out in 2013	Potato (Ermes)	ITALY (Europe South) Frazione Montariolo	764 g a.s./ha 753 g a.s./ha 740 g a.s./ha (7-d interval) 500 g/L SC	BBCH 43†; 25-07-2013 BBCH 43†; 01-08-2013 BBCH 43†; 09-08-2013	27	Tuber (BBCH 47-48†)	<0.01	<0.02	Chlorothalonil Whole tubers: mean = 95% RSD = NA (n = 2 in 0.01 – 0.1 mg/kg spiking range) R182281 (SDS3701) Whole tubers: mean = 99% RSD = NA (n = in 0.02 – 0.20 mg/kg spiking range)
Report: RAU-021-13 Study : RAU-021-13 Trial: F/CH13/PO02 Study to GLP - Study carried out in 2013	Potato (Agatha)	FRANCE (Europe South) Midi Pyrénées	766 g a.s./ha 777 g a.s./ha 766 g a.s./ha (7-d interval) 500 g/L SC	BBCH 41-43; 12-06-2013 BBCH 43; 19-06-2013 BBCH 43; 26-06-2013	28	Tuber (BBCH 49)	<0.01	<0.02	
Report: RAU-022-14 Study : RAU-022-14 Trial: F/CH14/PO01 Study to GLP - Study carried out in 2014	Potato (Kennebec)	ITALY (Europe South) Ronco all'Adige	776 g a.s./ha 776 g a.s./ha 761 g a.s./ha (7-d interval) 500 g/L SC	BBCH 41; 12-06-2014 BBCH 44; 19-06-2014 BBCH 47; 26-06-2014	28	Tuber (BBCH 49)	<0.01	<0.02	Chlorothalonil Whole tubers: mean = 96% RSD = 6.2% (n = 10 in 0.01 – 0.1 mg/kg spiking range) R182281 (SDS3701) Whole tubers: mean = 97% RSD = 6.4% (n = 10 in 0.02 –0.20 mg/kg spiking range)
Report: RAU-022-14 Study : RAU-022-14 Trial: F/CH14/PO02 Study to GLP - Study carried out in 2014	Potato (Bintje)	FRANCE (Europe South) Marsillargues	788 g a.s./ha 794 g a.s./ha 788 g a.s./ha (7-d interval) 500 g/L SC	BBCH 42; 06-06-2014 BBCH 43†; 13-06-2014 BBCH 43; 20-06-2014	28	Tuber (BBCH 48†)	<0.01	<0.02	

Unless otherwise stated residues of chlorothalonil and R182281 in untreated samples were < LOQ of 0.01 and 0.02 mg/kg respectively.

†Growth stages were expressed in terms of the foliage in the report. These have been expressed in the terms of the tuber for consistency across all trials.

NA = not applicable

B.7.4 Feeding studies

Products from barley, wheat and potato may form a part of global livestock diet in the EU. Therefore, the median and maximum dietary burdens were calculated for different groups of livestock using OECD guideline 505 and the OECD Guidance document on residues in livestock, series on pesticides No 73.

Barley straw and grain; wheat straw and grain; distillers grain, brewers grain, wheat meal; wheat milled by-products; potato culls; potato process waste and potato dried pulp are considered part of the dietary burden for livestock in EU.

Barley and wheat forage and silage are not considered relevant as the proposed uses for chlorothalonil are on cereals for grain production only.

The input values for chlorothalonil are shown in table B.7.4-1 and for SDS-3701 in table B.7.4-3, and are obtained from the supervised residue trials in B.7.3.

Since no residues are expected in potatoes, no processing factor has been used for the input value of potato process waste and dried pulp. The LOQ-STMR has been used for these potato by-products. The same applies to ‘wheat, milled by-products’: residues in wheat grain are <LOQ and, therefore, also no processing factor has been used for the ‘wheat, milled by-products’. For ‘brewer’s grain, dried’ and ‘distiller’s grain, dried’ as a worst-case the STMR for barley grain has been used. No processing factor is known for ‘brewer’s grain’ and ‘distiller’s grain’. Although grain itself is already a rather dry product, concentration of residues could theoretically occur during production of brewer’s grain and distiller’s grain, taking into account the yielding factor (see also the default processing factor of 3.3 according to PROFILe 3.0). However, for barley, all processing factors are <1 for both chlorothalonil and SDS-3701. Therefore, using the STMR for barley grain in the dietary burden calculation seems a reasonable worst-case assumption. Furthermore, for wheat, all processing factors for chlorothalonil and SDS-3701 in grain are ≤1 (except for wheat bran and flour). It should be noted that in some cases wheat bran may be added at the end of the process leading to distiller’s grain. Nevertheless, since residues in wheat grain are <LOQ, the STMR for barley grain still appears to be an acceptable conservative value.

The dietary burden has been calculated for chlorothalonil (table B.7.4-2) and SDS-3701 separately (table B.7.4-4). Subsequently, since both chlorothalonil residues as well as SDS-3701 residues could be present in the feed at the same time, the combined residue has been calculated and expressed as chlorothalonil by applying the molecular weight conversion factor of 1.07, as follows:

Combined residues = chlorothalonil residues + (SDS-3701 residues x 1.07).

The combined residues input values for the dietary burden calculation are presented in table B.7.4-5. The calculation of livestock exposure for the combined residues is presented in table B.7.4-6 and confirms that this livestock exposure is indeed the sum of the two separate calculations.

From the separate calculations, a ratio between chlorothalonil and SDS-3701 can be derived (table 7.4-7). The ratio can also be determined based on the median of the ratio’s of the selected residue values for both grain and straw in the supervised residue trials:

Barley: 5x 1:1; 3x 1.5:1; 1.6:1; 5x 2:1; 3.2:1; 3.7:1; 3.8:1; 3.9:1; 4:1; 4.5:1; 2x 5:1; **5.3:1**; 2x 6.8:1; 7.5:1; 9.6:1; 10:1; 10.5:1; 11.5:1; 12.3:1; 14.7:1; 15:1; 18:1; 18.6:1; 18.9: 1; 2x 19:1; 20:1; 22.5:1; 24:1; 24.4:1; 25.5:1; 30:1; 50:1

Wheat: 2x 1:1; 1.8:1; 2:1; 2.7:1; 2.8:1; 2x 4:1; **6.3:1**; 8.5:1; 12.8:1; 15.7:1; 16.5:1; 17.5:1; 23:1; 27.1:1; 27.5:1

When residues were <LOQ for both chlorothalonil and SDS-3701, then no ratio has been calculated.

In addition, the separate ratio's for grain and straw can also be shown (more ratio's are shown below than before the peer review, since more wheat straw samples are now considered acceptable concerning storage stability):

Barley grain: 4x 1:1; 2x 1.5:1; 4x **2:1**; 4:1; 18:1; 19:1

Barley straw: 1:1; 1.5:1; 1.6:1; 2:1; 3.2:1; 3.7:1; 3.8:1; 3.9:1; 4.5:1; 2x 5:1; 5.3:1; 2x 6.8:1; 7.5:1; **9.6:1; 10:1;** 10.5:1; 11.5:1; 12.3:1; 14.7:1; 15:1; 18.6:1; 18.9:1; 19:1; 20:1; 22.5:1; 24:1; 24.4:1; 25.5:1; 30:1; 50:1

Wheat grain: 2x **1:1**; 2:1

Wheat straw: 1.8:1; 2.7:1; 2.8:1; 3.4:1; 3.5:1; 2x 4:1; 6:1; 6.3:1; 7.1:1; 8.5:1; 10:1; 11.1:1; 12.4:1; **12.8:1; 13.1:1**; 13.2:1; 15.5:1; 15.7:1; 16.5:1; 17.1:1; 17.5:1; 17.9:1; 2x 23:1; 27.1:1; 2x 27.5:1; 29:1; 29.3:1

Values in bold depict the median value.

Based on these separate ratio's, it can be observed that the ratio for straw is considerably higher than for grain.

The results of the dietary burden calculation for the combined residues show that the trigger value of 0.004 mg/kg bw/d is exceeded for all groups of livestock, except for pigs.

Table B.7.4-1: Chlorothalonil input values for dietary burden calculation

Commodity	Median dietary burden		Maximum dietary burden	
	Input value (mg/kg)	Comment	Input value (mg/kg)	Comment
Barley, straw	1.5	STMR from NEU	5.7	HR from NEU
Barley, grain	0.02	STMR from NEU	n.a.	
Wheat, straw	0.87 ~~0.98~~	STMR from NEU ~~(based on 6 trials)~~	9.9 ~~2.1~~	HR from SEU
Wheat, grain	<0.01	STMR	n.a.	
Wheat gluten, meal	<0.002	STMR grain x PF 'wheat, gluten feed meal' (<0.01 x <0.20)	n.a.	

Commodity	Median dietary burden		Maximum dietary burden	
	Input value (mg/kg)	Comment	Input value (mg/kg)	Comment
Wheat, milled by-products	<0.01	STMR wheat grain	n.a.	
Brewer's grain, dried	0.02	STMR barley grain	n.a.	
Distiller's grain, dried	0.02	STMR barley grain	n.a.	
Potato, culls	<0.01	STMR potato	<0.01	HR
Potato, process waste	<0.01	STMR potato	n.a.	
Potato, dried pulp	<0.01	STMR potato	n.a.	

Table B.7.4-2: Calculation of livestock exposure for chlorothalonil

		Maximum dietary burden (mg/kg bw/d)	Median dietary burden (mg/kg bw/d)	Highest contributing commodity	Max dietary burden (mg/kg DM)	Intake >0.004 mg/kg bw/d
Ruminant	Beef cattle	0.055 ~~47~~	0.013	~~Barley~~ Wheat straw	2.30 ~~1.97~~	Yes
	Dairy cattle	0.088 ~~76~~	0.021	~~Barley~~ Wheat straw	2.29 ~~1.96~~	Yes
	Ram/Ewe	0.151 ~~29~~	0.035	~~Barley~~ Wheat straw	4.54 ~~3.88~~	Yes
	Lamb	0.193 ~~64~~	0.044	~~Barley~~ Wheat straw	4.53 ~~3.87~~	Yes
Pig/Swine	Breeding	0.001	0.001	Potato, process waste	0.05	No
	Finishing	0.001	0.001	Potato, culls	0.04	No
Poultry	Broiler	0.002	0.002	Potato, culls	0.02	No
	Layer	0.079 ~~24~~	0.008 ~~9~~	~~Barley~~ Wheat straw	1.15 ~~0.34~~	Yes
	Turkey	0.002	0.002	Potato, culls	0.02	No

Table B.7.4-3: SDS-3701 input values for dietary burden calculation

Commodity	Median dietary burden		Maximum dietary burden	
	Input value (mg/kg)	Comment	Input value (mg/kg)	Comment
Barley, straw	0.10	STMR from NEU	1.1	HR from NEU
Barley, grain	0.02	STMR from NEU	n.a.	
Wheat, straw	0.06 ~~0.07~~	STMR from NEU/SEU ~~(based on 6 trials)~~	0.58 ~~0.12~~	HR from SEU
Wheat, grain	<0.01	STMR	n.a.	
Wheat gluten, meal	<0.01	STMR grain x PF 'wheat, gluten feed meal' (<0.01 x <1)	n.a.	
Wheat, milled by-products	<0.01	STMR wheat grain	n.a.	
Brewer's grain, dried	0.02	STMR barley grain	n.a.	
Distiller's grain, dried	0.02	STMR barley grain	n.a.	
Potato, culls	<0.02	STMR potato	<0.02	HR

Commodity	Median dietary burden		Maximum dietary burden	
	Input value (mg/kg)	Comment	Input value (mg/kg)	Comment
Potato, process waste	<0.02	STMR potato	n.a.	
Potato, dried pulp	<0.02	STMR potato	n.a.	

Table B.7.4-4: Calculation of livestock exposure for SDS-3701

		Maximum dietary burden (mg/kg bw/d)	Median dietary burden (mg/kg bw/d)	Highest contributing commodity	Max dietary burden (mg/kg DM)	Intake >0.004 mg/kg bw/d
Ruminant	Beef cattle	0.011	0.003	Barley straw	0.47	Yes
	Dairy cattle	0.017	0.004	Barley straw	0.45	Yes
	Ram/Ewe	0.027	0.004	Barley straw	0.81	Yes
	Lamb	0.034	0.005	Barley straw	0.79	Yes
Pig/Swine	Breeding	0.002	0.002	Potato, process waste	0.09	No
	Finishing	0.002	0.002	Potato, culls	0.06	No
Poultry	Broiler	0.002	0.002	Potato, culls	0.03	No
	Layer	0.006	0.002	~~Barley~~ Wheat straw	0.09	Yes
	Turkey	0.002	0.002	Potato, culls	0.03	No

Table B.7.4-5: Combined residues of chlorothalonil and SDS-3701, expressed as chlorothalonil, input values for dietary burden calculation

Commodity	Median dietary burden	Maximum dietary burden
	Input value (mg/kg)	Input value (mg/kg)
Barley, straw	1.61	6.88
Barley, grain	0.04	n.a.
Wheat, straw	0.93 ~~1.05~~	10.52 ~~2.23~~
Wheat, grain	<0.02	n.a.
Wheat gluten, meal	<0.01	n.a.
Wheat, milled by-products	<0.02	n.a.
Brewer's grain, dried	0.04	n.a.
Distiller's grain, dried	0.04	n.a.
Potato, culls	<0.03	<0.03
Potato, process waste	<0.03	n.a.
Potato, dried pulp	<0.03	n.a.

Table B.7.4-6: Calculation of livestock exposure for combined residues

		Maximum dietary burden (mg/kg bw/d)	Median dietary burden (mg/kg bw/d)	Highest contributing commodity	Max dietary burden (mg/kg DM)	Intake >0.004 mg/kg bw/d
Ruminant	Beef cattle	0.061 ~~59~~	0.017	~~Barley~~ Wheat straw	2.54 ~~46~~	Yes
	Dairy cattle	0.097 ~~4~~	0.026	~~Barley~~ Wheat straw	2.52 ~~44~~	Yes
	Ram/Ewe	0.164 ~~58~~	0.040	~~Barley~~ Wheat straw	4.91 ~~74~~	Yes
	Lamb	0.207 ~~1~~	0.050	~~Barley~~ Wheat straw	4.87 ~~72~~	Yes
Pig/Swine	Breeding	0.003	0.003	Potato, process waste	0.14	No
	Finishing	0.003	0.003	Potato, culls	0.10	No
Poultry	Broiler	0.004	0.004	Potato, culls	0.05	No
	Layer	0.085 ~~30~~	0.011 ~~2~~	~~Barley~~ Wheat straw	1.25 ~~0.44~~	Yes
	Turkey	0.004	0.004	Potato, culls	0.06	Yes

Table B.7.4-7: Calculation of ratio between chlorothalonil and SDS-3701 in exposure to livestock

		Chlorothalonil Maximum dietary burden (mg/kg bw/d)	SDS-3701 Maximum dietary burden (mg/kg bw/d)	Ratio chlorothalonil : SDS-3701	Chlorothalonil median dietary burden (mg/kg bw/d)	SDS-3701 median dietary burden (mg/kg bw/d)	Ratio chlorothalonil : SDS-3701
Ruminant	Beef cattle	0.055 ~~47~~	0.011	~~4.3~~ 5:1	0.013	0.003	4.3:1
	Dairy cattle	0.088 ~~76~~	0.017	~~4.5~~ 5.2:1	0.021	0.004	5.3:1
	Ram/Ewe	0.151 ~~29~~	0.027	~~4.8~~ 5.6:1	0.035	0.004	8.8:1
	Lamb	0.193 ~~64~~	0.034	~~4.8~~ 5.7:1	0.044	0.005	8.8:1
Pig/Swine	Breeding	0.001	0.002	1:2	0.001	0.002	1:2
	Finishing	0.001	0.002	1:2	0.001	0.002	1:2
Poultry	Broiler	0.002	0.002	1:1	0.002	0.002	1:1
	Layer	0.079 ~~24~~	0.006	~~4~~ 13.2:1	0.008 ~~9~~	0.002	4~~.5~~:1
	Turkey	0.002	0.002	1:1	0.002	0.002	1:1

B.7.4.1 Poultry

No feeding studies were submitted for Annex I listing of chlorothalonil during the initial peer review. Furthermore, no additional studies have been submitted for the sake of the renewal of chlorothalonil. Since the highest exposure according to the maximum dietary burden (0.030 mg/kg bw/d) is 53 times lower than the dose level in the new metabolism study with chlorothalonil, residue levels in poultry commodities are expected to remain below the enforcement LOQ in eggs and tissues. Therefore, no hen feeding study is required.

B.7.4.2 Ruminants

During the initial peer review, no feedings studies with ruminants were available. In the Addendum 14 to the DAR a ruminant feedings study has been evaluated. Furthermore, additional studies are now available.

B.7.4.2.1 Feeding study with chlorothalonil and SDS-3701 in lactating cows

Previous evaluation	In Addendum 14 to DAR. In addition, residue concentrations are summarised per animal for the renewal of chlorothalonil (table B.7.4.2.1-1).
RMS remark	Acceptable

Characteristics

reference	:	Wiedmann and Kenyon (1995)	exposure	:	28 consecutive days
type of study	:	livestock feeding study	dose	:	0, 1.5, 3, 9, 30 mg as/kg feed chlorothalonil
year of execution	:	1994			
test substance	:	15:1 blend of chlorothalonil (purity 98.9%) and SDS-3701(4-hydroxy-2,5,6-trichloro-1,3-dicyano-benzene; purity 99.2%)			0, 0.1, 0.2, 0.6, 2 mg as/kg feed SDS-3701
route	:	oral (diet)	vehicle	:	capsules containing a 15:1 blend of both substances
species	:	lactating cows	GLP statement	:	yes
group size	:	4	guideline	:	no guideline in force

[1] corresponds to 0, 0.065, 0.13, 3.9, 13 mg/kg bw/d for chlorothalonil and 0, 0.004, 0.009, 0.026, 0.087 mg/kg bw/d for SDS-3701 (based on an assumed body weight of 550 kg and a average feed consumption of 24 kg/d)

Study design

Chlorothalonil and its metabolite SDS-3701 were administered orally (as capsules) as a 15:1-blend (93.29% Chlorothalonil, 6.38% SDS-3701) to lactating Holstein cows. Five groups with four animals/group received the following doses once per day over a period of 28 consecutive days: 0, 1.5 mg chlorothalonil/0.1 mg SDS-3701/kg feed, 3 mg chlorothalonil/0.2 mg SDS-3701/kg feed, 9 mg chlorothalonil/0.6 mg SDS-3701/kg feed, 30 mg chlorothalonil/2 mg SDS-3701/kg feed. In total 5 cows were replaced during the study due to low milk production.

Milk was collected twice daily, morning and evening milk was pooled in proportion to the milk production. Composite samples were taken from day 3, 6, 9, 12, 15, 18, 21, 24, 27. Milk samples from day 9, 15, 21 and 27 were separated into cream and skimmed milk. All samples were frozen. Within 24h after the last administration at day 28, animals were sacrificed (zero withdrawal) and samples from round muscle, loin muscle, liver, kidneys, perirenal and omental fat were taken from all animals. The metabolite SDS-3701 in the samples was determined after its derivatisation to its methyl ether applying GC-ECD.

Results

Residue levels in milk collected twice daily over the period of treatment (28 d) reached a plateau level depending on the dose level. A rapid increase in the milk occurred during the first 10 days of dosing. In the group dosed with 1.5 mg chlorothalonil/0.1 mg SDS-3701/kg a plateau was reached within 9 days. In the other groups from day 10 till day 28 the levels still increased slightly, reaching the plateau levels within 24d, 17d and 22d after 3 mg chlorothalonil/0.2 mg SDS-3701/kg feed, 9 mg chlorothalonil/0.6 mg SDS-3701/kg feed and 30 mg chlorothalonil/2 mg SDS-3701/kg feed, respectively.

The maximum levels of SDS-3701 in bovine tissues, milk and milk products are summarised in table B.7.8.1.1.

Table B.7.8.1.1 Maximum residue levels in milk (products) and tissues from dairy cattle dosed for 28 d with a blend of chlorothalonil and SDS-3701

Residue found	Tissue sample	0 mg as/kg feed	1.5/0.1[1)] mg as/kg feed	3.0/0.2 mg as/kg feed	9.0/0.6 mg as/kg feed	30/2.0 mg as/kg feed
SDS-3701 (mg/kg)	whole milk	<0.01	0.04	0.10	0.31	0.65
	cream	<0.01	0.04	0.09	0.19	0.44
	skimmed milk	<0.01	0.04	0.08	0.19	0.42
	muscle (round or loin)	<0.01	n.d.	0.02	0.09	0.24
	fat	<0.01	0.03	0.07	0.08	0.85
	liver	<0.01	0.03	0.04	0.18	0.55
	kidneys	<0.01	0.14	0.28	0.55	1.19

[1)] dose of chlorothalonil/dose of SDS-3701

Conclusions

A feeding study was performed with different concentrations of a 15:1 blend of chlorothalonil/SDS-3701 in lactating cows over a period of 28 days.

After administration of 1.5/0.1 and 3/0.2 mg as/kg chlorothalonil/SDS-3701, respectively, the SDS-3701 residue levels in milk were 0.04 and 0.1 mg/kg. Processing of the milk samples did not result in concentration of SDS-3701 in either of the fractions, skimmed milk and cream. After administration of 1.5/0.1 and 3/0.2 mg as/kg chlorothalonil/SDS-3701, respectively, the highest residue levels were found in the kidneys (0.14 and 0.28 mg/kg); residue levels were much lower in fat (0.03 and 0.07 mg/kg), liver (0.03 and 0.04 mg/kg) and muscle (not detectable and 0.02 mg/kg). The residue levels found in the milk and tissues and organs of groups dosed at higher levels (9.0/0.6 and 30/2.0 mg as/kg chlorothalonil/SDS-3701) were approximately proportional to the administered dose.

Guidelines and limitations

There were no deviations from the guideline (Lundehn document), with the following exception. It was reported that the stability of SDS-3701 in milk, muscle, liver and fat was investigated in a separate study (Ricerca Inc. study number 5927-93-0329). This study showed that under frozen conditions residues of SDS-3701 were stable in whole milk for one year, while low losses (≤17%) were seen in muscle, body fat and liver. The report of the stability study has been submitted and evaluated (see B.7.6.3.2; storage stability of SDS-3701 in cow´s milk and tissues). The report is considered acceptable.

Additional table B.7.4.2.1-1 for RAR: Residue levels for each individual animal in milk and tissues from dairy cattle dosed for 28 d with a blend of chlorothalonil and SDS-3701

Tissue sample	0 mg as/kg feed	1.5/0.1[1)] mg as/kg feed	3.0/0.2 mg as/kg feed	9.0/0.6 mg as/kg feed	30/2.0 mg as/kg feed
Whole milk[2)]	4x <0.01	2x 0.02; 2x 0.03	0.04; 2x 0.07; 0.08	0.15; 2x 0.21; 0.31	0.50; 0.53; 0.54; 0.55
Muscle (round or loin)[3)]	4x <0.01	4x <0.01	3x 0.01; 0.02	Loin: 2x 0.04; 0.05; 0.07 Round: 2x 0.04; 0.05; 0.09	Loin: 0.10; 0.11; 0.15; 0.24 Round: 0.11; 0.14; 2x 0.15
Fat (omental or perirenal)[4)]	4x <0.01	Omental: 2x 0.02; 2x 0.03 Perirenal: 2x 0.01; 2x 0.02	Omental: 0.01; 0.03; 0.05; 0.07 Perirenal: 0.02; 0.03; 2x 0.05	Omental: 0.01; 0.02; 0.05; 0.06 Perirenal: 0.03; 0.06; 0.07; 0.08	Omental: 0.09; 0.14; 0.25; 0.36 Perirenal: 0.47; 0.53; 0.81; 0.85
Liver	4x <0.01	3x 0.02; 0.03	3x 0.02; 0.04	2x 0.13; 2x 0.18	0.37; 0.39; 0.47; 0.55
Kidneys	4x <0.01	2x 0.13; 2x 0.14	0.13; 0.18; 0.22; 0.28	0.39; 0.49; 0.52; 0.55	0.76; 0.89; 0.95; 1.19

[1)] dose of chlorothalonil/dose of SDS-3701

[2)] results for milk are obtained from the day that it was concluded in the results that the plateau was reached, i.e. for dose 1.5/0.1 day 9; for dose 3.0/0.2 day 24; for dose 9/0.6 day 17; and for dose 30/2.0 day 22.

[3)] the highest residues are chosen as input values in the excel-sheet for MRL calculations, i.e. the residues for round muscle for dose 9/0.6; and the residues for loin muscle for dose 30/2.0.

[4)] the residues for perirenal fat are used as input for renal fat in the excel-sheet for MRL calculations; the residues for omental fat are used as input for subcutaneous fat in the excel-sheet for MRL calculations.

B.7.4.2.2 Feeding study with chlorothalonil in beef cattle

Previous evaluation	Submitted for the purpose of renewal
RMS remark	Acceptable

Report:	K-CA 6.4.2/02. Dever M (2008); The determination of tissue residues as measured by the major metabolite 4-hydroxy-2,5,6-trichloro-1,3-dicyanobenzene in liver, kidney, muscle and fat following daily ingestion by cattle of chlorothalonil over a 28 day period. Veterinary Health Research PTY Ltd. Report No SICB1880, Syngenta File No. R044686_11199.

Guidelines

Commission Directive 96/68/EC

US EPA Residue Chemistry Test Guidelines OPPTS 860.1480.

GLP

The study was carried out according to the principles of Good Laboratory Practice.

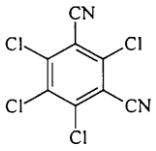
EXECUTIVE SUMMARY

Beef cattle were dosed for 28 consecutive days with chlorothalonil in the feed at a concentration of 0.1, 1.5 or 12.7 mg chlorothalonil/kg bw/day. Three additional cows served as controls.

Within 24 hours of the final dose animals were sacrificed and samples of muscle, liver, kidney, perirenal fat and subcutaneous fat were taken. Two animals from the highest dosing group were sacrificed 14 days after the last dose and samples of round muscle, loin muscle, liver, kidney, perirenal fat and sub-cutaneous fat were taken. Samples were analysed for residues of 4-hydroxy-2,5,6-trichloro-1,3-dicyanobenzene (R182281).

Residues of R182281 were found in all tissues for all dose levels with the exception of muscle and fat samples from the lowest dose level. Highest residues of R182281 were found in liver and kidney; ranging from 0.008 to 1.04 mg/kg and 0.024 to 1.51 mg/kg, respectively, depending on the dose level. Residues of R182281 decreased after a 14 day depuration period, although they were still significant. Residues of R182281 showed a broadly linear relationship to dosing level in animal tissues.

A1. Test Materials

Structure	
Common name	Chlorothalonil
CAS Number	1897-45-6
Batch number	F343 (98.6%)
Stability of test compound	The test substance is assumed to be stable for the duration of the dosing period

A2. Test Facilities

In-life phase	Veterinary Health Research Pty Ltd, New South Wales, Australia.
Analytical phase	Veterinary Health Research Pty Ltd, New South Wales, Australia.

A2. Test Animals

Species	Angus and Angus cross male castrates
Age	18 months
Weight at dosing	400-452 kg

Number of animals	14 (3 for low and medium dose, 5 for high dose, 3 control)
Acclimation period	14 days
Diet and water	Mixed ration calculated daily. Residual feed weighed and discarded daily. Fresh drinking water *ad libitum*
Housing	Indoors, in individual stalls

B. STUDY DESIGN

B1. Experimental conditions

Dosing regime	**Group**	**Treatment**	**Dietary concentration (mg/kg)**	**Dose rate (mg/day)***
	Group 1	Control	0	0
	Group 2	Chlorothalonil	0.1	41.1-44.5
	Group 3	Chlorothalonil	1.5	635 - 698
	Group 4	Chlorothalonil	12	5136 - 5555
Timing	Once per day			
Duration	28 consecutive days			
Depuration	14 days			
Method	Mixed into feed			

*Animals were weighed on a weekly basis and the bodyweight data used to determine the actual dose rate in the following week. The average feed intake data were used to calculate the actual dose rate administered.

B2. Sample Collection

Within 24 hours of the final dose animals were sacrificed and samples of muscle, liver, kidney, perirenal fat and subcutaneous fat were taken. Two animals from the highest dosing group were sacrificed 14 days after the last dose and samples of round muscle, loin muscle, liver, kidney, perirenal fat and sub-cutaneous fat were taken.

All samples were stored frozen for up to 11 months before analysis.

B3. Analytical Phase

Residues of R182281 were determined by an analytical method based on the QuEChERS method: The method involved extraction of the samples with acetonitrile containing 1% acetic acid, followed by the addition of anhydrous magnesium sulphate and sodium acetate and further homogenisation. The extracts were centrifuged and the supernatant removed. The supernatant was evaporated into dryness and re-dissolved in acetonitrile: water (1:1 v/v). Residues of R182281 were determined by LC-MS, using ion *m/z* 245 for quantification and *m/z* 247 for confirmation. The LOQ was 0.005 mg/kg.

II. RESULTS AND DISCUSSION

Recovery data for the method of analysis are presented in Table 7.4.2.2-1. Samples from the feeding study were analysed against matrix matched standards and linearity was demonstrated in liver, kidney, muscle and fat matrices over the concentration range equivalent to 0.005 - 0.075 mg/kg (r^2 > 0.996).

Table 7.4.2.2-1: Summary of recovery data for R182281 in products of animal origin

Commodity	Fortification concentration (mg/kg)	Quantified against matrix matched standards		
		No samples	Mean recovery (%)	RSD (%)
Bovine liver	0.005	7	116	10
	0.01	7	101	10
	0.025	7	103	4
	0.075	7	107	6
Bovine kidney	0.005	7	114	4
	0.01	7	106	4
	0.025	7	89	7
	0.075	7	92	9
Bovine muscle	0.005	7	106	8
	0.01	7	90	9
	0.025	7	85	7
	0.075	7	89	6
Bovine fat	0.005	7	94	14
	0.01	7	87	7
	0.025	7	113	11
	0.075	7	99	11

No residues of R182281 at or above 0.005 mg/kg were found in muscle or fat samples for the lowest dose level.

Maximum residues of R182281 found in the medium and high dose levels were, respectively, 0.01 and 0.06 mg/kg in muscle and 0.026 and 0.078 mg/kg in fat.

Following the lowest dose, maximum residues of R182281 were 0.008 mg/kg in liver and 0.024 mg/g in kidney.

For the medium and highest dose levels, maximum residues of R182281 were, respectively, 0.087 and 1.04 mg/kg in liver, and 0.39 and 1.51 mg/kg in kidney.

For the highest dose level, residues of R182281 decreased after a 14 day depuration period, although were still significant. Based on the residue levels from the lowest dose group, the residue levels in animal tissues from the other dose groups show a reasonably linear relationship.

The results in tissues are summarised in Table 7.4.2.2-2.

Table 7.4.2.2-2: Residues of R182281 in tissues of beef cows

Tissue		R182281 Residue Levels in Tissues (mg/kg)			
		0.1 mg/kg	1.5 mg/kg	12 mg/kg	12 mg/kg (after depuration)
Muscle		3 x <0.005	0.006, <0.005, 0.01	0.051, 0.061, 0.043	0.021, 0.034
	Mean	<0.005	0.007	0.052	0.028
Liver		0.008, <0.005, 0.006	0.071, 0.058, 0.087	0.881, 1.04, 0.323	0.460, 0.486
	Mean	0.006	0.072	0.748	0.473
Kidney		0.024, 0.015, 0.020	0.392, 0.273, 0.332	1.450, 1.510, 1.290	1.310, 1.080
	Mean	0.020	0.332	1.416	1.195
Perirenal Fat		3 x <0.005	0.014, <0.005, 0.008	0.078, 0.049, 0.061	0.020, 0.044
	Mean	<0.005	0.009	0.063	0.032
Sub-cutaneous Fat		3 x <0.005	0.026, 0.016, 0.025	0.052, 0.073, 0.070	0.050, 0.081
	Mean	<0.005	0.022	0.065	0.066

III. CONCLUSIONS

Residues of R182281 were found in all tissues for all dose levels with the exception of muscle and fat samples from the lowest dose level. Highest residues of R182281 were found in liver and kidney; ranging from 0.008 to 1.04 mg/kg and 0.024 to 1.51 mg/kg, respectively, depending on the dose level. Residues of R182281 decreased after a 14 day depuration period, although they were still significant. Residues of R182281 showed a broadly linear relationship to dosing level in animal tissues.

Remark RMS

The substance was administered in the feed. OECD 505 (Jan 2007) requires analysis of the feed to confirm dose concentrations. However, the study was executed in March 2007 and the protocol was inspected in 2006. Moreover, this is not required according to Sanco 7031/VI/95 rev 4. Therefore, the study is considered acceptable.

B.7.4.2.3 Feeding study with chlorothalonil in lactating cows; SDS-3701 levels in milk

Previous evaluation	Submitted for the purpose of renewal
RMS remark	Acceptable

Report:	K-CA 6.4.2/03. Rogers G (2008); The determination of the major metabolite 4-hydroxy-2,5,6-trichloro-1,3-dicyanobenzene residues in bovine milk following daily ingestion of chlorothalonil by lactating dairy cows over a 28 day period. Veterinary Health Research PTY Ltd. Report No SICB1859, Syngenta File No. R044686_11200.

Guidelines

Commission Directive 96/68/EC

US EPA Residue Chemistry Test Guidelines OPPTS 860.1480.

GLP

The study was carried out according to the principles of Good Laboratory Practice.

EXECUTIVE SUMMARY

Dairy cattle were dosed for 28 consecutive days with chlorothalonil in the feed at a concentration of 0.1, 1.5 or 12.7 mg chlorothalonil/kg bw/day. Three additional cows served as controls.

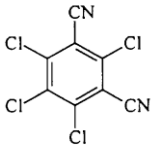
Milk was collected each day in the morning and evening and combined to provide a daily sample for individual cows. The milk from two cows in the highest dose group was collected for a further 14 days after dosing ceased. Samples were analysed for residues of 4-hydroxy-2,5,6-trichloro-1,3-dicyanobenzene (R182281).

Maximum residues of R182281 found in milk for the low, medium and high dose levels were, 1.2, 150 and 231 µg/L, respectively.

Maximum residues of R182281 found in cream for the low, medium and high dose levels were, 17.5, 247 and 336 µg/L, respectively.

For the highest dose level, residues of R182281 in whole milk decreased after a 14 day depuration period, although they were still significant. Residues in cream increased during the depuration period.

A1. Test Materials

Structure	
Common name	Chlorothalonil
CAS Number	1897-45-6
Batch number	F343 (98.6%)
Stability of test compound	The test substance is assumed to be stable for the duration of the dosing period

A2. Test Facilities

In-life phase	A commercial dairy farm, Dayboro 4521, Queensland, Australia.
Analytical phase	Veterinary Health Research Pty Ltd, New South Wales, Australia.

A2. Test Animals

Species	Holstein/Friesian/Jersey
Age	At least 2 years
Weight at dosing	445-655 kg
Number of animals	14 (3 for low and medium dose, 5 for high dose, 3 control)
Acclimation period	14 days
Diet and water	Pasture, *ad libitum* (open grazing paddocks). High energy supplement offered during milking. Fresh drinking water *ad libitum.*
Housing	Open grazing paddocks

B. STUDY DESIGN

B1. Experimental conditions

Dosing regime	Group	Treatment	Dietary concentration (mg/kg)*	Dose rate (mg/kg bodyweight/day)**
	Group 1	Control	0	0
	Group 2	Chlorothalonil	2	0.1
	Group 3	Chlorothalonil	30	1.53
	Group 4	Chlorothalonil	250	12.7
Timing	Once per day			
Duration	28 consecutive days			
Depuration	14 days			
Method	Orally, via syringe, diluted in 1% methylcellulose in water			

* On the assumption of a 550 kg animal consuming 24 kg dry matter/28 kg fresh feed daily.
**Animals were weighed on a weekly basis and the bodyweight data used to determine the actual dose rate in the following week.

B2. Sample Collection

All cows were milked individually morning and evening. The morning and evening milk was amalgamated to give a composite daily sample for each cow. A sub-sample of each day's milk was refrigerated for at least 12 hours to allow separation of cream from milk. Milk samples from two cows from the highest dose group were collected for a further 14 days after the last dose was administered. Following the depuration period all cows were returned to the dairy herd.
All samples were stored frozen for up to 10 months before analysis.

B3. Analytical Phase

Residues of R182281 were determined by an analytical method based on the QuEChERS method: The method involved extraction of the samples with acetonitrile containing 1% acetic acid, followed by the addition of anhydrous magnesium sulphate and sodium acetate and further homogenisation. The extracts were centrifuged and the supernatant removed. The supernatant was evaporated into dryness and re-dissolved in acetonitrile: water (1:1 v/v). Residues of R182281 were determined by LC-MS, using ion *m/z* 245 for quantification and *m/z* 247 for confirmation. The LOQ was 5 µg/L in both milk and cream.

II. RESULTS AND DISCUSSION

Recovery data for the method of analysis are presented in Table 7.4.2.3-1. Samples from the feeding study were analysed against matrix matched standards and linearity was demonstrated in milk and cream matrices over the concentration range equivalent to 5 - 250 µg/L ($r^2 > 0.999$).

Table 7.4.2.3-1: Summary of recovery data for R182281 in milk and cream

Commodity	Fortification concentration (µg/L)	No. samples	Recovery (%)	Mean (%)	RSD (%)
Milk	5	6	105, 104, 105, 102, 103, 95	102	3.7
	10	5	100, 101, 98, 92, 96	97	3.7
	25	6	100, 99, 95, 95, 95, 93	96	2.8
	75	5	89, 84, 86, 87, 84	86	2.5
	250	6	93, 88, 97, 93, 95, 89	93	3.7
	Overall	28	-	95	6.5
Cream	5	6	98, 76, 85, 86, 86, 90	87	8.6
	10	6	99, 96, 90, 97, 98, 97	96	3.3
	25	6	102, 102, 97, 95, 96, 96	98	3.2
	75	6	100, 98, 93, 92, 92, 92	95	3.8
	250	6	87, 97, 92, 99, 96, 97	95	4.7
	Overall	30	-	94	6.2

No residues of R182281 above 5 µg/L were found in control samples.

Maximum residues of R182281 found in milk for the low, medium and high dose levels were, 1.2, 150 and 231 µg/L, respectively.

Maximum residues of R182281 found in cream for the low, medium and high dose levels were, 17.5, 247 and 336 µg/L, respectively.

For the highest dose level, residues of R182281 in whole milk decreased after a 14 day depuration period, although were still significant. Residues in cream increased during the depuration period.

The results are summarised in Table 7.4.2.3-2.

Table 7.4.2.3-2: Residues of R182281 in milk and cream*[1)]

Day		Whole milk residues (µg/L)			Cream residues (µg/L)		
		0.1 mg/kg bw/day	1.5 mg/kg bw/day	12 mg/kg bw/day	0.1 mg/kg bw/day	1.5 mg/kg bw/day	12 mg/kg bw/day
0	Range	3 x <0.2	3 x <0.2	<0.2 – 0.6	3 x < 0.2	3 x <0.2	5 x <0.2
	Mean	<0.2	<0.2	0.3	<0.2	<0.2	<0.2
3	Range	-	-	44.0 – 117	-	-	56.3 – 130
	Mean	-	-	69.8	-	-	107.8
4	Range	<0.2 – 1.2	4.8 – 36.1	46.8 – 136	-	-	-
	Mean	0.6	14.4	86.6	-	-	-
7	Range	<0.2 – 0.3	8.9 – 46.6	67.5 – 216	-	-	33.2 – 210
	Mean	0.3	21.7	126.2	-	-	127.7
11	Range	-	-	58.8 – 231	-	-	66.7 – 233
	Mean	-	-	149.4	-	-	157.7
12	Range	<0.2 – 1.2 (<0.2; 0.9; 1.2)	14.0 - 150	85.3 – 179	-	-	-

Day		Whole milk residues (µg/L)			Cream residues (µg/L)		
		0.1 mg/kg bw/day	1.5 mg/kg bw/day	12 mg/kg bw/day	0.1 mg/kg bw/day	1.5 mg/kg bw/day	12 mg/kg bw/day
	Mean	0.8	59.5	136.1	-	-	-
14	Range	-	-	-	5.1 – 17.5	18.0 - 205	69.6 – 214
	Mean	-	-	-	10.2	99.7	156.1
15	Range	-	-	95.9 – 192 (95.9; 2x 159; 180; 192)	-	-	83.2 – 212
	Mean	-	-	157.2	-	-	173.6
17	Range	<0.2-0.5	11.6 – 37.0	107 – 191	-	-	-
	Mean	0.3	21.8	148.4	-	-	-
19	Range	-	-	106 – 182	-	-	119 – 202
	Mean	-	-	144.8	-	-	169.0
21	Range	<0.2 – 0.4	14.3 – 57.0 (14.3; 17.6; 57.0)	107 – 198	-	-	-
	Mean	0.3	29.6	151.6	-	-	-
23	Range	-	-	99.3 – 180	-	-	46.4 – 226
	Mean	-	-	133.7	-	-	172.1
24	Range	<0.2 - 0.3	18.7 – 46.9	79.1 – 176	-	-	-
	Mean	0.2	29.2	119.7	-	-	-
26	Range	<0.2 – 0.6	13.2 – 55.6	22.9 – 187	-	-	-
	Mean	0.3	29.1	105.1	-	-	-
27	Range	-	-	2.3-215	-	-	62.4 – 232
	Mean	-	-	114	-	-	145.6
28	Range	<0.2 – 0.8	15.9 – 52.9	7.9 – 183	<0.2 – 16.7	29.4 - 247	124 – 237
	Mean	0.5	29.3	119.6	5.7	127.5	167.8
29	Range	-	-	83.0 – 195	-	-	186 – 199
	Mean	-	-	139	-	-	192.5
30	Range	-	-	74.6 – 165	-	-	203 – 269
	Mean	-	-	119.8	-	-	236.0
31	Range	-	-	76.2 – 168	-	-	236 – 282
	Mean	-	-	122.1	-	-	259.0
32	Range	-	-	54.0 – 124	-	-	178 – 323
	Mean	-	-	89.0	-	-	250.5
33	Range	-	-	54.5 -92.2	-	-	245 – 336
	Mean	-	-	73.4	-	-	290.5
34	Range	-	-	41.6 – 90.7	-	-	-
	Mean	-	-	66.2	-	-	-
37	Range	-	-	22.3 – 57.8	-	-	-
	Mean	-	-	40.1	-	-	-
40	Range	-	-	12.4 – 27.9	-	-	-

Day		Whole milk residues (µg/L)			Cream residues (µg/L)		
		0.1 mg/kg bw/day	1.5 mg/kg bw/day	12 mg/kg bw/day	0.1 mg/kg bw/day	1.5 mg/kg bw/day	12 mg/kg bw/day
	Mean	-	-	20.2	-	-	-
42	Range	-	-	8.6 – 18.9	-	-	-
	Mean	-	-	13.8	-	-	-

*< values were used as their numerical value for calculation of mean, i.e. <0.2 = 0.2
[1)] results for milk are obtained from the day that the plateau was reached, i.e. for dose 0.1 day 12; for dose 1.5 day 21; for dose 12 day 15 (in the excel sheet only 4 values can be entered, while there are 5 values for the highest dose: the 4 highest values are selected as a worst-case).

III. CONCLUSIONS

Maximum residues of R182281 found in milk for the low, medium and high dose levels were, respectively, 1.2, 150 and 231 µg/L.

Maximum residues of R182281 found in cream for the low, medium and high dose levels were, respectively, 17.5, 247 and 336 µg/L.

For the highest dose level, residues of R182281 in whole milk decreased after a 14 day depuration period, although they were still significant. Residues in cream increased during the depuration period.

Remark RMS

The substance was administered via syringe. OECD 505 (Jan 2007) requires analysis of the liquid in the syringe to confirm dose concentrations. However, the study was executed in April 2007 and the protocol was inspected in 2006. Moreover, this is not required according to Sanco 7031/VI/95 rev 4. Therefore, the study is considered acceptable.

B.7.4.3 Pigs

The metabolism of chlorothalonil in ruminants was similar to that seen in the rat. Metabolism and feeding studies in pigs are not required, as data for ruminants can be used to address the potential for residues in pigs. See B.7.2.4 for further argumentation.

B.7.4.4 Fish

For the time being there are no agreed test guidelines for the estimation of the dietary burden of pesticide residues for fish or for the design and conduct of fish metabolism studies. Therefore, no fish metabolism studies have been conducted (see also SANCO/10181/2013 rev 2.1). Furthermore, regarding the representative uses, potato, wheat and barley grain are considered potential ingredients for fish feed. Since the low residue levels in these commodities, a significant exposure to fish is unlikely. See B.7.2.5 for further argumentation.

B.7.5 Effects of processing

During the initial peer review of chlorothalonil, the magnitude of chlorothalonil residues in processed crops has been investigated in several field trials. The evaluation of these trials is copied from the DAR into this RAR. However, storage stability has not been assessed in these studies. Furthermore, SDS-3701 has often been measured in these studies, but no transfer factors have been calculated. In addition, SDS-46851 has sometimes been measured, but no transfer factors were calculated. The reliability of these studies is therefore questionable.

In addition, dietary exposure studies were available in the processing paragraph of the original DAR, in which residue levels in crops were analysed after packing, shipping and preparation for sale, and subsequently compared to the residue levels at harvest. These studies have not been copied into the RAR, since these studies are not part of the data requirements and not relevant for the renewal of chlorothalonil. For the sake of the renewal of chlorothalonil, a study investigating the nature of the residue has been submitted. Furthermore, additional studies regarding the magnitude of residues in processed commodities have been submitted for the renewal of chlorothalonil.

B.7.5.1 Nature of the residue

Previous evaluation	Submitted for the purpose of renewal, but already evaluated in an Evaluation Report submitted by The Netherlands for the Article-12 MRL-review, from which the evaluation is copied into this RAR
RMS remark	Acceptable

Characteristics

Reference	:	Grout, S.J., 2002 (IIA 6.5.1-01)	GLP	:	yes
Type of study	:	nature of the residue	Guideline	:	Lundehn (Appendix E, 7035/VI/95 rev. 5)
Year of execution	:	2001-2002	Acceptability	:	acceptable
Test substance	:	[phenyl -^{14}C] chlorothalonil (radiochemical purity ≥98.0%, Batch. 98-54.1 and 98-54.2)			

Study design

The behaviour of [phenyl-^{14}C] chlorothalonil was studied under conditions simulating pasteurisation, baking/brewing/boiling and sterilisation.

Aliquots (~30-50 µL) of a solution of [phenyl-^{14}C] chlorothalonil in acetonitrile were added to tubes (two per treatment) containing 5 mL of ammonium citrate buffer solutions (0.1 M) of pH 4, pH 5 or pH 6 (nominal concentration 5 mg/L). The following incubations were performed (all in the dark):

- ammonium citrate buffer, 20 minutes, 90°C, pH 4
- ammonium citrate buffer, 60 minutes, 100°C, pH 5
- ammonium citrate buffer, 20 minutes, 120°C, pH 6

An artefact was observed in the initial pH 5 and pH 6 experiments using ammonium citrate buffer solutions. This artefact was demonstrated to be a compound which was not formed by hydrolysis but presumably by reaction with ammonium from the ammonium citrate buffer. In order to clarify this

further, additional experiments were performed using different buffer solution (sodium acetate buffer) and/or different incubation conditions. The additional incubations covered the following variants (all in the dark):

- ammonium citrate buffer, 20 minutes, 120°C, pH 4
- ammonium citrate buffer, 20 minutes, 90°C, pH 6
- sodium acetate buffer, 20 minutes, 120°C, pH 6

In all cases control tubes were included under the same conditions of pH, incubation time and buffer type, but incubated at room temperature. After incubation, the radioactivity in the test solutions was quantified by LSC and identified by direct normal and reversed phase TLC. The identity of the artefact (following purification using normal and reversed phase SPE) was confirmed by LC-MS/MS. Temperature was determined during incubation and pH prior to incubation. The recorded values confirmed the target values.

Results

The results are summarised in Table B.7.7.1-01. Mass balances were 95-110%. No degradation was observed in the controls incubated at room temperature. No degradation was observed at pH 4 and 90°C, but at pH 5 and 100°C some degradation occurred (the only degradation product >5% AR was R182281, 19% AR). During incubation at pH 6 and 120°C in acetate buffer, significant degradation occurred, and degradation products >5% AR were R182281 (59% AR) and R613363 (15% AR). During incubation at pH 6 and 120°C in citrate buffer, degradation of chlorothalonil was nearly complete, but in addition to R182281 (48% AR) and R613363 (23% AR), a product was formed identified to be 4-amino-2,5,6-trichloroisophthalonitrile. This product is formed by nucleophilic substitution of the 4-chloride ion with an amino group. This product could not be formed by direct hydrolysis and would not be formed during normal processing conditions. Its formation is the result of using an ammonium citrate salt in the buffer solution. The results from the incubation at pH 6 and 120°C in citrate buffer are therefore not considered in the study conclusion.

Table B.7.7.1-01 Identification of radioactivity after incubation of [phenyl-^{14}C] chlorothalonil under conditions simulating pasteurization, baking/brewing/boiling and sterilization (% AR, duplicate means unless indicated)

pH	temp. (°C)	time (min)	buffer soln	mass balance	chloro-thalonil	R182281	R613636	Artefact (C)	Un-known's	Remain-der
4	90	20	citrate	109	105	1.9	-	-	-	1.7
5	100	60	citrate	107	81	19	3.4	1.4	-	2.3
6	120	20	citrate	110	3.1	48	23	28	5.9(B)	2.9
4	120	20	citrate(A)	95	73	17	2.3	0.4	0.4	1.3
6	90	20	citrate(A)	97	85	5.3	2.8	2.1	0.4	1.0
6	120	20	acetate	103	26	59	15	-	1.7	0.5

(A) Results from a single tube.

(B) Consists of at least 6 discrete components, none >2.4% AR.

(C) Identified to be 4-amino-2,5,6-trichloroisophthalonitrile.

Conclusions

Chlorothalonil was stable under conditions simulating pasteurisation, but showed increased degradation under conditions simulating baking/brewing/boiling and sterilisation. Degradation products under conditions simulating baking/brewing/boiling and sterilisation, respectively, were R182281 (19% and 59% AR) and R613363 (3.4% and 15% AR).

Guidelines & Limitations

The study is acceptable.

B.7.5.2 Distribution of the residue in peel and pulp

The distribution of the residues in peel/pulp is not relevant for barley and wheat. Tomato is separated into peel and pulp during processing. Studies in tomato, including measurements of distribution of the residue in peel and pulp, are presented below in B.7.5.3. Potatoes can be peeled during processing, however, as residues in whole tubers were <LOQ for both chlorothalonil and R182281, processing data are not required.

B.7.5.3 Magnitude of residues in processed commodities

B.7.5.3.1 Processing of potatoes

Previous evaluation	In DAR
RMS remark	Acceptable

Characteristics

reference	:	Zeneca, King et al., 1993	treatment	:	field application
type of study	:	processing study	rate	:	5 applications of 1.13 kg a.i/ha and 4 applications of 11.3 kg a.i/ha
year of execution	:	1992	formulation	:	Bravo 720 (54.0% chlorothalonil)
test substance	:	chlorothalonil	GLP statement	:	yes
crop/commodity	:	potatoes	guidelines	:	not applicable

Study design

Field grown potato plants (Superiors) were treated with Bravo 720 during the growing season; five times with 1.13 kg a.i./ha and subsequently four times with the exaggerated rate of 11.3 kg a.i/ha (approximately ten times the intended use rate). The potatoes were harvested 21 days after the last application and either analysed for the presence of residues or processed. Processing comprised washing and peeling. The peels were filtered off (wet peels) and dried (dry peels). The peeled potatoes were subdivided and either french fried, chipped, or prepared into potato granules. Samples from the processing efforts were homogenized by chopping, mixing by hand and grinding, and processed/ analysed.

Results and conclusions

Chlorothalonil and SDS-46851 levels all remained below the LOD of 0.01 and 0.03 mg/kg, respectively, in potato tubers (unwashed raw agricultural commodity), washed tubers, french fries, wet and dry peels, chips and potato granules. Only trace amounts of SDS-3701 were detected in wet and dry peels, whilst SDS-3701 levels were below the LOD of 0.01 mg/kg in all other matrixes.

Remark RMS: No robust processing factors can be derived, since residues were almost always below LOD.

B.7.5.3.2 Processing of green beans and tomatoes

Previous evaluation	In DAR
RMS remark	Acceptable

Characteristics

reference	:	Zeneca, Marks, 1983	treatment	:	fortified
type of study	:	processing study	rate	:	-
year of execution	:	1979	formulation	:	acetone
test substance	:	^{14}C-uniformly ring labelled chlorothalonil (specific activity 3.8 x 10^4 dpm/µg, radiochemical purity 99.2%)	GLP statement	:	no
crop/commodity	:	green beans and potatoes	guidelines	:	not applicable

Study design

The effect of cooking on chlorothalonil levels in water, green beans, and tomatoes was evaluated. Samples of water, fresh chopped green beans or tomatoes were fortified with ^{14}C-uniformly labelled chlorothalonil (specific activity 3.8 x 10^4 dpm/µg, radiochemical purity 99.2%) and cooked by boiling for 10 minutes under each of the following types of cooking conditions:

- 600 ml open beaker
- 500 ml flat bottom flask equipped with a condenser for refluxing
- 500 ml flat bottom flask equipped with a distillation column
- 6 quart pressure cooker loosely covered
- 6 quart pressure cooker sealed

Samples were analysed for the presence of residues by extraction, partitioning, and LSC or TLC/autoradiography.

Results

The results are summarized in Table 7.7.1.2.

Table 7.7.1.2 Effect of cooking on chlorothalonil levels in water, tomatoes and green beans

Cooking conditions	Loss (%)		
	water	**tomatoes**	**green beans**
Open beaker	96.8	98.0	93.8
Reflux	0.6	0.0	0.0
Distillation	0.0	7.3	4.7
Pot without cover		84.9[1]	93.0[1]
Pot with cover		88.8[1]	89.0[1]

1 measurement in triplicate

Conclusions

The typical cooking operations with open or loosely sealed vessels resulted in a loss through volatilization of 85-98% of the chlorothalonil amended to water, tomatoes or green beans. The chlorothalonil was found to degrade to varying degree to metabolites such as SDS-3701 and SDS-19221 and to trace amounts of uncharacterized degradation products. When chlorothalonil-fortified tomatoes or green beans were cooked in a tightly-sealed pressure cooker, no loss or degradation of chlorothalonil was detected.

B.7.5.3.3 Processing of snapbeans

Previous evaluation	In DAR
RMS remark	Acceptable

Characteristics

reference	:	Zeneca, Ballee et al., 1980	treatment	:	field application
type of study	:	processing study	rate	:	-
year of execution	:	1979	formulation	:	Bravo 6F (54% chlorothalonil) and Bravo 500 (40.4% chlorothalonil)
test substance	:	chlorothalonil	GLP statement	:	no
crop/commodity	:	snapbeans	guidelines	:	not applicable

Study design

Field grown snapbean plants were either treated with Bravo 6F or with Bravo 500. At the normal ripening stage the snapbeans were harvested and transported to a processing factory, where they

were either air cleaned, washed, blanched, and canned, or washed twice, sliced, chopped, and steam/water blanched before freezing. Samples of the snapbeans from the various processing stages were assayed in duplicate for chlorothalonil and SDS-3701 levels.

Results

The chlorothalonil levels, and the respective transfer factors are presented in tables 6.7.1.3.1 and 6.7.1.3.2. SDS-3701 levels were below the LOD of 0.01 mg/kg in all samples.

Table 7.7.1.3.1 Chlorothalonil levels in snapbean samples collected during the processing for canning and concomitant transfer factors

Product	Chlorothalonil (mg/kg)	transfer factor
Load sample	0.84	-
After air cleaning	0.54	0.64
After washing	<0.01	<0.012
After blanching	<0.01	<0.012
After canning	<0.01	<0.012

Table 7.7.1.3.2 Chlorothalonil levels in snapbean samples collected during the processing for freezing and concomitant transfer factors

Product	Chlorothalonil (mg/kg)	transfer factor
Load sample	0.78	-
Beans after first wash[1]	0.16	0.21
After second wash	0.09	0.12
Sliced and chopped	0.10	0.13
After steam or water blanching	<0.01	<0.013

1 waste included after first wash

Conclusions

Canned and frozen snapbeans contained no detectable levels of chlorothalonil or SDS-3701.

B.7.5.3.4 Processing in cherries

Previous evaluation	In DAR
RMS remark	Acceptable

Characteristics

reference	:	Zeneca, Stallard et al., 1977	treatment	:	field application
type of study	:	processing study	rate	:	6 applications at 2.52 kg a.i/ha
year of execution	:	1977	formulation	:	Bravo 6F (54% chlorothalonil)
test substance	:	chlorothalonil	GLP statement	:	no
crop/commodity	:	cherries	guidelines	:	not applicable

Study design

Field grown cherries were treated six times with Bravo 6F at a rate of 2.52 kg a.i./ha. The cherries were harvested 7 days after the last application and held in a cooler at 4°C until processing. The cherries were washed with cold water for two hours, pitted and canned either with or without sugar and samples were collected and analysed to determine the chlorothalonil and SDS-3701 levels.

Results

The chlorothalonil levels in cherry samples collected during processing are presented in Table 7.7.1.4. SDS-3701 levels were at or below the LOD of 0.01 mg/kg in all samples.

Table 7.7.1.4 Chlorothalonil levels in cherry samples collected during processing and concomitant transfer factors

Product	Chlorothalonil (mg/kg)	transfer factor
Whole cherries- field sample	2.74	-
Washed- unpitted cherries	0.52	0.19
Washed- pitted cherries	0.38	0.14
Canned cherries (with water)	0.03	0.01
Canned cherries (with 25% sugar water)	0.03	0.01

Conclusions

Chlorothalonil or SDS-3701 were not concentrated into canned products during the processing of cherries. Major residue reductions occurred at each step of processing.

B.7.5.3.5 Processing in peaches

Previous evaluation	In DAR
RMS remark	Acceptable

Characteristics

reference	:	Zeneca, Ballee et al., 1977	treatment	:	field application
type of study	:	processing study	rate	:	six applications of 2.52 kg a.i/ha
year of execution	:	1976	formulation	:	Bravo 6F (54% chlorothalonil)
test substance	:	chlorothalonil	GLP statement	:	no
crop/commodity	:	peaches	guidelines	:	not applicable

Study design

Field grown peaches were treated six times with Bravo 6F at a rate of 2.52 kg a.i./ha. The peaches were harvested 7 days after the last application. The peaches received an initial cold water wash, followed by a caustic bath peeling wash (18% NaOH- 70°C). The peaches were subsequently spray washed and puree was canned. The collected samples were analysed for chlorothalonil and SDS-3701 levels.

Results

The chlorothalonil and SDS-3701 levels in peach samples, collected during processing, are presented in Table 7.7.1.5.

Table 7.7.1.5 Chlorothalonil residues in peach samples collected during processing and concomitant transfer factors

Product	Chlorothalonil (mg/kg)	SDS-3701 (mg/kg)	transfer factor Chlorothalonil
Whole peaches- field sample	12.9	n.r	-
Whole peaches after water wash	5.86	n.r	0.45
Whole peaches after caustic bath	0.21	0.13	0.02
Canned peach puree	<0.01	<0.01	<0.0008

n.r not reported

Conclusions

Major reductions in chlorothalonil levels occurred during the processing of peaches. No chlorothalonil or SDS-3701 was detected in the final product (canned peach puree).

B.7.5.3.6 Processing in grapes

Previous evaluation	In DAR
RMS remark	Acceptable

Characteristics

reference	:	Zeneca, Prince et al., 1994	treatment	:	field application
type of study	:	processing study	rate	:	7 applications of 3.3 kg a.i/ha
year of execution	:	1993	formulation	:	Bravo 720 (53.8% chlorothalonil)
test substance	:	chlorothalonil	GLP statement	:	yes
crop/commodity	:	grapes (White Riesling)	guidelines	:	not applicable

Study design

Grapes (White Riesling) were treated with Bravo 720, three times during (pre-)bloom and four times at fruit maturation at a rate of 3.3 kg a.i/ha/application (2-3 times the rate proposed for Europe) at 7-10 day intervals. The grapes were harvested 30 days after the last application. The grapes were either processed into raisins, wet and dry pomace, or into juice and samples were analysed for chlorothalonil and SDS-3701 levels.

Results

The chlorothalonil residues in samples collected during processing are presented in Table 7.7.1.6.

Table 7.7.1.6 Chlorothalonil residues in samples collected during the processing of grapes and concomitant transfer factors

Product	Chlorothalonil (mg/kg)	transfer factor	SDS-3701 (mg/kg)	transfer factor
Grapes	9.42	-	0.03	-
Raisins	4.22	0.45	0.02	0.67
Wet Pomace	15.30	1.62	0.03	1.0
Dry Pomace	12.32	1.31	0.12	4.0
Fresh juice	2.20	0.23	<0.01	<0.33

Conclusions

Some concentration of chlorothalonil occurred during the processing of grapes into wet and dry pomace. A reduction in chlorothalonil occurred during the processing into raisins and into fresh grape

juice. During processing of grapes into wet pomace, the SDS-3701 levels remained unchanged. SDS-3701 levels markedly increased during processing into dry pomace. No SDS-3701 was detected in fresh grape juice and a slight reduction in SDS-3701 levels occurred during processing into raisins.

B.7.5.3.7 Processing in apples

Previous evaluation	In DAR
RMS remark	Acceptable

Characteristics

reference	: Zeneca, Kenyon et al., 1983	treatment	:	field application
type of study	: processing study	rate	:	a single application of 19 kg a.i/ha or 3.8 kg a.i/ha
year of execution	: 1982	formulation	:	Bravo 500 (40.4% chlorothalonil)
test substance	: chlorothalonil	GLP statement	:	no
crop/commodity	: apples	guidelines	:	not applicable

Study design

Apple trees were treated with Bravo 500 according to the proposed use of a single early season application of 19 kg a.i/ha at the 0.6 cm green stage of bud expansion or with a single 3.8 kg ai/ha treatment 7 days before harvest (spike). An untreated plot was also included. Three separate lots of apples were processed into frozen apple slices, apple sauce, apple juice, hammer milled apples and apple cider and also into apple peels/cores and apple pomace potentially used for cattle feed. Two samples of each product were analysed for chlorothalonil and SDS-3701.

Results

The chlorothalonil levels in samples collected during the processing of apples are presented in Table 7.7.1.7. SDS-3701 levels were all around or below the LOD of 0.01 mg/kg, except in dry pomace, in which levels up to 0.08-0.09 mg/kg were determined.

Table 7.7.1.7 Chlorothalonil levels in apple samples collected during processing and concomitant transfer factors

Product	**Bravo SAT**[1] **(mg/kg)**	**transfer factor**	**Bravo exaggerated rate (mg/kg)**	**transfer factor**
Apples (unwashed)	<0.01	-	4.49	-
Apples (washed)	<0.01	n.a	3.82	0.85

Product	Bravo SAT[1] (mg/kg)	transfer factor	Bravo exaggerated rate (mg/kg)	transfer factor
Apple slices	<0.01	n.a	0.17	0.04
Unsweetened applesauce	<0.01	n.a	0.02	0.004
Sweetened applesauce	<0.01	n.a	0.02	0.004
Wet pomace	0.04	> 4.0	13.01	2.9
Dry pomace	0.19	> 19.0	45.55	10.1
Apple juice (canned)	<0.01	n.a	0.10	0.02
Hammer milled apples	0.03	>3.0	4.23	0.94
Apple cider	<0.01	n.a	2.95	0.66
Peels/cores	0.03	> 3.0	17.76	4.0

n.a not applicable

1 single early season application

Conclusions

The processing of chlorothalonil treated apples into wet and dry pomace resulted in a 3 to 4 and 10 to 20 fold increase in chlorothalonil levels, respectively. In addition, peels/cores contained approximately 3 to 4 times higher levels of chlorothalonil when compared to the raw (unwashed) agricultural product. SDS-3701 was only detected in dry pomace obtained from spike-treated apples. Chlorothalonil levels were reduced by the processing of apples into apple slices, apple sauce, and apple juice (transfer factors of 0.04, 0.004, and 0.02, respectively).

B.7.5.3.8 Processing in wheat grain

Previous evaluation	In DAR
RMS remark	Acceptable

Characteristics

reference	:	Zeneca, Stallard et al., 1983	treatment	:	field application
type of study	:	processing study	rate	:	3 applications of 5.1 kg a.i/ha

year of execution	:	1977	formulation	:	Bravo 6F (54% chlorothalonil)
test substance	:	chlorothalonil	GLP statement	:	no (a Quality Assurance Statement was included in the report)
crop/commodity	:	wheat grain	guidelines	:	not applicable

Study design

Wheat was treated three times with Bravo 6F at a rate of 5.1 kg a.i/ha/application. The wheat was harvested 24 days after the last application. The wheat was milled, resulting in reduction flour (53%), break flour (13%), bran (20%), shorts (11%) and a milling loss (3%). Duplicate subsamples were analysed for chlorothalonil and SDS-3701.

Results and conclusions

No chlorothalonil or SDS-3701 were detected in grain samples (LOD: 0.01 mg/kg for both compounds) or in any processed product (LOD: 0.03 mg/kg for both compounds).

Remark RMS: No robust processing factors can be derived, since residues were almost always below LOD.

B.7.5.3.9 Processing in tomatoes

Previous evaluation	In DAR
RMS remark	Acceptable

Characteristics

reference	:	Zeneca, Szalkowski et al., 1980	treatment	:	field application
type of study	:	processing study	rate	:	7 applications of 2.5 kg a.i/ha or 5 kg a.i/ha
year of execution	:	1980	formulation	:	Bravo 500 (40.4% chlorothalonil)
test substance	:	chlorothalonil	GLP statement	:	no
crop/commodity	:	tomatoes	guidelines	:	not applicable

Study design

Two plots of tomato plants were treated with a total of 7 applications of chlorothalonil, formulated as Bravo 500. The application schedule was initiated at the flowering stage and continued through maturity. The application rates amounted to 2.5 kg a.i/ha (maximum recommended rate) for one plot and to 5 kg a.i/ha (exaggerated rate) for the second plot. An untreated plot was also included. The treated tomatoes were harvested immediately after the last application. Three separate lots of tomatoes were processed into pomace, juice and paste. Two samples of each product were analysed for chlorothalonil and SDS-3701.

Results

The chlorothalonil levels in samples collected during the processing of tomatoes are presented in

Table 7.7.1.9. SDS-3701 levels were below 0.05 mg/kg in all samples.

Table 7.7.1.9 Chlorothalonil levels (in mg/kg) in samples collected during the processing of tomatoes and concomitant transfer factors

Product	Bravo recommended rate	transfer factor	Bravo exaggerated rate	transfer factor
Tomatoes (unwashed)	2.51	-	4.69	-
Tomatoes (washed)	0.65	0.26	1.20	0.26
Pomace	2.23	0.89	3.82	0.81
Juice	0.02	0.008	0.78	0.17
Condensate	<0.0003	<0.0001	<0.0003	<0.00006
Paste	<0.01	<0.004	0.02	0.004

Conclusions

The processing of tomatoes into various products resulted in lower chlorothalonil levels compared to the raw (unwashed) agricultural product.

B.7.5.3.10 Processing in cucumbers

Previous evaluation	In DAR
RMS remark	Acceptable

Characteristics

reference	:	Zeneca, King et al., 1987	treatment	:	field application
type of study	:	processing study	rate	:	two applications of 2.7 kg a.i/ha
year of execution	:	1985	formulation	:	Bravo 500 (40.4% chlorothalonil)
test substance	:	chlorothalonil	GLP statement	:	yes
crop/commodity	:	cucumbers	guidelines	:	not applicable

Study design

Cucumber plants were treated twice with Bravo 500 at the recommended rate of 2.7 kg a.i/ha/application. The cucumbers were harvested 12 hours after the last application. The cucumbers

were washed twice, sprayed, sliced and brined, and pickles and juice were canned (both cold and hot). Samples from each product were analysed for chlorothalonil, SDS-3701 and SDS-46851.

Results

The chlorothalonil levels determined in the individual fractions resulting from the processing of cucumbers are presented in Table 7.7.1.10. SDS-3701 levels were close to or below 0.01 mg/kg in all samples. SDS-46851 was only detected occasionally.

Table 7.7.1.10 Chlorothalonil levels in samples collected during the processing of cucumbers and concomitant transfer factors

Product	Chlorothalonil (mg/kg)	transfer factor
Field cucumbers	1.32	-
Washed cucumbers	0.71	0.54
Post rinse	0.52	0.39
Brined sliced pickle	0.38	0.29
Brine solution	0.0069	0.005
Cold canned pickles	0.11	0.08
Cold canned juice	0.0065	0.005
Hot canned pickles	0.02	0.015
Hot canned juice	<0.0005	<0.0004

Conclusions

There was no evidence of concentration of chlorothalonil into any processing fraction. The amount of chlorothalonil residue found on cucumbers declined 98% when cucumbers were processed according to normal commercial practice.

B.7.5.3.11 Processing in carrots

Previous evaluation	In DAR
RMS remark	Acceptable

Characteristics

reference	:	Zeneca, King et al., 1990	treatment	:	field application
type of study	:	processing study	rate	:	11 applications of 1.7 kg a.i/ha or of 16.8 kg a.i/ha
year of execution	:	1988	formulation	:	Bravo 720 (54% chlorothalonil)
test substance	:	chlorothalonil	GLP statement	:	yes
crop/commodity	:	carrots	guidelines	:	not applicable

Study design

Two plots of carrots were treated with a total of 11 applications of chlorothalonil, formulated as Bravo 720. The application rates amounted to 1.7 kg a.i/ha (recommended rate) in one plot and 16.8 kg a.i/ha (exaggerated rate) in the second plot respectively. An untreated plot was also included. The treated carrots were harvested immediately after the last application. Three separate lots of carrots were peeled, cooked, pureed and canned. Two samples of each product were analysed for chlorothalonil, SDS-3701, and SDS-46851.

Results

Chlorothalonil levels of 0.04 and 2.23 mg/kg were determined in the raw agricultural product upon recommended and exaggerated rate treatments, respectively. The SDS-3701 levels in the raw agricultural product were below the LOD of 0.01 mg/kg upon recommended rate treatment and 0.06 mg/kg upon exaggerated rate treatment. SDS-46851 was not detected in the raw agricultural product. In processed fractions, none of the compounds exceeded the LOD (Chlorothalonil and SDS-3701: LOD of 0.01 mg/kg; SDS-46851: LOD of 0.03 mg/kg).

B.7.5.3.12 Processing in citrus

Previous evaluation	In DAR
RMS remark	Acceptable

Characteristics

reference	:	Zeneca, Ballee et al., 1980	treatment	:	field application
type of study	:	processing study	rate	:	see Table 7.7.12.1
year of execution	:	1973	formulation	:	Bravo 6F (54% chlorothalonil)
test substance	:	chlorothalonil	GLP statement	:	no
crop/commodity	:	limes, oranges, grapefruit and lemons	guidelines	:	not applicable

General study design

Limes, oranges, grapefruit and lemons were all treated with Bravo 6F. Table 7.7.1.12.1 summarizes information concerning the treatment rate, the number of applications and pre-harvest intervals (PHI):

Table 7.7.1.12.1 Treatment rate, number of applications and pre-harvest intervals citrus fruit

Citrus crop	Application rates (kg a.i/ha)	No. of applications	PHI (days)
limes	17 25	2 2	18 18
oranges	11.8 17.6	3 3	10 10
grapefruit	9.3 13.9	2 2	251 251
lemons	16.8 25.3	2 2	110 110

Following harvest, the fruits were processed into various products and samples of unwashed and washed whole fruit, chopped peel (not for lemons), chopped peel frits (oranges and limes only), finisher pulp (oranges and limes only), juice, peel liquor (oranges and limes only), molasses (oranges and limes only), cold pressed oil and dried pulp and peel were analysed for chlorothalonil and SDS-3701.

Results and conclusions

The ranges in chlorothalonil levels in samples resulting from the processing of citrus are presented in Table 7.7.1.12.2.

Table 7.7.1.12.2 Chlorothalonil ranges (in mg/kg) in samples collected during the processing of citrus and concomitant transfer factors

Product	Bravo recommended rate	maximal transfer factor	Bravo exaggerated rate	maximal transfer factor
Whole fruit (unwashed)	<0.01-3.53	-	<0.01-6.22	-
Whole fruit (washed)	<0.01-1.25	>1.0 (lemons)	<0.01-1.23	n.a.

Product	**Bravo recommended rate**	**maximal transfer factor**	**Bravo exaggerated rate**	**maximal transfer factor**
Chopped peel	<0.01-0.25	<0.33 (oranges)	<0.01-0.47	0.08 (limes)
Peel frits	0.01-0.90	0.33 (oranges)	0.16-1.34	0.22 (limes)
Finisher pulp	<0.01-0.01	<0.33 (oranges)	0.01	0.003 (oranges)
Juice	<0.01	<0.33 (oranges)	<0.01	<0.003 (oranges)
Peel liquor	<0.01	<0.33 (oranges)	0.01-0.03	0.005 (limes)
Molasses	<0.01-0.03	<0.33 (oranges)	<0.01-0.06	0.01 (limes)
Cold pressed oil	0.04-10.6	>4.0 (grapefruits and lemons)	0.04-192	30.9 (limes)
Dried pulp and peel	<0.01-0.05	>1.0 (lemons)	<0.01-0.10	n.a.

n.a. not applicable

The major part of the chlorothalonil present on the whole citrus fruits was removed by the initial washing process. The remaining chlorothalonil on the fruit was concentrated in the cold pressed citrus oil. No chlorothalonil was detected in any citrus juice and dried pulp and peel contained trace amounts of residues. SDS-3701 levels were around or below 0.04 mg/kg for all products, except for cold pressed oil, which contained up to 0.24 mg SDS-3701/kg. SDS-3701 was not determined in the raw agricultural products.

B.7.5.3.13 Processing in peanuts

Previous evaluation	In DAR
RMS remark	Acceptable

Characteristics

reference	:	Zeneca, Kenyon et al., 1986	treatment	:	field application
type of study	:	processing study	rate	:	11 or 13 applications of 1.24 kg a.i/ha
year of execution	:	1985	formulation	:	Bravo 500 (40.4% chlorothalonil)
test substance	:	chlorothalonil	GLP statement	:	yes
crop/commodity	:	peanuts	guidelines	:	not applicable

Study design

Two plots of peanuts were treated with either a total of 11 (recommended treatment) or 13 (exaggerated treatment) applications of chlorothalonil, formulated as Bravo 500 at a rate of 1.24 kg a.i/ha/application. An untreated plot was also included. The treated peanuts were harvested 16 (recommended use) and 6 days (exaggerated treatment) after the last application. Three separate lots of peanuts were shelled and pressed into crude oil and presscake. The presscake and crude oil were expelled into crude oil and press cake and into refined oil and soapstock respectively. Two samples of each product were analysed for chlorothalonil, SDS-3701, and SDS-46851.

Results

The chlorothalonil levels in samples collected during the processing of peanuts are presented in Table 7.7.1.13. SDS-3701 levels were around 0.1 mg/kg in hulls following both treatment regimes and showed no concentration upon processing. SDS-46851 levels amounted up to 0.3 mg/kg in peanut nutmeats and hulls following both treatments and also showed no concentration upon processing.

Table 7.7.1.13 Chlorothalonil levels (in mg/kg) in samples collected during the processing of peanuts and concomitant transfer factors

Product	Bravo recommended rate	transfer factor	Bravo exaggerated rate	transfer factor
Peanut nutmeats (directly from field)	<0.01	-	0.01	-
Peanut hulls (directly from field)	0.29	-	0.40	-
Peanut nutmeats (just prior to processing)	0.03	-	0.02	-
Peanut hulls (just prior to processing)	0.25	-	0.19	-

Product	Bravo recommended rate	transfer factor	Bravo exaggerated rate	transfer factor
Peanut nutmeats (processing fractions)	<0.01	n.a	0.02	1.0
Peanut hulls (processing fractions)	0.58	2.3	0.22	1.16
Presscake (expeller)	<0.01	n.a	0.01	0.5
Presscake (solv. ext.)	<0.01	n.a	<0.01	<0.5
Peanut crude oil (expeller)	<0.01	n.a	<0.01	<0.5
Peanut crude oil (solv. ext.)	0.02	>2.0	0.01	0.5
Peanut refined oil	<0.01	n.a	<0.01	<0.5
Peanut soapstock	<0.01	n.a	<0.01	<0.5

n.a not applicable

Conclusions

There was no concentration of chlorothalonil residues upon processing.

B.7.5.3.14 Processing in plums

Previous evaluation	In DAR
RMS remark	Acceptable

Characteristics

reference	:	Zeneca, Prince et al., 1993	treatment	:	field application
type of study	:	processing study	rate	:	4 or 5 applications of 3.4 kg a.i/ha (Bravo 720) or 5 applications of 1.42 kg a.i/ha (Bravo 825)
year of execution	:	1992	formulation	:	Bravo 720 (53.6% chlorothalonil) and Bravo 825 (81.2% chlorothalonil)
test substance	:	chlorothalonil	GLP statement	:	yes
crop/commodity	:	plums	guidelines	:	not applicable

Study design

Plum trees were either treated with 4 or with 5 applications of Bravo 720 at a rate of 3.4 kg a.i/ha/application with 132 day and 99 day pre-harvest intervals (PHI). Additionally, another plot of plum trees was treated with 5 applications of Bravo 825 (batch: ASC-66518-0101-1207) at a rate of 1.42 kg a.i/ha/application with a PHI of 99 days. Four separate lots of plums were processed into dry prunes and reconstituted prunes. Two samples of each product were analysed for chlorothalonil and SDS-3701.

Results

The chlorothalonil levels measured in the individual fractions and the transfer factors are presented in Table 7.7.1.14. The prunes from plums treated with Bravo 720 and Bravo 825 and harvested 99 days after the last application (5 weeks after shuck-split) were over-dried before they were reconstituted. Sufficient fruit for a second attempt was only available for the plums treated with Bravo 825. SDS-3701 levels were all below the LOD of 0.01 mg/kg.

Table 7.7.1.14 Total chlorothalonil levels (in mg/kg) and concomitant transfer factors in plums and processed products

Product	**Bravo 720 4 appl./ PHI 132 days**	**maximal transfer factor[1]**	**Bravo 720 5 appl.[2]/ PHI 99 days**	**transfer factor[1]**	**Bravo 825 5 appl./ PHI 99 days**	**transfer factor[1]**
Raw plums	<0.01-0.05	-	0.01-0.03	-	<0.01	-
Raw plums (second					<0.01	-

Product	Bravo 720 4 appl./ PHI 132 days	maximal transfer factor[1]	Bravo 720 5 appl.[2]/ PHI 99 days	transfer factor[1]	Bravo 825 5 appl./ PHI 99 days	transfer factor[1]
process)[3]						
Dry prunes	<0.01	<0.2	<0.01	<0.33-<1.0	<0.01	n.a
Dry prunes (second process)[3]					<0.01	n.a
Reconst. prunes	<0.01	<0.2	<0.01	<0.33-<1.0	<0.01	n.a
Reconst. prunes (second process)[3]					<0.01	n.a

1 the mean residue concentration was used to calculate the transfer factor

2 atypical set of prunes, due to over-drying

3 a second analysis was performed due to an over-dried first lot

n.a not applicable

Conclusions

Chlorothalonil levels on raw plums ranged from 0.01-0.05 mg/kg. No residues were detected in any of the processing products.

B.7.5.3.15 Processing in winter squash

Previous evaluation	In DAR
RMS remark	Acceptable

Characteristics

reference	:	Zeneca, King et al., 1990	treatment	:	field application
type of study	:	processing study	rate	:	eleven applications of 2.5 kg a.i/ha
year of execution	:	1988	formulation	:	Bravo 720 (54% chlorothalonil)
test substance	:	chlorothalonil	GLP statement	:	yes
crop/commodity	:	winter squash	guidelines	:	not applicable

Study design

Field grown winter squash plants were treated eleven times with Bravo 720 at a rate of 2.5 kg a.i/ha/application. The winter squash was harvested 1.75 to 2.75 hours after the last application. The winter squash was peeled, milled and partially cooked (squash finisher). Duplicate subsamples were analysed for chlorothalonil, SDS-3701 and SDS-46851.

Results

The chlorothalonil levels in samples collected during the processing of winter squash are presented in Table 7.7.1.15. SDS-3701 and SDS-46851 levels were 0.02 and 0.06 mg/kg in the raw agricultural product and showed no concentration upon processing.

Table 7.7.1.15 Chlorothalonil levels in samples collected during the processing of winter squash and concomitant transfer factors

Product	Chlorothalonil (mg/kg)	transfer factor
Squash	3.23	-
Peeled squash	<0.01	<0.003
Milled squash	<0.01	<0.003
Squash finisher	<0.01	<0.003
Finisher waste	<0.01	<0.003
Squash waste	0.15	0.05
Baby food	<0.01	<0.003

Conclusions

No concentration of chlorothalonil residues occurred in any of the processing fractions.

B.7.5.3.16 Processing in soybeans

Previous evaluation	In DAR
RMS remark	Acceptable

Characteristics

reference	:	Zeneca, Kenyon et al., 1987	treatment	:	field application
type of study	:	processing study	rate	:	3 or 4 applications of 1.68 kg a.i/ha
year of execution	:	1986	formulation	:	Bravo 720 (53.7% chlorothalonil)
test substance	:	chlorothalonil	GLP statement	:	yes
crop/commodity	:	soybeans	guidelines	:	not applicable

Study design

Field grown soybean plants were either treated with a total of 3 (recommended treatment) or 4 (spike treatment) applications of chlorothalonil, formulated as Bravo 720. The application rate amounted 1.68 kg a.i/ha/application. An untreated plot was also included. The treated soybeans were harvested 68 and 13 days after the last application.

Three separate lots of soybeans were dried and then separated into kernels and hulls. The kernels were processed by solvent extraction into meal, oils (crude and refined) and soapstock. Samples were analysed for chlorothalonil, SDS-3701 and SDS-46851.

Results

The chlorothalonil levels in samples collected during the processing of soybeans are presented in Table 7.7.1.16. SDS-3701 and SDS-46851 levels were below 0.05 and 0.1 mg/kg, respectively, in all edible products.

Table 7.7.1.16 Chlorothalonil levels (in mg/kg) in samples collected during the processing of soybeans and concomitant transfer factors

Product	Bravo recommended use	transfer factor	Bravo exaggerated treatment	transfer factor
Whole soybeans (pre-drying)	<0.01	-	0.02	-
Whole soybeans (post-drying)	<0.01	n.a	0.02	1.0
Soybean kernels	<0.01	n.a	<0.01	<0.5
Soybean hulls	0.03	>3.0	0.07	3.5
Soybean Meal	<0.01	n.a	<0.01	<0.5

Product	Bravo recommended use	transfer factor	Bravo exaggerated treatment	transfer factor
Crude soybean oil	<0.01	n.a	0.03	1.5
Refined soybean oil	<0.01	n.a	<0.01	<0.5
Soybean soapstock	<0.01	n.a	<0.01	<0.5

Conclusions

There was no concentration of chlorothalonil residues in any edible processed product.

B.7.5.3.17 Processing in corn

Previous evaluation	In DAR
RMS remark	Acceptable

Characteristics

reference	:	Zeneca, Fitzgerald et al., 1993	treatment	:	field application
type of study	:	processing study	rate	:	seven applications of 1.68 kg a.i/ha
year of execution	:	1992	formulation	:	Bravo 720 (53.6% chlorothalonil)
test substance	:	chlorothalonil	GLP statement	:	yes
crop/commodity	:	corn	guidelines	:	not applicable

Study design

Field grown corn plants were treated seven times with Bravo 720 at the exaggerated rate of 1.68 kg a.i/ha/application at 7 day intervals. Corn grain and fodder samples were harvested 45 days after the last application.

The corn grain was processed into grain dust, grits, meal, flour, starch, wet and dry milled presscake and wet and dry milled crude and refined oil. Duplicate samples of each product were analysed for chlorothalonil and SDS-3701.

Results and conclusions

Chlorothalonil and SDS-3701 were only detected in corn fodder (4.41 and 0.04 mg/kg, respectively) and corn grain dust (0.06 and 0.01 mg/kg, respectively). Chlorothalonil and SDS-3701 levels were below 0.01 mg/kg in all processed products.

Remark RMS: No robust processing factors can be derived, since residues were almost always below LOD.

B.7.5.3.18 Processing in tomatoes

Previous evaluation	Submitted for the purpose of renewal, but already evaluated in an Evaluation Report submitted by The Netherlands for the Article-12 MRL-review, from which the evaluation is copied into this RAR. However, the evaluation on storage stability is re-assessed based on new storage stability studies submitted for the renewal.
RMS remark	Acceptable Furthermore, the remark in the original conclusion regarding the analytical method has now been addressed.

Report:	K-CA 6.5.3/01. Gardinal P. (2007), Chlorothalonil (R44686): Residue study on outdoor tomatoes and processed tomato products in southern France. Syngenta, Jealott's Hill International Research Centre, Bracknell, United Kingdom. Syngenta Report Number 05-6039. (Syngenta File No: R44686/4093).

Characteristics

Reference	:	Gardinal,2007 (IIA 6.5.3, IIA 6.5.4, IIIA 8.5.3, IIIA 8.5.4)	GLP	:	Yes
Type of study	:	Effect on residue levels (processing)	Guideline	:	7029/VI/95 rev. 5 (Lundehn)
Year of execution	:	2005-2006	Acceptability	:	Acceptable
Test substance	:	chlorothalonil 500 SC; batch no.SIP4C40916			

Study design

A field trial with tomato was conducted in Bias, Southern France with 3 foliar broadcast applications (7 day interval) of chlorothalonil (formulated as 500 SC) at a rate of 7500 g a.i/ha (3N), totalling 22500 g a.s./ha (3N total rate of intended use). Tomatoes were harvested at 3 DALA and processed to juice, puree and canned tomato.

Storage and storage stability

Tomatoes were processed after a storage- and transport period of 4 days (chilled storage). Tomatoes and processed fractions were stored at -20°C for a period of max ~11 months prior to analysis. The storage period is within the demonstrated storage stability period for chlorothalonil and R182281 (Vol. 1, 2.7.1) in watery matrices. It has been shown that R613636 is stable for 24 months in processed fractions of tomato (juice, paste and puree).

Analytical method

Analysis of tomatoes and processed fractions for chlorothalonil and R182281 was performed using method GRM005.01A (GC/MS (chlorothalonil) and LC-MS/MS (R182281), LOQ 0.01 mg/kg). This method is similar to method RAM 320.01, which was validated in a number of commodities, including tomato, with a LOQ of 0.01 mg/kg (Addendum 1 of DAR of January 2001). The method involved homogenisation of tomatoes in the presence of sulphuric acid and is acceptable. Concurrent procedural recoveries for chlorothalonil were included at 0.01-20 mg/kg (tomatoes) and were between

70 and 105% (i.e. acceptable). Concurrent procedural recoveries for R182281 were included at 0.01-1.0 mg/kg (tomatoes) and were between 82 and 113% (i.e. acceptable).
Analysis of tomatoes and processed fractions for R613636 was performed using method RAM 464/01 (LC-MS/MS, LOQ 0.01) which was similar to GRM005.01A, except for the cleanup step (SPE with HLB cartridges instead of C8 cartridges). Concurrent procedural recoveries for R613636 were included at 0.01-0.2 mg/kg (tomatoes) and were between 68 and 107% (i.e. acceptable).
References to validation reports for methods GRM005.01A and RAM 464/01 were provided (studies by Chaggar (2006) and Crook *et al* (2005)).

Transfer and yield factors

Transfer factors (i.e. processing factors) were calculated for chlorothalonil, for the first processing product (i.e. washed tomatoes) and the end products for the different processes. For the same products yield factors were calculated for R182281, a compound which may be formed during processing. Since the toxicological endpoints for R182281 are different from those of chlorothalonil, the yield factor Y was calculated as Y = A / B, where A is the residue of R182281in the processed product and B the residue of chlorothalonil in the initial sample of tomatoes. The yield factors calculated this way are worst case since they include the residue of R182281 already present in the tomatoes before processing. The same calculation was used to determine yield factors for R613636, where possible.

Results

For chlorothalonil, R182281 and R613636 residues, in tomatoes and processed tomato fractions, and processing factors and yield factors (first processing product and end products only), see Table B.7.7.2.3-07.
During processing to juice, the mean residue of chlorothalonil in the end product (juice, after pasteurisation) was 1.04 mg/kg. Processing to puree resulted in a mean residue of <0.01 mg/kg in the end product, puree, after sterilisation. The mean residues in the end products after canning were <0.01 mg/kg for both solid and liquid portion, after sterilisation.
The processing factors for washed tomatoes were similar for all processes and averaged 0.30. Mean processing factors for end products were <0.001 for puree and canned tomatoes and 0.11 for juice.
Yield factors for R182281 in washed tomatoes were <0.001 for all processes, R613636 was not analysed in washed tomatoes, therefore no yield factor could be calculated.
Mean yield factors for R182281 in end products varied from 0.002 (juice after pasteurisation) to 0.013 (puree after sterilisation) and were 0.003 and 0.004, respectively, for the solid and liquid portion after sterilisation of canned tomatoes. For the end products of all processes the mean yield factors for R613636 were <0.001.

Table B.7.7.2.3-07 Residues and processing/yield factors in tomato and processed tomato products after 9 treatments with chlorothalonil (formulated as 500 SC) at 3 x 7500 g a.s./ha.

Tomato product	Residues (mg/kg)			Processing factor (chlorothalonil)[1]	Yield factor (R182281)[1]	Yield factor (R613636)[1]
	chlorothalonil	**R182281**	**R613636**			
Tomatoes before processing	9.6[2]	0.02[2]	n.a.			
Juicing						
washed tomatoes[3]	2.8, 2.5, 2.9, 2.7	<0.01 (4x)	n.a.	0.28 (0.26-0.30)	<0.001 (<0.001-<0.001)	-
crushed tomatoes	2.5	<0.01	n.a.			
wet pomace sieved	3.1	0.03	n.a.			
dry pomace[3]	12[4], 13, 10, 12	0.25, 0.31, 0.28, 0.36	<0.01, <0.01, <0.01, <0.01			
raw juice, sieved	2.9	0.01	n.a.			
juice, before pasteurisation	3.1	0.01	n.a.			
juice, after pasteurisation[3]	1.1, 1.0, 0.87, 1.2	0.02, 0.02, 0.02, 0.03	<0.01, <0.01, <0.01, <0.01	0.11 (0.09-0.13)	0.002 (0.002-0.003)	<0.001 (<0.001-<0.001)
puree production						
washed tomatoes[3]	3.4, 4.0, 2.3, 2.5	<0.01, <0.01, <0.01, <0.01	n.a.	0.32 (0.24-0.42)	<0.001 (<0.001-<0.001)	-
crushed tomatoes	3.5	<0.01	n.a.			
reduced tomatoes	0.05	0.24	<0.01			
sieved tomatoes	<0.01	0.15	<0.01			
wet pomace, sieved	0.09	0.38	<0.01			
puree, before sterilisation	<0.01	0.14	<0.01			
puree, after sterilisation[3]	<0.01, <0.01, <0.01, <0.01	0.15, 0.13, 0.12, 0.11	<0.01, <0.01, <0.01, <0.01	<0.001 (<0.001-<0.001)	0.013 (0.011-0.016)	<0.001 (<0.001-<0.001)
Canning						

Tomato product	Residues (mg/kg)			Processing factor (chlorothalonil)[1]	Yield factor (R182281)[1]	Yield factor (R613636)[1]
	chlorothalonil	R182281	R613636			
washed tomatoes[3]	3.9, 2.8, 1.6, 2.7	<0.01, <0.01, <0.01, <0.01	n.a.	0.29 (0.17-0.41)	<0.001 (<0.001-<0.001)	-
peeled tomatoes	0.24	<0.01	<0.01			
peels	28	0.03	<0.01			
solid portion, before sterilisation	0.43	<0.01	<0.01			
liquid portion before sterilisation	2.0	<0.01	<0.01			
solid portion, after sterilisation[3]	<0.01, <0.01, <0.01, <0.01	0.02, 0.04, 0.04, 0.05	<0.01, <0.01, <0.01, <0.01	<0.001 (<0.001-<0.001)	0.004 (0.002-0.005)	<0.001 (<0.001-<0.001)
liquid portion, after sterilisation[3]	<0.01, <0.01, <0.01, <0.01	0.04, 0.02, 0.02, 0.02	<0.01, <0.01, <0.01, <0.01	<0.001 (<0.001-<0.001)	0.003 (0.002-0.004)	<0.001 (<0.001-<0.001)

[1] mean processing or yield factor, with range in parentheses.

[2] mean of 2 field samples.

[3] results of balance study and 3 follow-up studies.

[4] mean of 2 analyses.

Conclusions

Following 3 foliar spray applications of chlorothalonil (formulated as 500 SC) at 7500 g a.s./ha (3N) on tomato, residues of chlorothalonil were 9.6 mg/kg in whole fruit. Mean residues for washed tomatoes were 2.84 mg/kg. Mean residues in juice were 1.04 mg/kg and in puree and canned tomatoes (solid and liquid fraction) <0.01 mg a.s./kg. Associated mean processing factors were 0.30 (washing), 0.11 (juice) and <0.001 for canned tomatoes and puree.

In addition, suitability of methods GRM005.01A and RAM 464/01 for the analysis of chlorothalonil, R182281 and R613636 in tomatoes should be addressed (submission of (interim) reports of studies by Chaggar (2006) and Crook *et al* (2005)).

Guidelines & Limitations

1. Samples of tomato and processed fractions were stored frozen for a period of ~11 months prior to analysis. Stability for up to 4 years at -7°C or below was confirmed in various crops including tomato. No storage stability in processed commodities was reported, however storage stability for chlorothalonil as stated above can be extrapolated to processed tomato fractions.
2. All other comments were incorporated in the above summary.

B.7.5.3.19 Processing in barley

Previous evaluation	Submitted for the purpose of renewal, but already evaluated in an Evaluation Report submitted by The Netherlands for the Article-12 MRL-review, from which the evaluation is copied into this RAR. However, the evaluation on storage stability is re-assessed based on new storage stability studies submitted for the renewal.
RMS remark	~~Acceptable~~ For chlorothalonil and SDS-3701 acceptability of the study is pending submission of storage stability data in cereal grain.

Report:	K-CA 6.5.3/02. Simon P. (2007), Chlorothalonil: Residue study on barley and processed barley products in Germany. Syngenta Agro GmbH, Technologiepark 1-5, D-63477 Maintal, Germany. Syngenta Report Number gba243004. (Syngenta File No: R44686/4112).

Characteristics

Reference	:	Simon, 2007 (IIA 6.5.3, IIA 6.5.4, IIIA 8.5.3, IIIA 8.5.4)	GLP	:	Yes
Type of study	:	Effect on residue levels (processing)	Guideline	:	7035/VI/95 (Lundehn)
Year of execution	:	2004-2007			
Test substance	:	chlorothalonil 500 SC (A7867A); batch no. YAL-2-A23-A, 505 g a.s./L and batch no. YAL-3-C24-A, 512 g a.s./kg	Acceptability	:	Acceptable

Study design

A field trial with spring barley (variety Barke) was conducted in Mochau-Lüttewitz, Germany, with 2 foliar applications (29 day interval) of chlorothalonil (formulated as 500 SC) at rates of 3 kg a.s./ha (3N). Barley was harvested from the field trial at 35 DALA, transported at the same day to two processing facilities (Köthen and Berlin) and stored at ambient temperature for about 2 months until processing to pot barley and beer.

Methods of analysis

Barley and processed fractions were analysed for chlorothalonil and R182281 by method RAM 365/02 (GC/MS, LOQ 0.01 mg/kg) and for R613636 by method 464/01 (LC-MS/MS, LOQ 0.01 mg/kg). Concurrent procedural recoveries were included for all three analytes at ≥0.01 mg/kg for grain, offal, malt, malt sprouts, spent grain, abrasion dust, pot barley, wort, flocs and beer. Recoveries were acceptable: 61-104% (chlorothalonil, mean 87%, RSD 11%), 85-124% (R182281, mean 101%, RSD 12%) and 83-115% (R613636, mean 98%, RSD 13%). All residue data in the Table below were uncorrected for recovery of the analytical method.

Stability during homogenisation

During the preparation of the monograph, there were concerns about potential losses of chlorothalonil during sample preparation and homogenisation (reporting table from ECCO 111 residue peer review meeting [28.6.01]). Syngenta prepared a statement to address these concerns (Lister N and Smith M, chlorothalonil comments relating to metabolism and residues, ERA3753, 21 August 2001). It was

demonstrated that losses of chlorothalonil were prevented by performing sample homogenisation in the presence of 10% v/w 0.1M sulphuric acid. This procedure was not followed during homogenisation of the samples of all studies. In the evaluation table of 29.09.2004, under point 5.3, it was concluded that *"instability of chlorothalonil during homogenisation is not an issue for the matrices of the only remaining use claimed by Syngenta (wheat grain and straw)"*. For the comparable dry matrices barley grain, sieved or cleaned grain, offal, abrasion dust and pot barley, instability should therefore not be an issue. The last step in the preparation of malt is kiln drying, which stops all enzymatic processes, including that leading to endogenous binding of chlorothalonil during sample preparation. Hence also the results of the processed samples are valid in this respect.

Storage and storage stability

Barley and processed fractions were stored at ≤-18°C for a period of up to 22 months prior to analysis for chlorothalonil and R182281, and up to 12 months prior to analysis for R613636.

Storage stability of chlorothalonil and metabolite SDS-3701 (R182281) has been discussed during the expert Peer Review Meeting (#164). It was concluded that based on the available data storage stability of chlorothalonil in cereal grain has been demonstrated for 62 days. For metabolite SDS-3701 no storage stability data in cereal grain in available and the meeting concluded that extrapolation from other high starch matrix commodities is not acceptable. A data gap is set. It is therefore concluded that the residue trials in barley with chlorothalonil and SDS-3701 are not acceptable since the storage time of the samples exceeds the demonstrated storage stability.

~~It is sufficiently demonstrated that chlorothalonil and R182281 are stable in high starch commodities for at least 22 months (Vol. 1, 2.7.1).~~ R613636 is shown to be stable in several processed grain commodities for at least 24 months (Vol. 1, 2.7.1).

Residues in control samples

Residues in control samples produced from grain from untreated plots were below the LOQ of 0.01 mg/kg for all three analytes except in a few cases for intermediate products (grain after cleaning, 0.01 mg/kg chlorothalonil; two samples of offal, 0.02 & 0.04 mg/kg chlorothalonil and 0.02 mg/kg R182281) and in one case for an end product (pot barley from Köthen, 0.01 mg/kg R182281).

Transfer and yield factors

Transfer factors were calculated for chlorothalonil for the end products beer and pot barley. For the same products yield factors were calculated for R182281, a compound which may be formed during processing. Since the toxicological endpoints for R182281 are different from those of chlorothalonil, the yield factor Y was calculated as Y = A / B, where A is the residue of R182281 in the processed product and ~~P~~ B the residue of chlorothalonil in the initial sample of grain. The yield factors calculated this way are worst case since they include the residue of R182281 already present in the initial sample of "grain as received" or "grain before hulling".

In all calculations the initial residue of chlorothalonil was taken to be the value determined in the grain as received or (where determined in pot barley production) grain before hulling. Where residues in end products were <0.01 mg/kg, 0.01 mg/kg was used in the calculation.

Results

The residues in barley and processed barley fractions and the processing and yield factors are shown in Table B.7.7.2.9-1. The potential processing product R613361 was not found above the LOQ in any process product and yield factors were not calculated. The transfer factors for chlorothalonil were <0.04, <0.04, <0.05 and <0.05 in beer (mean <0.05) and 0.22, 0.22, 0.18 and 0.25 in pot barley (mean 0.22). The yield factors for R182281 were <0.04, <0.04, <0.05 and <0.05 in beer (mean <0.05) and 0.11, <0.05, 0.07 and 0.07 in pot barley (mean 0.08).

Table B.7.7.2.9-01 Residues and processing factors in barley and processed barley products

processing step	crop part	residue (mg/kg) a.s.	residue (mg/kg) R182281	residue (mg/kg) R613636	~~Transfer~~ Processing factor a.s.	Yield factor R182281	Processing factor R182281
field sample	grain	0.90	0.06	na[(B)]			
cleaning(A)	grain as received[(A)]	0.19	0.18	na			
	grain after cleaning	0.46	0.21	na			
	offal	8.7	4.3	na			
beer (balance study, Kothen)	grain after sieving	0.31	0.14	na			
	grain before malting	0.26	0.22	na			
	malt after drying	<0.01	0.1	<0.01			
	malt sprouts	0.02	0.08	<0.01			
	malt before brewing	<0.01	0.07	<0.01			
	spent grain	<0.01	0.09	<0.01			
	wort before cooking	<0.01	0.02	<0.01			
	flocs (spent hops)	<0.01	0.24	<0.01			
	wort after cooking	<0.01	0.02	<0.01			
	young beer	<0.01	0.01	<0.01			
	spent yeast	<0.01	0.31	<0.01			
	beer	<0.01	<0.01	<0.01	<0.05	<0.05	<0.06
pot barley (balance study, Kothen)	grain before hulling	0.27	0.28	na			
	abrasion dust	0.54	1.2	<0.01			
	pot barley	0.06	0.02	<0.01	0.32 ~~0.22~~	0.11	0.11
beer follow-up 1, Kothen)	malt after drying	<0.01	0.12	na			
	malt before brewing	<0.01	0.09	<0.01			
	beer	<0.01	<0.01	<0.01	<0.05	<0.05	<0.06
pot barley follow-up 1, Kothen)	abrasion dust	0.59	1.3	<0.01			
	pot barley	0.06	<0.01	<0.01	0.32 ~~0.22~~	<0.05	<0.06
cleaning(A)	grain as received[(A)]	0.28	0.21	na			
	grain after cleaning	0.45	0.22	na			
	offal	8.2	4.8	na			

processing step	crop part	residue (mg/kg)			~~Transfer~~ Processing factor a.s.	Yield factor R182281	Processing factor R182281
		a.s.	R182281	R613636			
beer	malt after drying	<0.01	0.09	<0.01			
follow-up 2,	malt before brewing	<0.01	0.08	<0.01			
Berlin)	beer	<0.01	<0.01	<0.01	<0.04	<0.04	<0.05
pot barley	abrasion dust	0.62	0.97	<0.01			
follow-up 2,	pot barley	0.05	0.02	<0.01	0.18	0.07	0.095
Berlin)							
beer	malt after drying	<0.01	0.10	<0.01			
follow-up 3,	malt before brewing	<0.01	0.08	<0.01			
Berlin)	beer	<0.01	<0.01	<0.01	<0.04	<0.04	<0.05
pot barley	abrasion dust	0.75	1.3	<0.01			
follow-up 3,	pot barley	0.07	0.02	<0.01	0.25	0.07	0.095
Berlin)							

(A) The grain was stored at ambient temperature for 2 months prior to the start of processing.

(B) na = not analysed.

Conclusions

The transfer factors for chlorothalonil were <0.04, <0.04, <0.05 and <0.05 in beer (mean <0.05) and 0.22, 0.22, 0.18 and 0.25 in pot barley (mean 0.22). The yield factors for R182281 were <0.04, <0.04, <0.05 and <0.05 in beer (mean <0.05) and 0.11, <0.05, 0.07 and 0.07 in pot barley (mean 0.08). R613636 was not found in any processed product (<0.01 mg/kg).

However, the acceptability of the results is pending the demonstration of storage stability of chlorothalonil and SDS-3701 (R182281) in cereal grain.

Guidelines & Limitations

Comments were incorporated in the above summary.

B.7.5.3.20 Processing in barley

Previous evaluation	Submitted for the purpose of renewal
RMS remark	~~Acceptable~~ The acceptability of the results is pending the demonstration of storage stability of chlorothalonil and SDS-3701 (R182281) in cereal grain.

Report:	K-CA 6.5.3/03. North N. (2014), Chlorothalonil – Residue study on barley and processed products in Germany and Southern France in 2011. Eurofins Agroscience Services Ltd., Derbyshire, United Kingdom. Syngenta Report Number S11-00524-REG, File No: A7867A_11251)

Guidelines

FAO Guidelines on Producing Pesticide Residues Data from Supervised Trials (Rome, 1990).

Commission of the European Communities: General Recommendations for the Design, Preparation and Realization of Residue Trials (SANCO 7029/V1/95 rev. 5 22/7/1997).
Commission of the European Communities, Processing Studies; (SANCO 7035/V1/95 rev. 5 22/7/1997)
OECD Guidelines for the Testing of Chemicals: OECD Test Guideline 508: Magnitude of the Pesticide Residues in Processed Commodities.
European Commission Guidance Document on Residue Analytical Method (SANCO/825/00 revision 8.1, 16 Nov 2010).

GLP

The study was carried out according to the principles of Good Laboratory Practice.

Executive Summary

Two residue trials on barley were conducted in Germany and southern France during 2011. Two applications of chlorothalonil were applied to each plot as A7867A, a suspension concentrate (SC) formulation at a nominal rate of 3 kg a.s./ha (= 4 times intended rate) separated by a 10 or 17 day interval. Samples of mature barley grain were harvested 42 days after the last application of A7867A and analysed to determine residues of chlorothalonil and R182281.
Samples of mature barley grain were processed into pearl barley, brewing malt, beer, pot barley and barley flour. One balance study and three follow-up studies were carried out for each process. The processed samples were analysed for chlorothalonil and R182281.

Separate mass balances and transfer factors for chlorothalonil and for R182281 were calculated. Sufficient data is available to allow transfer factors to be calculated for chlorothalonil and R182281 residues from barley into beer, pot barley and other barley processed commodities. It is concluded that residues of chlorothalonil and R182281 would not be expected to concentrate in beer, pearl barley, pot barley or flour.

A1. Test Materials

Test Material	A7867A
Description	Suspension concentrate formulation containing chlorothalonil
Purity	495 g/L
Batch number	SAV0L00018
Stability of test compound	The test substance is assumed to be stable for the period of use in the study

A2. Test Facilities

Field trials	Niedersachen, Germany	Midi Pyrénnées, France
Processing phase	Eurofins Agroscience Services GmbH, Carl-Goerdeler-Weg 5, D-21684 Stade, Germany	
Analytical phase	Eurofins Agroscience Services Ltd., Slade Lane, Wilson, Melbourne, Derbyshire DE73 8AG, UK	

B. STUDY DESIGN AND METHODS

B1. Processing phase

In field trials, commercially grown barley was treated twice (at growth stage BBCH 55 and at BBCH 65 or 69) with a foliar spray of A7867A, at nominal application rates of 3.0 kg chlorothalonil/ha. The interval between the applications was 10 or 17 days.

Mature barley grain was harvested 42 days after the last application and used for the production of pearl barley, beer and pot barley.

Prior to each of the balance and follow-up processing studies, uncleaned barley grain from each trial was analysed to give a pre-processed residue value.

The uncleaned barley grain was cleaned using a 'sample cleaner' and samples of cleaned barley grain were taken (samples of shrivelled grain and impurities were also taken for the balance study).

Samples were stored for 5-8 months before processing.

Storage stability of chlorothalonil and metabolite SDS-3701 (R182281) has been discussed during the expert Peer Review Meeting (#164). It was concluded that based on the available data storage stability of chlorothalonil in cereal grain has been demonstrated for 62 days. For metabolite SDS-3701 no storage stability data in cereal grain in available and the meeting concluded that extrapolation from other high starch matrix commodities is not acceptable. A data gap is set. It is therefore concluded that the residue trials in barley with chlorothalonil and SDS-3701 are not acceptable since the storage time of the samples exceeds the demonstrated storage stability.

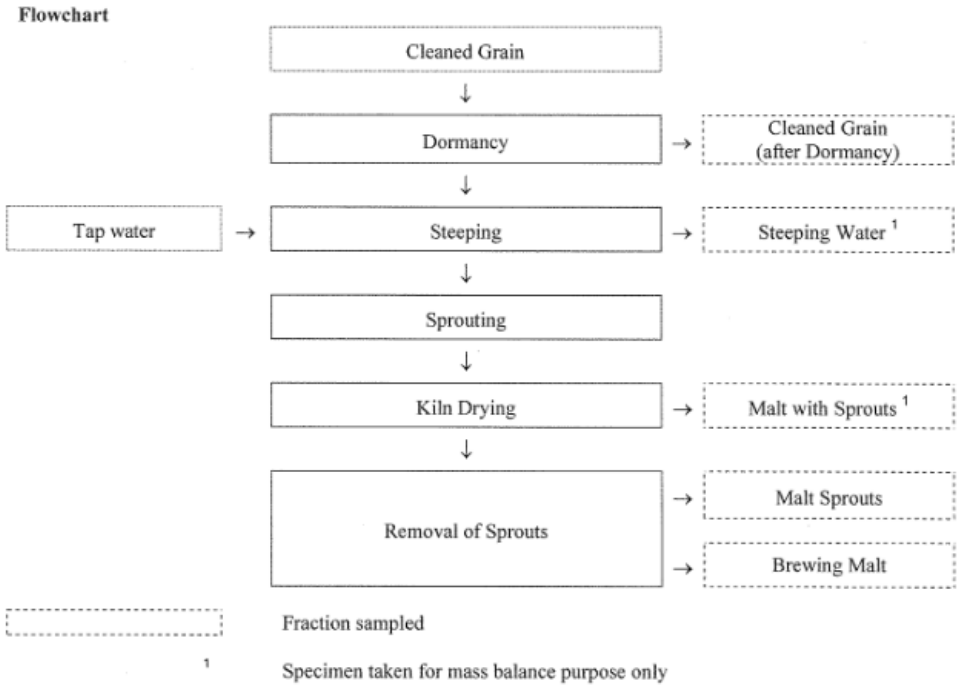
Pot barley, pearl barley and flour

The moisture content of the grain was tested and found to be 15% therefore a conditioning step was not required. The cleaned barley grain was decorticated at different abrasion rates (8.2-12% for pot barley and 25-31% for pearl barley) and fractions of pot barley and bran, and pearl barley and 'rub off' were taken. Some of the pot barley fraction was milled to give flour.

Malting

Cleaned grain was stored cooled for 63-85 days. After dormancy the grain was steeped by covering with water for 23-24 hours, after which the water was removed. For the sprouting process which was conducted in a climatic exposure cabinet, the grains were turned by hand periodically and moistened with tap water. The temperature during sprouting was 10-14°C. The emergence period was 6-8 days. After sprouting, the green malt was kiln dried in a drying oven at 50-60°C for 6 hours. The drying temperature was then elevated for approximately 8 hours to 85-100°C, followed by a generally constant temperature of 85-100°C for another 9-11 hours. After kiln drying the malt sprouts were separated mechanically. Fractions of malt sprouts and brewing malt were taken.

Figure 7.5.3.20-1 Flowchart for malting



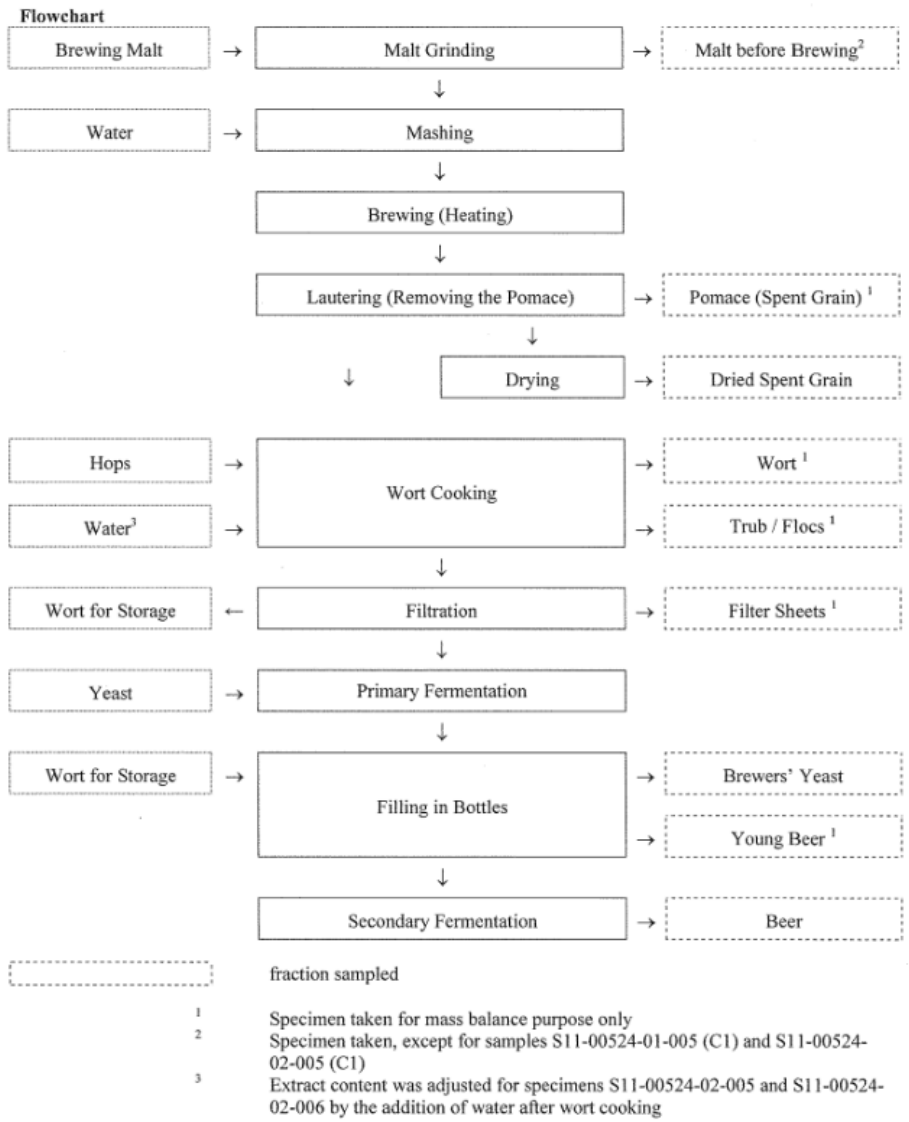
Beer

The brewing malt was stored for 1-6 days at 4.5-7.0°C until brewing. The malt was ground and mixed with warm water to produce a mash. The mash temperature was slowly raised to 78°C with resting periods and then rinsed with water at 80-82°C. The wort was then cooked for 80 minutes during which hops were added. The deposited 'flocs' ('trub' or 'hops draff') were removed. The remaining wort was filtered and cooled. Fermentation was started by addition of yeast solubilised in wort and lasted for 7 days, after which the beer was stored in bottles for 14 days for a secondary fermentation.

The following samples were taken for analysis in order to determine the residue accountability from the mass balance:

Uncleaned grain, cleaning impurities, cleaned grain, cleaned grain after storage, 'rub-off', pearl barley, cleaned grain after dormancy, steeping water, malt with sprouts, malt sprouts, brewing malt, malt before brewing, spent grain, dried spent grain, wort, trub/flocs, brewer's yeast, young beer, beer, bran, pot barley and barley flour.

Figure 7.5.3.20-2 Flowchart for brewing



Transfer factors were determined from the mass balance and follow up studies for the following samples:

Cleaned grain, pearl barley, pot barley, barley flour, brewing malt, dried spent grain, brewer's yeast and beer.

The chlorothalonil and R182281 residue accountability for the brewing process was calculated from a mass balance study, and transfer factors for the processed commodities were determined from this and the additional follow up studies.

Sampled grain and processed samples were stored frozen for a maximum period of 14 months from sampling to analysis, which is covered by storage stability studies (Vol. 1, 2.7.2).

B2. Analytical Phase

Samples of the raw agricultural commodity (grain) and various processed fractions were analysed for chlorothalonil and R182281 using method GRM005.01A. The method involved extraction with acetone/5M sulphuric acid (95:5 v/v), dilution with water followed by SPE clean up for chlorothalonil or taking up in acetonitrile:water (50/50 v/v) for R182281 with minor modifications. For trob and flocs an additional extract purification step using florisil was employed for chlorothalonil. Subsequently, analysis was performed by gas chromatography with mass selective detection (GC-MSD) or chlorothalonil and LC-MS/MS for R182281. The LOQ was 0.01 mg/kg for both analytes in all commodities. A full method description and validation data are presented in B.5.2.1 (K-CA 4.2/01-K-CA 4.2/07). For the processing fractions the analytical method was successfully validated in the current report.

II. RESULTS AND DISCUSSION

Procedural recoveries for the method of analysis are presented in Table 7.5.3.20-1 for the processing matrices.

Table 7.5.3.20-1: Summary of recovery data for chlorothalonil and R182281 in processed matrices of barley grain

Commodity	Fortification concentration (mg/kg)	Chlorothalonil			Quantified against matrix matched standards		
		No samples	Mean recovery (%)	RSD (%)	No samples	Mean recovery (%)	RSD (%)
Pearly barley	0.01	5	99	1.7	5	107	4
	0.1	5	101	0.8	5	108	7
Brewing malt	0.01	5	103	0.8	5	108	4
	0.1	5	102	2.2	5	98	2
Wort	0.01	5	97	2.3	5	103	3
	0.1	5	105	2.0	5	98	3
Brewer's yeast	0.01	5	96	1.6	5	103	2
	0.1	5	98	3.4	5	101	7
Young beer	0.01	5	98	3.9	5	109	5
	0.1	5	100	3.8	5	95	5

Residues of chlorothalonil in barley grain specimens taken at normal commercial harvest were 1.05 and 1.53 mg/kg, while residues of R182281 were 0.03 and 0.04 mg/kg. A summary of the measured residues from the various processed fractions is given in Table 7.5.3.20-2. The mean transfer factors for each commodity for chlorothalonil and R182281 were calculated and are presented in Table 7.5.3.20-3 and Table 7.5.3.20-4, respectively.

Table 7.5.3.20-2: Summary of chlorothalonil and R182281 residues in barley processed commodities from trials in Germany and southern France

Commodity	Residues (mg/kg)							
	Balance 1[a]		Follow-up 1[a]		Follow-up 2[b]		Follow-up 3[b]	
	chloro-thalonil	R182281	chloro-thalonil	R182281	chloro-thalonil	R182281	chloro-thalonil	R182281
Cleaning								
Mean barley grain (RAC)	0.97	0.12	0.76	0.11	0.82	0.11	0.82	0.08
Cleaned grain	0.61	0.09	0.41	0.06	0.40	0.05	0.60	0.07
Impurities	3.99	0.83	-	-	-	-	-	-
Pearl barley								
Cleaned grain after storage	0.65	0.11	0.40	0.11	0.49	0.08	0.58	0.08
Rub- off	1.05	0.72	-	-	-	-	-	-
Pearl barley	0.11	0.02	0.13	0.02	0.04	<0.01	0.10	0.01
Malt								
Cleaned grain after dormancy	0.58	0.10	0.27	0.07	0.70	0.08	0.63	0.08
Steeping water	0.02	0.03	-	-	-	-	-	-
Malt with sprouts	0.02	0.07	-	-	-	-	-	-
Malt sprouts	0.06	0.12	0.06	0.12	0.06	0.16	0.09	0.23
Brewing malt	0.02	0.06	0.02	0.04	0.02	0.05	0.02	0.06
Beer								
Malt before brewing	0.01	0.05	<0.01	0.04	<0.01	0.05	0.01	0.06
Spent grain	<0.01	0.08	-	-	-	-	-	-
Dried spent grain	<0.01	0.04	<0.01	0.02	<0.01	0.04	<0.01	0.05
Wort	<0.01	0.02	-	-	-	-	-	-
Trub/flocs	<0.01	0.03	-	-	-	-	-	-
Brewer's yeast	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
Young beer	<0.01	0.02	-	-	-	-	-	-
Beer	<0.01	0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
Pot barley/flour								
Bran	2.21	1.10	-	-	-	-	-	-
Pot barley	0.23	0.05	0.16	0.05	0.11	0.03	0.10	0.02
Barley flour	0.07	0.07	0.06	0.07	0.05	0.03	0.06	0.04

--: not analysed in follow up study.
a: trial in Germany;
b: trial in France

Table 7.5.3.20-3: Summary of chlorothalonil transfer factors into processed barley products

Commodity	Transfer Factor	Median Transfer Factor
Cleaned grain	0.63, 0.54, 0.49, 0.73	0.59
Pearl barley	0.11, 0.17, 0.05, 0.12	0.12
Pot barley	0.24, 0.21, 0.13, 0.12	0.17
Barley flour	0.07, 0.08, 0.06, 0.07	0.07
Brewing malt	0.02, 0.03, 0.02, 0.02	0.02
Dried spent grain	<0.01, <0.01, <0.01, <0.01	<0.01
Brewer's yeast	<0.01, <0.01, <0.01, <0.01	<0.01
Beer	<0.01, <0.01, <0.01, <0.01	<0.01

Transfer factor = residue in processed commodity/mean residue in uncleaned grain (e.g. for cleaned grain 0.61/0.97 = 0.63)

Table 7.5.3.20-4: Summary of R182281 transfer factors into processed barley products

Commodity	Transfer Factor	Median Transfer Factor
Cleaned grain	0.75, 0.55, 0.45, 0.88	0.65
Pearl barley	0.17, 0.18, <0.09, 0.13	0.15
Pot barley	0.42, 0.45, 0.27, 0.25	0.35
Barley flour	0.58, 0.64, 0.27, 0.50	0.54
Brewing malt	0.50, 0.36, 0.45, 0.75	0.48
Dried spent grain	0.33, 0.18, 0.36, 0.63	0.35
Brewer's yeast	<0.08, <0.09, <0.09, <0.13	<0.09
Beer	0.08, <0.09, <0.09, <0.13	0.09

Transfer factor = residue in processed commodity/mean residue in uncleaned grain (e.g. for cleaned grain 0.09/0.12 = 0.75)

Residues of chlorothalonil and R182281 in the pre-processed barley grain samples of individual samples were 0.70 to 1.05 mg/kg and 0.07 to 0.12 mg/kg, respectively. No residues of chlorothalonil or R182281 were found at or above the LOQ (0.01 mg/kg) in any of the untreated barley grain or untreated processed commodities.

Processing into pearl barley

Residues of both chlorothalonil and R182281 were reduced during pearl barley production; residues of chlorothalonil in pearl barley were 0.04 to 0.13 mg/kg (mean transfer factor of 0.11) and residues of R182281 were <0.01 to 0.02 mg/kg (mean transfer factor of < 0.14). A mass balance of 100% was achieved; 72% of the initial chlorothalonil residue and 94% of the initial R182281 was recovered during the process.

Processing into pot barley and flour

For pot barley production residues of chlorothalonil and R182281 were also reduced; residues of chlorothalonil in pot barley were 0.10 to 0.23 mg/kg (mean transfer factor of 0.18) and residues of R182281 were 0.02 to 0.05 mg/kg (mean transfer factor of 0.35). For barley flour chlorothalonil

residues were 0.05 to 0.07 mg/kg (mean transfer factor of 0.07) and residues of R182281 were 0.03 to 0.07 mg/kg (mean transfer factor of 0.50). The mass balance during pot barley and barley flour production was 100% of the initial barley grain mass; 73% of the initial chlorothalonil residue and 95% of the initial R182281 was recovered during the process.

Processing into beer

No residues of chlorothalonil were found above 0.01 mg/kg in the treated beer samples. Residues of R182281 in beer were <0.01 to 0.01 mg/kg. The mass balance during beer production was 102% of the initial barley grain mass; 66% of the initial chlorothalonil residue and 93% of the initial R182281 residue was recovered during the process.

III. CONCLUSIONS

Sufficient data is available to allow transfer factors to be calculated for chlorothalonil and R182281 residues from barley into beer, pot barley and other barley processed commodities. It is concluded that residues of chlorothalonil and R182281 would not be expected to concentrate in beer, pearl barley, pot barley or flour.

However, the acceptability of the results is pending the demonstration of storage stability of chlorothalonil and SDS-3701 (R182281) in cereal grain.

B.7.5.3.21 Processing in barley

Previous evaluation	Submitted for the purpose of renewal
RMS remark	Not acceptable, ~~except~~ for metabolite R613636. The acceptability of the results is pending the demonstration of storage stability of chlorothalonil and SDS-3701 (R182281) in cereal grain.

Report:	K-CA 6.5.3/04. Sala A. (2014h), Determination of chlorothalonil and its metabolites SDS3701 and R613636 residues in raw agricultural commodity barley and processed commodity (pot barley, brewing malt, beer) following two applications of chlorothalonil 500 SC. Research Centre BioSpheres, Salerano sul Lambro, Italy. Syngenta File No: R044686_11189. Report Number RAU-008-14.

Guidelines

Regulation (EC) No. 1107/2009 of the European Parliament and of the Council of 21 October 2009, concerning the placing of plant protection products on the market and repealing Council Directives 78/117/EEC and 91/414/EEC.

EU Guidance documents on residue analytical methods SANCO/3029/99, rev. 4 (11/07/2000)

EC guidance document 1607/VI/97 rev.2, 10/6/1999

GLP

The study was carried out according to the principles of Good Laboratory Practice.

Executive Summary

Outdoor barley grown in field trials in Italy and Poland was treated with Chlorothalonil 500 SC. The plots were sprayed twice with nominal application rates of 3.0 kg chlorothalonil/ha (= 4 times intended rate). Samples of mature barley grain were harvested 40 or 54 days after the last application and analysed to determine residues of chlorothalonil, R182281 and R613636.

Additional samples of the mature barley grain taken at 40 or 54 days from the treated plots were used for the production of pot barley, brewing malt and beer, and the processed samples were analysed for residues of chlorothalonil, R182281 and R613636.

Transfer factors for the various processed fractions were calculated. The study showed that residues of chlorothalonil and R182281 would not be expected to concentrate in beer, brewing malt or pot barley. Residues of R613636 were below the LOQ (0.01 mg/kg) in the grain before processing and all processed products.

A1. Test Materials

Test Material	Chlorothalonil 500 SC
Description	Suspension concentrate formulation containing chlorothalonil
Purity	507 g/L
Batch number	PN1911
Stability of test compound	The test substance has been shown to be stable under the storage and test conditions of the study

A2. Test Facilities

Field trial	Italy	Poland
Malting and brewing	Staphyt Processing, MAS la Paluzette, F-34590, France	
Pot barley production	INRA, 2 Place Viala, 34060 Montpellier, France	
Analytical phase	Biospheres Residues Analysis Unit, Via Vittoria Veneto, 26857 Salerano sul Lambro (LO), Italy	

B. STUDY DESIGN AND METHODS

B1. Processing phase

In two field trials, barley was treated twice with a foliar spray of the formulation at nominal application rates of 3.0 kg chlorothalonil/ha. The interval between the applications was 9-14 days (trial 1: BBCH 39 and 61; trial 2: BBCH 59 and 69).

Mature barley grain was harvested at the two sites 40 or 54 days after the last application and used for the production of beer, brewing malt and pot barley. Samples were stored for about 2 months before processing.

The grains were prepared by sieving to retain grains with a minimum diameter of 2.5 mm. The sieved grain was steeped at a temperature of 18°C for 47 hours then germinated at 16^{o}C for approximately 5 days. The germinated grain was then dried by raising the temperature from 30° to 80°C over a period of 24 hours. Brewing malt specimens were taken.

The malt was milled before mashing. Ground malt was mixed with water at 45°C and the pH was adjusted to 5.5 by the addition of lactic acid. The mashing process took place in three stages: firstly at 45°C for 20 minutes, secondly at 64°C for 20 minutes and thirdly at 74°C for 30 minutes. Hops were added and the wort was cooked at 100°C for approximately 1.5 hours. The wort was cooled, yeast was added and left to ferment at 12°C for 15-16 days until a stable density was obtained. Beer specimens were taken.
For pot barley, the grains were passed through a husker twice and the hulls were recovered. Pot barley specimens were taken.
Samples were stored frozen for maximally 6 months before analysis, which is acceptable for metabolite R613636. ~~, which is coverd by storage stability studies (Vol. 1, 2.7.2).~~
Storage stability of chlorothalonil and metabolite SDS-3701 (R182281) has been discussed during the expert Peer Review Meeting (#164). It was concluded that based on the available data storage stability of chlorothalonil in cereal grain has been demonstrated for 62 days. For metabolite SDS-3701 no storage stability data in cereal grain in available and the meeting concluded that extrapolation from other high starch matrix commodities is not acceptable. A data gap is set. It is therefore concluded that the residue trials in barley with chlorothalonil and SDS-3701 are not acceptable since the storage time of the samples exceeds the demonstrated storage stability.

B2. Analytical Phase

Samples of the raw agricultural commodity (grain) and various processed fractions were analysed for chlorothalonil, R182281 and R613636. The analytical method used (validated in study BIU-016-14) involved extraction with acidified ethyl acetate followed by analysis using a gas chromatograph equipped with µ-ECD detector for chlorothalonil. For R182281 the analytical method consisted of extraction with methanol and analysis with LC-MS/MS. R613636 was analysed after extraction with acidified acetone, dilution with water and solid phase extraction by means of LC-MS/MS. The LOQ was 0.01 mg/kg for chlorothalonil and R613636, and 0.02 mg/kg for R182281 in all commodities. Full method descriptions and validation data are presented in B.5.1.2.5 (K-CA 4.1.2/32). For the processing fractions the analytical method for chlorothalonil and R182281 was successfully validated in the current report. No procedural recoveries are available for R613636.
Transfer factors for the processed commodities were determined.

II. RESULTS AND DISCUSSION

Procedural recoveries for the method of analysis are presented in Table 7.5.3.21-1 for the processing matrices.

Table 7.5.3.21-1: Summary of recovery data for chlorothalonil and R182281 in processed matrices of barley grain

Commodity	Fortification concentration (mg/kg)	Chlorothalonil		R182281	
		Mean recovery (%) n=3	RSD (%) n=3	Mean recovery (%) n=3	RSD (%) n=3
Pot barley	0.01	95	1.0	99	7
	0.1	99	0.6	99	1
Brewing malt	0.01	85	2.7	86	21[1]
	0.1	100	4.1	108	9
Beer	0.01	91	4.5	90	3
	0.1	96	1.2	98	2

[1] The RSD is above the limit of 20%, but is not considered to impair the reliability of the results.

A summary of the measured residues from the various fractions for the processed fraction is given in Table 7.5.3.21-2.

Table 7.5.3.21-2: Summary of chlorothalonil, R182281 and R613636 residues in barley and processed commodities from trials conducted in Italy and Poland

Trial:	Residues (mg/kg)					
	Italy			Poland		
Commodity	Chlorothalonil	R182281	R613636	Chlorothalonil	R182281	R613636
Grain (RAC)	0.39	0.04	<0.01	0.21	0.04	<0.01
Grain for processing	0.36	0.05	<0.01	0.11	0.04	<0.01
Pot barley	<0.01	<0.02	<0.01	0.04	<0.02	<0.01
Brewing malt	0.02	<0.02	<0.01	<0.01	<0.02	<0.01
Beer	<0.01	<0.02	<0.01	<0.01	<0.02	<0.01

The mean transfer factors for each commodity for chlorothalonil and R182281 were calculated and are presented in Table 7.5.3.21-3. Transfer factors for R613636 were not calculated as residues in the grain before processing and in all processed commodities were below the LOQ (0.01 mg/kg).

Table 7.5.3.21-3: Summary of transfer factors into processed barley products

	Chlorothalonil		R182281	
Commodity	Transfer Factors	Mean Transfer Factor	Transfer Factors	Median Transfer Factor
Pot barley	<0.03, 0.36	<0.20	<0.4, <0.5	<0.5
Brewing malt	0.06, <0.09	<0.07	<0.4, <0.5	<0.5
Beer	<0.03, <0.09	<0.06	<0.4, <0.5	<0.5

Transfer factor = residue in processed commodity/mean residue in grain for processing (e.g. for brewing malt 0.02/0.36 = 0.06).

Residues of chlorothalonil and R182281 did not concentrate in pot barley, brewing malt or beer.

III. CONCLUSIONS

Sufficient data is available to allow transfer factors to be calculated for chlorothalonil residues from barley into beer, pot barley and brewing malt. It can be concluded that residues of chlorothalonil and R182281 would not be expected to concentrate in beer, brewing malt or pot barley.
However, the acceptability of the results is pending the demonstration of storage stability of chlorothalonil and SDS-3701 (R182281) in cereal grain.

Residues of R613636 were below the LOQ (0.01 mg/kg) in the grain before processing and all processed products.

Remark RMS: Since no procedural recoveries are available for R613636, results for this metabolite are considered as not acceptable.

B.7.5.3.22 Processing in wheat

Previous evaluation	Submitted for the purpose of renewal, but already evaluated in an Evaluation Report submitted by The Netherlands for the Article-12 MRL-review, from which the evaluation is copied into this RAR
RMS remark	~~Acceptable.~~ The acceptability of the results is pending the demonstration of storage stability of chlorothalonil and SDS-3701 (R182281) in cereal grain.

Report:	K-CA 6.5.3/05. Gill JP and Sutra G. (2001), Residue levels in wheat and processed wheat products from trials carried out in France during 1999. Zeneca Study No 99JH076. Syngenta Report Number RJ3094B. (Syngenta File No: R44686/2186).

Characteristics

Reference	:	Gill & Sutra, 2001 (IIA 6.5.3, IIIA 8.5.3)	GLP	:	Yes
Type of study	:	Effect on residue levels (processing)	Guideline	:	-
Year of execution	:	1999-2000	Acceptability	:	Results reliable but not useful (initial residue too low in both trials)
Test substance	:	chlorothalonil 500 SC (YF10934); batch no. 882, 523 g a.s./L			

Study design

Two field trials with wheat were conducted in Beine Nauroy and Faverolles in Northern France (see Table B.7.7.6.2.22/02), with 2 foliar applications of chlorothalonil 500 SC at rates of 1.1 kg a.s./ha (1N). Wheat grain was harvested at 52 and 39 days after the last application, respectively, processed to various fractions (see Table below) and wholemeal flower was baked into bread using two different practical processes. The processes were performed on a pilot scale and mimicked typical commercial practices. Prior to processing the wheat grain was stored at the processing facility for 2 months at 15°C.

Methods of analysis

Wheat and processed fractions were analysed for chlorothalonil by method RAM 320/01 (GC/MS, LOQ 0.01 mg/kg). The LOQ was validated prior to the study in the process products white flour and wholemeal bread. Concurrent procedural recoveries were included at 0.02-0.2 mg/kg (grain), 0.02 mg/kg (wholemeal flour, white flour, bran and cleaned offal) and 0.02-0.05 mg/kg (wholemeal bread). Concurrent recoveries were acceptable: 72-108%. The reported LOQ was 0.01 mg/kg, although concurrent validation was only provided at a level of 0.02 mg/kg. The reported study results are accepted since (1) Pre-test validation in cereal grain for RAM 320/01 reported in Addendum 1 of the DAR and that in white flour and wholemeal bread was performed at the same laboratory which conducted the analytical phase of the processing trials, and was found to be acceptable at 0.01 mg/kg; (2) Results of concurrent validation at 0.02 mg/kg were good. (3) The peak at 0.02 mg/kg was

sufficiently high, so that at 0.01 mg/kg a well resolved peak would be expected; (4) The apparent residue in untreated samples was insignificant.

All residue data in the Table below were uncorrected for recovery of the analytical method.

Stability during homogenisation

During the preparation of the monograph, there were concerns about potential losses of chlorothalonil during sample preparation and homogenisation (reporting table from ECCO 111 residue peer review meeting [28.6.01]). Syngenta prepared a statement to address these concerns (Lister N and Smith M, chlorothalonil comments relating to metabolism and residues, ERA3753, 21 August 2001). It was demonstrated that losses of chlorothalonil were prevented by performing sample homogenisation in the presence of 10% v/w 0.1M sulphuric acid. This procedure was not followed during homogenisation of the samples of all studies. In the evaluation table of 29.09.2004, under point 5.3, it was concluded that *"instability of chlorothalonil during homogenisation is not an issue for the matrices of the only remaining use claimed by Syngenta (wheat grain and straw)"*. Instability should therefore not be an issue during this processing study, where only dry wheat matrices were analysed besides bread. The baking of bread however stops all enzymatic processes, including that leading to endogenous binding of chlorothalonil during sample preparation. Hence also the results of the bread samples are valid in this respect.

Storage and storage stability

Wheat and processed fractions were stored at ≤-18°C for up to 10 months prior to analysis for chlorothalonil. ~~*stable under frozen storage conditions (at -7°C or below) for at least 2 years (almonds nutmeats and hulls) to 4 years* In volume 3 of the DAR, under point B.7.6.2, it was demonstrated that *"chlorothalonil levels were (cherries, wheat, grain, tomatoes, cucumbers, carrots, soybeans and celery). In potatoes and peanuts, chlorothalonil levels tended to decrease upon long term storage, yet remained relatively stable during the first year and half year of storage, respectively."*. The above storage stability data include wheat grain, in which chlorothalonil is stable for 4 years.~~

Storage stability of chlorothalonil and metabolite SDS-3701 (R182281) has been discussed during the expert Peer Review Meeting (#164). It was concluded that based on the available data storage stability of chlorothalonil in cereal grain has been demonstrated for 62 days. For metabolite SDS-3701 no storage stability data in cereal grain in available and the meeting concluded that extrapolation from other high starch matrix commodities is not acceptable. A data gap is set. It is therefore concluded that the residue trials in wheat with chlorothalonil and SDS-3701 are not acceptable since the storage time of the samples exceeds the demonstrated storage stability.

Residues in control samples

Residues in all control samples produced from grain from untreated plots were <0.01 mg/kg.

Transfer factors

Transfer factors were calculated for chlorothalonil for all products. In all calculations the initial residue of chlorothalonil was taken to be the value determined in the grain pre-milling. In case the residue in

both the grain pre-milling and the processed product was <0.01 mg/kg, a processing factor was not calculated.

Results

The residues in wheat and processed wheat fractions and the processing factors are shown in the Table below. Transfer factors could only be estimated for the Faverolles trial, and in that case meaningful values were only determined for process fractions in which a significant increase in residue occurred. The process factors for chlorothalonil were 6 in bran and cleaned course bran, and <1 in all other products.

Table B.7.7.2.10-01 Residues and processing factors of chlorothalonil in wheat and processed wheat products

	Beine Nauroy trial		Faverolles trial	
crop part	**residue (mg/kg)**	**Transfer factor**	**residue (mg/kg)**	**Transfer factor**
grain (field)	<0.01		0.05	
grain pre-milling(D)	<0.01		0.01	
wholemeal flour	<0.01	na(C)	<0.01	<1
middlings	<0.01	na	<0.01	<1
break flour	<0.01	na	<0.01	<1
bran	<0.01	na	0.06	6
white flour	<0.01	na	<0.01	<1
cleaned course bran	<0.01	na	0.06	6
cleaned offal	<0.01	na	<0.01	<1
toppings	<0.01	na	<0.01	<1
type 550 flour	<0.01	na	<0.01	<1
bread (CBP)(A)	<0.01	na	<0.01	<1
bread (SMP)(B)	<0.01	na	<0.01	<1

(A) CBP: Chorleywood Bread process.

(B) SMP: Spiral Mixing Process.

(C) na = not applicable.

(D) Prior to processing the wheat grain was stored at the processing facility for 2 months at 15°C.

Conclusions

Beine Nauroy trial: initial residue and residues in process fractions too low (all <0.01 mg/kg).

Faverolles trial: processing factors 6 in bran and cleaned course bran, and <1 in all other products (initial residue 0.01 mg/kg, residue in process products <0.01 mg/kg).

However, the acceptability of the results is pending the demonstration of storage stability of chlorothalonil and SDS-3701 (R182281) in cereal grain.

Guidelines & Limitations

Comments were incorporated in the above summary. The study results are reliable but not useful since the initial residue was too low in both trials.

B.7.5.3.23 Processing in wheat

Previous evaluation	Submitted for the purpose of renewal
RMS remark	Acceptable for trial GB04-99-S079

Report:	K-CA 6.5.3/06. Gill JP and Myles P. (2001), Residue levels in wheat and processed wheat products from trials carried out in the UK during 1999. Zeneca, Jealott's Hill IRC,Bracknell Berkshire, UK. Zeneca Study No 99JH077. Syngenta Report Number RJ3095B. (Syngenta File No: R44686/2187).

Guidelines

Commission of the European Communities, Processing Studies; (SANCO 7035/V1/95 rev. 5 22/7/1997)

GLP

The study was carried out according to the principles of Good Laboratory Practice.

Executive Summary

Three residue field trials were conducted on wheat where two applications of a suspension concentrate formulation containing 500 g/L chlorothalonil, were made at a rate of 1.1 kg a.s./ha. Grain sampled at harvest was milled into white flour and wholemeal flour. The wholemeal flour was then used to bake bread by two typical commercial processes (the Chorleywood Bread Process and the spiral-mixing process). Wholemeal flour, middlings, break flour, bran, offal, toppings, white flour, type 550 flour and bread were analysed for residues of chlorothalonil.

Residues of chlorothalonil were not found above the LOQ of 0.01 mg/kg in flour or bread and would therefore not be expected to concentrate in these processed commodities. Residues were found to concentrate in bran, leading to a transfer factor of 4. Residues in the pre-processed grain were very low; a maximum residue of 0.02 mg/kg was found in the pre-processed grain and so the results of this study should be interpreted with caution.

A1. Test Materials

Test Material	YF10934
Description	Suspension concentrate formulation containing chlorothalonil
Purity	523 g/l
Batch number	882
Stability of test compound	The test substance has been shown to be stable under the storage and test conditions of the study

A2. Test Facilities

Field trials	UK
Milling and bread production	Campden and Chorleywood Food Research Association, Chipping Campden, Gloucestershire, GL55 6LD, UK
Analytical phase	Syngenta, Jealott's Hill International Research Centre, Bracknell, Berkshire, RG42 6EY, UK

B. STUDY DESIGN AND METHODS

B1. Processing phase

Three residue field trials were conducted on wheat in the UK during 1999 consisting of two decline trials and one harvest trial. In each trial two applications of a suspension concentrate formulation containing 500 g/L chlorothalonil, were made at a rate of 1.1 kg a.s./ha.

Grain sampled at harvest from the two decline trials (67 and 54 days after final application) were milled into white flour and wholemeal flour. Samples had been stored for 1-1.5 month before processing.

Storage stability of chlorothalonil and metabolite SDS-3701 (R182281) has been discussed during the expert Peer Review Meeting (#164). It was concluded that based on the available data storage stability of chlorothalonil in cereal grain has been demonstrated for 62 days.

The wholemeal flour was then used to bake bread by two typical commercial processes (the Chorleywood Bread Process and the spiral-mixing process). Samples of wholemeal flour, middlings, break flour, bran, offal, toppings, white flour, type 550 flour and bread were analysed for residues of chlorothalonil.

Samples were stored frozen for maximally 11 months, which is covered by storage stability studies (Vol. 1, 2.7.1)

B2. Analytical Phase

Samples of the raw agricultural commodity (grain) and various processed fractions were analysed for chlorothalonil using method RAM 320/01. The method involved extraction into acidified acetone followed by solid phase extraction and finally analysis by GC-MSD. The LOQ was 0.01 mg/kg for all commodities. A full method description and validation data are presented in B.5.1.2.5 (K-CA 4.1.2/22). and 6.5.3/05.

II. RESULTS AND DISCUSSION

A summary of the measured residues from the various processed fractions along with the calculated transfer factors are given in Table 7.5.3.23-1.

Table 7.5.3.23-1: Summary of chlorothalonil residues in wheat and processed wheat products

Commodity	Residues (mg/kg)			
	GB04-99-S078	Transfer Factor	GB04-99-S079	Transfer Factor
Grain	na[1]	Not applicable	0.02	-
Grain (Pre-milling)	<0.01		0.02	1
Wholemeal Flour	<0.01		0.01	0.5
Middlings	<0.01		<0.01	<0.5
Break Flour	<0.01		<0.01	<0.5
Bran	<0.01		0.08	4
White Flour	<0.01		<0.01	<0.5
Cleaned Course Bran	0.01	<1	0.06	3
Cleaned Offal	<0.01		0.03	1.5
Toppings	<0.01		<0.01	<0.5
Type 550 Flour	<0.01		<0.01	<0.5
Wholemeal Bread (CBP)[2]	<0.01		<0.01	<0.5
Wholemeal Bread (Spiral Mixed)	<0.01		<0.01	<0.5

[1] na – Not analysed

[2] CBP – Chorleywood Baking Process

Residues of chlorothalonil in grain and processed wheat products were all below the LOQ of 0.01 mg/kg for one trial. In the second trial residues of chlorothalonil were reduced in flour and bread compared to grain before processing. Residues were shown to concentrate in bran leading to a transfer factor of 4.

III. CONCLUSIONS

Residues of chlorothalonil would not be expected to concentrate in flour and bread. Residues were found to concentrate in bran, leading to a transfer factor of 4.

The applicant concluded that residues in the pre-processed grain were very low; a maximum residue of 0.02 mg/kg was found in the pre-processed grain and so the results of this study should be interpreted with caution.

Remark RMS: only the results from trial GB04-99-S079 have been used for calculation of transfer factors, since the residues in the pre-processed grain of trial GB04-99-S078 are <LOQ.

B.7.5.3.24 Processing in wheat

Previous evaluation	Submitted for the purpose of renewal
RMS remark	~~Acceptable~~ The acceptability of the results is pending the demonstration of storage stability of chlorothalonil and SDS-3701 (R182281) in cereal grain.

Report:	K-CA 6.5.3/07. North L., (2014a) Chlorothalonil – Residue study on wheat and processed products in Germany and northern France in 2011, Eurofins Agroscience Services Ltd, Melbourne, Derbyshire, UK. Report Number S11-00526-REG. (Syngenta File No. A7867A_11256)

Guidelines

FAO Guidelines on Producing Pesticide Residues Data from Supervised Trials (Rome, 1990).

Commission of the European Communities. General Recommendations for the Design, Preparation and Realization of Residue Trials (SANCO 7029/V1/95 rev. 5 22/7/1997).

Commission of the European Communities, Processing Studies; (SANCO 7035/V1/95 rev. 5 22/7/1997)

OECD Guidelines for the Testing of Chemicals : OECD Test Guideline 508: Magnitude of the Pesticide Residues in Processed Commodities.

European Commission Guidance Document on Residue Analytical Method (SANCO/825/00 rev. 8.1, 16 Nov 2010).

GLP

The study was carried out according to the principles of Good Laboratory Practice.

Executive Summary

Two residue trials on wheat were conducted in northern France and Germany during 2011. Two applications of chlorothalonil were applied to each plot as A7867A, a suspension concentrate (SC) formulation at a nominal rate of 3 kg a.s./ha (4 times intendend rate) separated by a 10 or 12 day interval. Samples of mature wheat grain were harvested 58 or 63 days after the last application of A7867A and analysed to determine residues of chlorothalonil and R182281.
Samples of mature wheat grain were processed into white flour (type 550), wholemeal flour, wholemeal bread, wheat germs, starch and gluten. One balance study and one follow-up study were carried out for each trial, covering each process. Therefore, a total of two balance and two follow-up studies were performed. The processed samples were analysed for residues of chlorothalonil and R182281.

Separate transfer factors for chlorothalonil and for R182281 were calculated. Mass balances were calculated for chlorothalonil only, due to the low levels of R182281 found in the samples before processing.
Sufficient data are available to allow transfer factors to be calculated for chlorothalonil and R182281 residues for wheat processed products. It is concluded that residues of chlorothalonil and R182281 would not be expected to concentrate in flour, bread, wheat germ, dried starch, dried gluten and gluten feed meal. Residues of chlorothalonil and R182281 would be expected to concentrate in course bran.

A1. Test Materials

Test Material	A7867A
Description	Suspension concentrate formulation containing chlorothalonil
Purity	495 g/L
Batch number	SAV0L00018
Stability of test compound	The test substance is assumed to be stable for the period of use in the study

A2. Test Facilities

Field trials	Württemberg, Germany	Loiret, France
Processing phase	Eurofins Agroscience Services GmbH, Carl-Goerdeler-Weg 5, D-21684 Stade, Germany	
Analytical phase	Eurofins Agroscience Services Ltd., Slade Lane, Wilson, Melbourne, Derbyshire DE73 8AG, UK	

B. STUDY DESIGN AND METHODS

B1. Processing phase

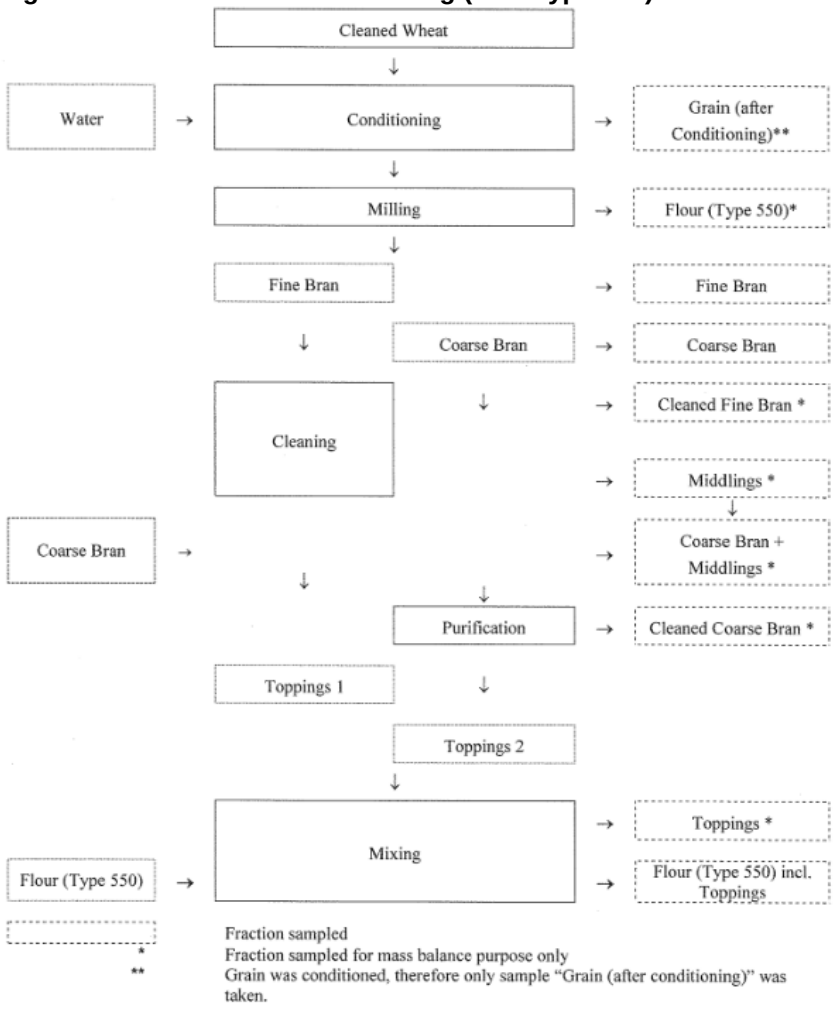
In field trials, commercially grown wheat was treated twice (at growth stage BBCH 53-57 and at BBCH 67-69) with a foliar spray of A7867A, at nominal application rates of 3.0 kg chlorothalonil/ha. The interval between the applications was 10 or 12 days.

Mature wheat grain was harvested 58 or 63 days after the last application and used for the production of white flour (type 550), wholemeal flour, wholemeal bread, wheat germ, starch and gluten.
Prior to each of the balance and follow-up processing studies, uncleaned wheat grain from each trial was analysed in duplicate to give a pre-processed residue value.
The un-cleaned wheat grain was cleaned using a 'sample cleaner' and for the balance studies, samples of cleaned wheat grain, impurities and shrivelled grain were taken for analysis. Part of the cleaned wheat was stored for subsequent processing to germ, starch and gluten.

White flour (type 550)

The cleaned grain was moistened overnight with tap water at 13.1°C – 19.9°C and the resulting conditioned grain had a moisture content of 15.7 – 16.7%. The conditioned grain was milled, producing flour (type 550), fine bran and coarse bran. The coarse bran was purified into cleaned coarse bran and 'toppings 2'. The fine bran was purified to produce cleaned fine bran, 'toppings 1' and 'middlings' fractions. The 'middlings' and coarse bran were then mixed at rate of 1:1 and the 'toppings 1 and 2' fractions were mixed to produce 'toppings'. Part of remaining 'toppings' and flour (type 550) were mixed.

Figure 7.5.3.24-1 Wheat milling (flour type 550)



Wholemeal flour

The cleaned grain was moistened overnight with tap water at temperature 13.1°C – 19.9°C and the resulting conditioned grain had a moisture content of 15.7 – 16.7%. The conditioned grain was milled to produce wholemeal flour.

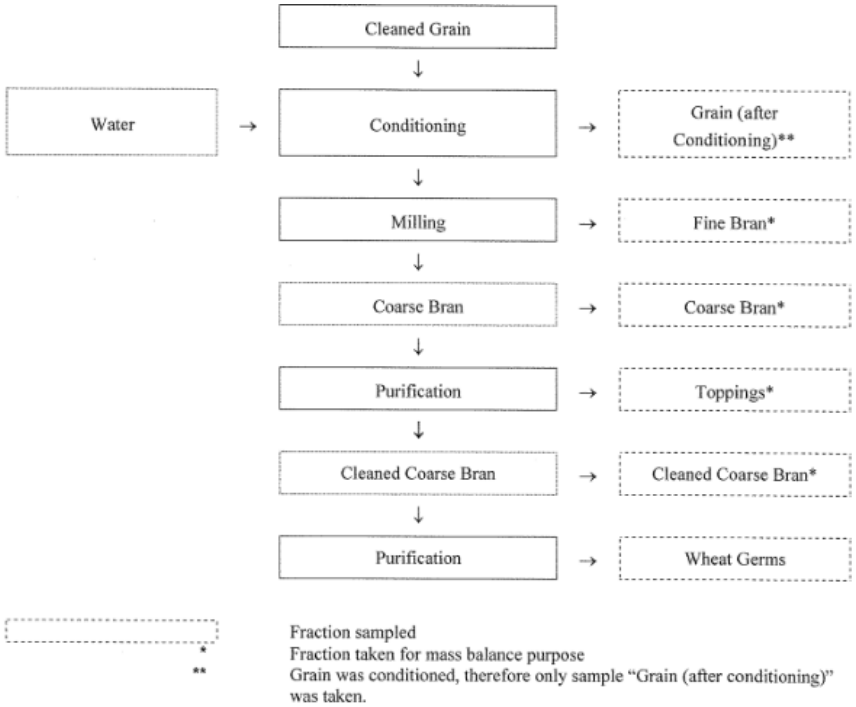
Wholemeal bread

Wholemeal flour was mixed with salt, sugar, plant fat, ascorbic acid, yeast and water in the kneading machine for 10 minutes. The dough was placed in an environmental cabinet, with a controlled climate at 24-27 °C and a relative humidity of 70-80% for 25 minutes. After the first fermentation process the dough was kneaded for 1 minute and then taken through a second fermentation process. After 15 minutes of fermentation, the dough was kneaded for an extra minute and then divided into loaves. The loaves were transferred into baking forms and placed under the same environmental conditions for 20 minutes. The dough was baked at 182-213°C for 24-26 minutes.

Wheat germ

The cleaned grain was moistened in a counter current mixer for 30 minutes and the resulting conditioned grain had a moisture content of 16.3 – 17.4%. The remaining conditioned grain was milled and the following fractions were produced: flour, fine bran, coarse bran. Only the coarse bran containing the germs was taken through the purification process. During the purification process cleaned coarse bran and 'toppings' were produced. The cleaned coarse bran fraction was cleaned again and the wheat germ fraction was taken.

Figure 7.5.3.24-2 Wheat germs flow chart

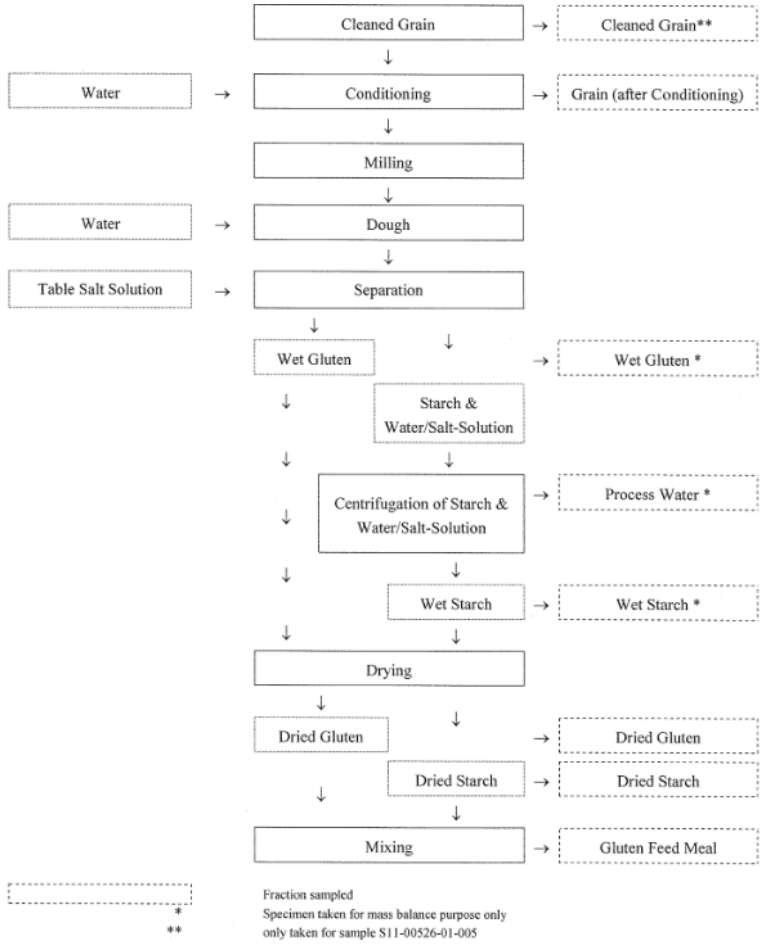


Starch and gluten

Flour (type 550) was mixed with water in a kneading machine for 5-10 minutes and then the dough was sprayed with 3% table salt solution in 4-6 washing steps. After each step, the starch/gluten

solution was drained into a vessel and retained for the starch/water centrifugation. The remaining fraction in the kneading machine, wet gluten, was sampled. The starch/water solution was centrifuged for 4 minutes and the fractions wet starch and process water were produced. Fractions of wet starch and wet gluten were dried to produce dried starch and dried gluten. Dried starch and dried gluten were mixed (ratio 1:1) to give gluten feed meal.

Figure 7.5.3.24-3 Starch and gluten flow chart



The following samples were taken for analysis in order to determine the residue accountability of chlorothalonil from the mass balance:

Uncleaned grain, cleaning impurities, shrivelled grain, cleaned grain, conditioned grain, fine bran, course bran, cleaned fine bran, 'middlings', coarse bran + 'middlings', cleaned coarse bran, 'toppings', flour type 550, wholemeal flour, flour type 550 + toppings, dough, wholemeal bread, wheat germ, wet starch, wet gluten, starch washing water, dried starch, dried gluten and gluten feed meal.

Transfer factors were determined from the mass balance and follow up studies for the following samples:

fine bran, course bran, flour type 550, wholemeal flour, dough, wholemeal bread, wheat germ, dried starch, dried gluten and gluten feed meal.

Samples were stored frozen for a maximum period of 12 months from sampling to analysis.~~, which is covered by storage stability studies (Vol. 1, 2.7.1).~~

Storage stability of chlorothalonil and metabolite SDS-3701 (R182281) has been discussed during the expert Peer Review Meeting (#164). It was concluded that based on the available data storage stability of chlorothalonil in cereal grain has been demonstrated for 62 days. For metabolite SDS-3701 no storage stability data in cereal grain in available and the meeting concluded that extrapolation from other high starch matrix commodities is not acceptable. A data gap is set. It is therefore concluded that the residue trials in wheat with chlorothalonil and SDS-3701 are not acceptable since the storage time of the samples exceeds the demonstrated storage stability.

B2. Analytical Phase

Samples of the raw agricultural commodity (grain) and various processed fractions were analysed for chlorothalonil and R182281 using method GRM005.01A with minor modifications. The method involved extraction with acetone/5M sulphuric acid (95:5 v/v), dilution with water followed by SPE clean up for chlorothalonil or taking up in acetonitrile:water (50/50 v/v) for R182281. The LOQ was 0.01 mg/kg for both analytes in all commodities. A full method description and validation data are presented in B.5.2.1 (K-CA 4.2/01-K-CA 4.2/07). As part of this study, the analytical method for chlorothalonil and R182281 was successfully validated in grain and its processing matrices.

II. RESULTS AND DISCUSSION

Procedural recoveries for the method of analysis are presented in Table 7.5.3.24-1 for grain and its processing matrices.

Table 7.5.3.24-1: Summary of recovery data for chlorothalonil and R182281 in wheat grain and its processed matrices

Commodity	Fortification concentration (mg/kg)	Chlorothalonil		R182281	
		Mean recovery (%) n=5	RSD (%) n=5	Mean recovery (%) n=5	RSD (%) n=5
Grain	0.01	99	4	102	17
	0.1	113	3	103	16
Flour	0.01	96	3	105	1
	0.1	96	10	94	4
Bran	0.01	86	2	93	5
	0.1	86	2	92	1
Dough	0.01	91	3	98	1
	0.1	99	1	100	2
Bread	0.01	92	2	95	5
	0.1	106	5	88	5
Dry gluten	0.01	91	5	109	5
	0.1	93	1	101	3
Dry starch	0.01	71	17	102	4
	0.1	84	1	104	1

A summary of the measured residues in the various processed fractions is given in Table 7.5.3.24-2.

Table 7.5.3.24-2: Summary of chlorothalonil and R182281 residues in wheat processed commodities from trials in Germany and northern France

Commodity	Residues (mg/kg)							
	Balance 1		Follow-up 1		Balance 2		Follow-up 2	
	chloro-thalonil	R182281	chloro-thalonil	R182281	chloro-thalonil	R182281	chloro-thalonil	R182281
Mean wheat grain (RAC)	0.07	0.02	0.04	0.01	0.04	<0.01	0.12	0.01
Cleaning								
Cleaned grain	0.07	0.02	0.05	0.02	0.03	<0.01	0.03	<0.01
Impurities	16.61	2.24	--	--	3.41	0.21	--	--
Shrivelled grain	0.16	0.04	--	--	0.13	0.02	--	--
White flour production								
Grain (conditioned)	0.04	0.02	0.03	0.02	0.01	<0.01	0.01	<0.01
Flour type 550	<0.01	<0.01	--	--	<0.01	<0.01	--	--
Fine bran	0.03	0.02	0.02	0.01	<0.01	<0.01	0.01	<0.01
Coarse bran	0.18	0.09	0.17	0.06	0.05	0.02	0.07	0.03
Cleaned fine bran	0.08	0.05	--	--	0.02	0.01	--	--
Middlings	0.03	0.02	--	--	<0.01	<0.01	--	--
Coarse bran + middlings	0.10	0.06	--	--	0.02	0.01	--	--
Cleaned coarse bran	0.12	0.07	--	--	0.04	0.02	--	--
Toppings	0.08	0.05	--	--	0.03	0.02	--	--
Flour type 550 + toppings	0.05	0.04	0.05	0.03	0.02	0.01	0.01	<0.01
Wholemeal flour and bread production								
Grain (conditioned)	0.05	0.02	0.03	0.02	0.01	<0.01	0.02	<0.01
Wholemeal flour	0.03	0.03	0.03	0.02	0.01	<0.01	<0.01	<0.01
Dough	<0.01	0.02	<0.01	0.01	<0.01	<0.01	<0.01	<0.01
Wholemeal bread	<0.01	0.02	<0.01	0.01	<0.01	<0.01	<0.01	<0.01
Wheat germ								
Grain (conditioned)	0.03	0.02	0.04	0.02	0.01	<0.01	0.02	<0.01
Fine bran	0.07	0.04	--	--	0.02	0.01	--	--
Coarse bran	0.13	0.06	--	--	0.06	0.02	--	--
Toppings	0.10	0.07	--	--	0.03	0.02	--	--
Cleaned coarse bran	0.13	0.07	--	--	0.04	0.02	--	--
Wheat germ	0.04	0.02	0.05	0.03	<0.01	<0.01	0.01	<0.01
Starch and gluten								
Grain (conditioned)	0.05	0.01	0.04	0.01	0.01	<0.01	0.02	<0.01
Wet starch	<0.01	<0.01	--	--	<0.01	<0.01	--	--
Wet gluten	<0.01	<0.01	--	--	<0.01	<0.01	--	--
Process water	<0.01	<0.01	--	--	<0.01	<0.01	--	--

Commodity	Residues (mg/kg)							
	Balance 1		Follow-up 1		Balance 2		Follow-up 2	
	chloro-thalonil	R182281	chloro-thalonil	R182281	chloro-thalonil	R182281	chloro-thalonil	R182281
Dried starch	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
Dried gluten	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
Gluten feed meal	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01

--: not analysed in follow up study.

Residues of chlorothalonil and R182281 in the individual pre-processed grain samples were in the range 0.02 to 0.14 mg/kg and <0.01 to 0.02 mg/kg, respectively. No residues of chlorothalonil or R182281 were found at or above the LOQ (0.01 mg/kg) in any of the untreated wheat grain or untreated processed commodities.

Processing into white flour (type 550)

Residues of chlorothalonil in cleaned wheat grain were 0.03 to 0.07 mg/kg, in fine bran were <0.01 to 0.03 mg/kg, in coarse bran were 0.05 to 0.18 mg/kg and in white flour type 550 including toppings were 0.01 to 0.05 mg/kg.

Residues of R182281 in cleaned wheat grain were <0.01 to 0.02 mg/kg, in fine bran were <0.01 to 0.03 mg/kg, in coarse bran were 0.02 to 0.09 mg/kg and in white flour type 550 including toppings were <0.01 to 0.04 mg/kg.

The mass balance during white flour type 550 including toppings production was 100% of the initial wheat grain mass; 80% of the initial chlorothalonil residue was recovered during the process.

Processing into wholemeal flour and bread

Residues of chlorothalonil in grain after conditioning were 0.01 to 0.05 mg/kg, were <0.01 to 0.03 mg/kg for wholemeal flour and were < 0.01 mg/kg in wholemeal bread.

Residues of R182281 in grain after conditioning were <0.01 to 0.02 mg/kg, were <0.01 to 0.03 mg/kg for wholemeal flour and were < 0.01 mg/kg to 0.02 mg/kg in wholemeal bread.

The mass balance during wholemeal flour production was 100% of the initial wheat grain mass; 77% to 104% of the initial chlorothalonil residue was recovered during the process. The mass balance during wholemeal bread production was 100% of the initial wheat grain mass; 76% to 103% of the initial chlorothalonil residue was recovered during the process.

Processing into wheat germ

Residues of chlorothalonil in grain after conditioning were 0.01 to 0.04 mg/kg and in wheat germ were <0.01 to 0.05 mg/kg. Residues of R182281 in grain after conditioning were <0.01 to 0.02 mg/kg and in wheat germ were <0.01 to 0.03 mg/kg.

The mass balance during wheat germ production was 99-100% of the initial wheat grain mass; 77-103% of the initial chlorothalonil residue was recovered during the process.

Processing into starch and gluten

Residues of both chlorothalonil and R182281 were < 0.01 mg/kg in all dried starch, dried gluten and gluten feed meal samples. The mass balance during gluten feed meal production was 99-100% of the initial wheat grain mass; 79- 105% of the initial chlorothalonil residue was recovered during the process.

The mean transfer factors for each commodity for chlorothalonil and R182281 were calculated and are presented in Table 7.5.3.24-3 and Table 7.5.3.24-4, respectively.

Table 7.5.3.24-3: Summary of chlorothalonil transfer factors into processed wheat products

Commodity	Transfer Factor	Median Transfer Factor
Coarse bran	2.57, 1.25, 4.25, 0.58	1.91
Fine bran	0.43, <0.25, 0.50, 0.08	0.34
White flour (550) + toppings	0.71, 0.50, 1.25, 0.08	0.61
Wholemeal flour	0.43, 0.25, 0.75, <0.08	0.34
Wholemeal bread	<0.14, <0.25, <0.25, <0.08	<0.20
Wheat germs	0.57, <0.25, 1.25, 0.08	0.41
Dried starch	<0.14, <0.25, <0.25, <0.08	<0.20
Dried gluten	<0.14, <0.25, <0.25, <0.08	<0.20
Gluten feed meal	<0.14, <0.25, <0.25, <0.08	<0.20

Transfer factor = residue in processed commodity/mean residue in grain (e.g. for coarse bran 0.18/0.07 = 2.57)

Table 7.5.3.24-4: Summary of R182281 transfer factors into processed wheat products

Commodity	Transfer Factor*	Median Transfer Factor*
Coarse bran	4.5, 6, 3.0	4.5
Fine bran	1.0, 1.0, <1.0	1.0
White flour (550) + toppings	2.0, 3.0, <1.0	2.0
Wholemeal flour	1.5, 2.0, <1.0	1.5
Wholemeal bread	1.0, 1.0, <1.0	1.0
Wheat germs	1.0, 3.0, <1.0	1.0
Dried starch	<0.5, <1.0, <1.0	<1.0
Dried gluten	<0.5, <1.0, <1.0	<1.0
Gluten feed meal	<0.5, <1.0, <1.0	<1.0

Transfer factor = residue in processed commodity/mean residue in grain (e.g. for coarse bran 0.09/0.02 = 4.5)

* Data from Balance study 2 were not used in the calculation of transfer factors as residues of R182281 were <0.01 in the grain before processing.

Residues of chlorothalonil in coarse bran were higher (maximum 0.18 mg/kg) than in the wheat grain samples prior to processing. The mean transfer factor was 2.2, indicating that residues of chlorothalonil are concentrated in coarse bran. Residues of chlorothalonil in fine bran were lower (maximum 0.03 mg/kg) than in the wheat grain samples prior to processing, leading to an overall mean transfer factor of <0.32.

Residues of R182281 in coarse bran were higher (maximum 0.09 mg/kg) than in the wheat grain samples prior to processing leading to an overall mean transfer factor of 4.5. Residues of R182281 in fine bran were the same as in the wheat grain samples prior to processing.
Residues of chlorothalonil in both white and wholemeal flour were lower than in the wheat grain samples prior to processing indicating that residues do not concentrate in flour. Residues of R182281 were higher in white flour (maximum 0.04 mg/kg) than in the wheat grain samples prior to processing in two studies, but were <0.01 mg/kg in one study leading to an overall mean transfer factor of <2.0. Residues of R182281 in wholemeal flour were the same as in the wheat grain samples prior to processing.
Residues of chlorothalonil were < 0.01 mg/kg in wholemeal bread leading to overall transfer factors < 0.18. Residues of R182281 were the same or lower than in the wheat grain samples prior to processing leading to a mean transfer factor of <1.
Residues of chlorothalonil in wheat germ were lower (maximum 0.05 mg/kg) then wheat grain samples prior to processing, leading to a mean transfer factor <0.54.
Residues of R182281 in wheat germ were higher (maximum 0.03 mg/kg) than wheat grain samples prior to processing in one study; however were lower or the same than in the wheat grain samples prior to processing in the other studies. Overall the mean transfer factor was <1.6.
Residues of chlorothalonil and R182281 in dried starch, dried gluten and gluten feed meal were all <0.01 mg/kg, leading to overall mean transfer factors <0.8.

III. CONCLUSIONS

Sufficient data are available to allow transfer factors to be calculated for chlorothalonil and R182281 residues for wheat processed products. It is concluded that residues of chlorothalonil and R182281 would not be expected to concentrate in flour, bread, wheat germ, dried starch, dried gluten and gluten feed meal. Residues of chlorothalonil and R182281 would be expected to concentrate in coarse bran. However, the acceptability of the results is pending the demonstration of storage stability of chlorothalonil and SDS-3701 (R182281) in cereal grain.

B.7.5.3.25 Processing in wheat

Previous evaluation	Submitted for the purpose of renewal
RMS remark	~~Acceptable for chlorothalonil only~~ The acceptability of the results is pending the demonstration of storage stability of chlorothalonil and SDS-3701 (R182281) in cereal grain.

Report:	K-CA 6.5.3/08. Sala A. (2015), Determination of chlorothalonil and its metabolites SDS3701 and R613636 residues in raw agricultural commodity winter wheat and processed commodity (grain, flour, total bran, wholemeal flour, wholemeal bread, wheat germ) following two applications of chlorothalonil 500 SC. Syngenta File No: R044686_11359. Report Number RAU-007-14.

Guidelines

EU Guidance documents on residue analytical methods SANCO/3029/99, rev. 4 (11/07/2000)
EC guidance document 1607/VI/97 rev.2, 10/6/1999

GLP

The study was carried out according to the principles of Good Laboratory Practice.

Executive Summary

Outdoor wheat grown in field trials in Italy and Poland was treated with Chlorothalonil 500 SC. The plots were sprayed twice with nominal application rates of 3.0 kg chlorothalonil/ha. Samples of mature wheat grain were harvested 47 or 55 days after the last application and then analysed to determine residues of chlorothalonil, R182281 and R613636.

Additional samples of the mature wheat grain taken at 47 or 55 days from the treated plots were used for the production of white flour, wholemeal flour, bran, wheat germs and wholemeal bread, and the processed samples were analysed for residues of chlorothalonil, R182281 and R613636.

Residues in the pre-processed grain were very low; residues of R182281 and R613636 were below the LOQ in grain and all processed commodities meaning that transfer factors could not be determined. Transfer factors determined for the various process fractions were calculated for chlorothalonil only. The study showed that residues of chlorothalonil would not be expected to concentrate in white flour, wholemeal flour and wheat germs. Residues of chlorothalonil may be expected to concentrate in bran and wholemeal bread, although as residues in the pre-processed grain were very low the results of this study should be interpreted with caution.

A1. Test Materials

Test Material	Chlorothalonil 500 SC
Description	Suspension concentrate formulation containing chlorothalonil
Purity	507 g/L
Batch number	PN1911
Stability of test compound	The test substance has been shown to be stable under the storage and test conditions of the study

A2. Test Facilities

Field trial	Roccabianca (PR) Italy	Wielkopolska Poland
Milling and bread production	Biospheres Processing Laboratory, Via Vittoria Veneto, 26857 Salerano sul Lambro (LO), Italy	
Analytical phase	Biospheres Residues Analysis Unit, Via Vittoria Veneto, 26857 Salerano sul Lambro (LO), Italy	

B. STUDY DESIGN AND METHODS

B1. Processing phase

In 2014, in two field trials, wheat was treated twice with a foliar spray of the formulation at nominal application rates of 3.0 kg chlorothalonil/ha. The first application was at BBCH59 (Italyl) or BBCH39

(Poland), and the second application was at BBCH69. The interval between the applications was 9 (Italy) or 25 (Poland) days.
Mature wheat grain was harvested 47 or 55 days after the last application at normal commercial harvest, and used for the production of flour, bran, bread and wheat germ. Both processing trials were carried out as follow-up trial: only the starting (grain) and final final products (whole meal bread, whole meal flour, flour type 550 and wheat germs) obtained during the processing procedures were sampled.
The grains were milled and sieved to give white flour (550 type) and bran. Separate aliquots of grains were milled to give wholemeal flour that was used to bake bread by a typical commercial process.
To obtain wheat germs, the grains were spread homogenously in a thin layer, covered with water and left to germinate for 11-12 days. The wheat germs were then removed and dried at room temperature.

Figure 7.5.3.25-1 Process flow chart to get flour type 550

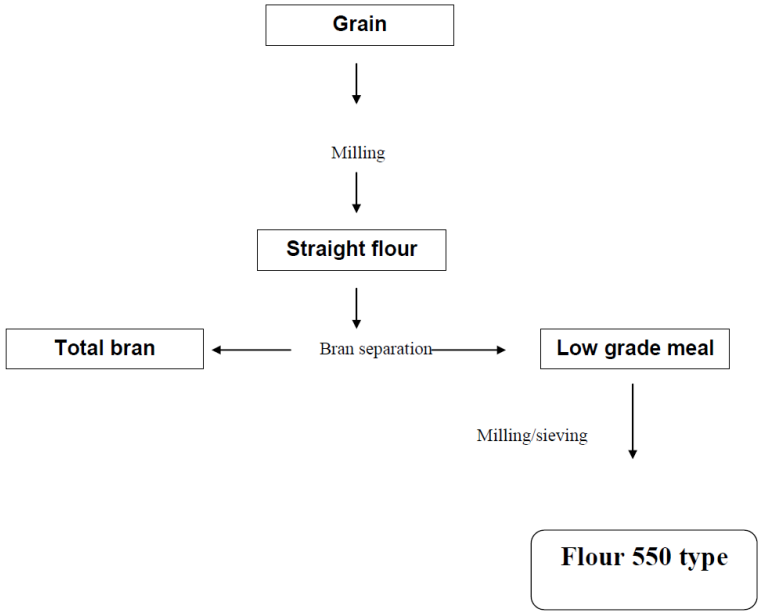


Figure 7.5.3.25-2 Process flow chart to get whole meal flour and whole meal bread

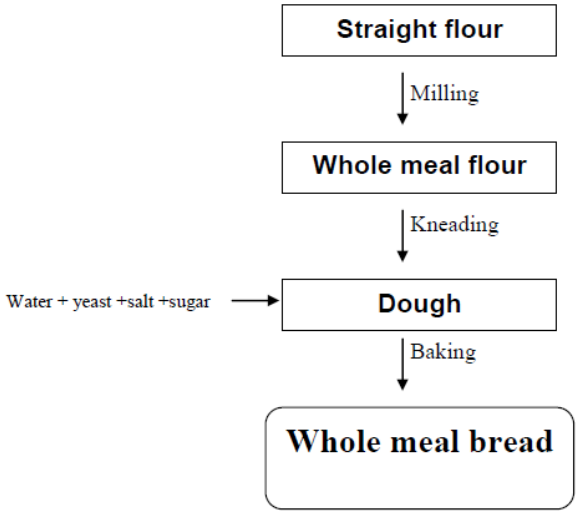
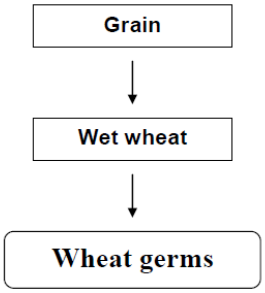


Figure 7.5.3.25-3 Process flow chart to get wheat germs



B2. Analytical Phase

Samples of the raw agricultural commodity (grain) and various processed fractions were analysed for chlorothalonil and R182281 using methods described in study BIU-014-14 (see 7.3.3), and for R613636 using an analytical method not further referenced. For chlorothalonil, method RAU076-00 consists in an extraction using ethyl acetate acidified, performed using an orbital shaker and an ultrasonic bath. Final analysis will be carried out using a Gas Chromatograph equipped with a µ-ECD detector. For R182281 the analytical method consisted of extraction with methanol and analysis with LC-MS/MS. For R613636, the samples were extracted with acidified acetone. After centrifugation the samples were diluted with ultrapure water and purified by solid phase extraction on an Oasis HLB cartridge. The purified extracts were then analysed with an HPLC system coupled with a triple quadrupole mass analyser (LC-MS/MS).

The LOQ was 0.01 mg/kg for chlorothalonil and R613636, and 0.02 mg/kg for R182281 in all commodities. Full method descriptions and validation data are presented in B.5.1.2.5 (K-CA 4.1.2/33). Transfer factors for the processed commodities were determined.

Samples were stored for maximally 3 months before the analysis of chlorothalonil, 5 months for R182281, and 6 months until analysis of R613636. These time periods are covered by storage stability studies for R613636 only(Vol. 1, 2.7.1).

Storage stability of chlorothalonil and metabolite SDS-3701 (R182281) has been discussed during the expert Peer Review Meeting (#164). It was concluded that based on the available data storage stability of chlorothalonil in cereal grain has been demonstrated for 62 days. For metabolite SDS-3701 no storage stability data in cereal grain in available and the meeting concluded that extrapolation from other high starch matrix commodities is not acceptable. A data gap is set. It is therefore concluded that the residue trials in wheat with chlorothalonil and SDS-3701 are not acceptable since the storage time of the samples exceeds the demonstrated storage stability.

II. RESULTS AND DISCUSSION

The accuracy of the method of analysis was evaluated by recoveries, which are presented in Table 7.5.3.25-1 for grain and its processing matrices. Procedural recoveries are presented in Table 7.5.3.25-2. The correlation coefficient R^2 was >0.99 for all analytes.

Table 7.5.3.25-1: Summary of recovery data for chlorothalonil and R182281 in wheat grain and its processed matrices

Commodity	Fortification	Chlorothalonil	R182281

	concentration (mg/kg)	Mean recovery % (individual values) n=3	RSD (%) n=3	Mean recovery % (individual values) n=3	RSD (%) n=3
Total bran	0.01	76.10 (85.45; 78.33; 64.52)	13.98	98.56 (95.13; 107.92; 92.64)	8.32
	0.1	91.54 (93.04; 92.57; 88.73)	2.59	83.30 (86.26; 78.63; 85.02)	4.91
Flour	0.01	82.07 (84.90; 81.40; 79.91)	3.12	88.78 (91.29; 89.63; 85.42)	3.41
	0.1	84.39 (89.24; 83.56; 80.35)	5.34	72.78 (66.40; 72.04; 79.90)	9.32
Whole meal flour	0.01	96.93 (99.53; 97.70; 93.55)	3.16	94.77 (104.82; 102.33; 77.16)	16.15
	0.1	93.16 (94.44; 90.41; 94.64)	2.57	100.28 (101.01; 96.38; 103.44)	3.58
Whole meal bread	0.01	98.94 (82.53; 115.74; 98.56)	16.79	89.04 (87.81; 85.12; 94.20)	5.24
	0.1	98.58 (89.28; 91.18; 88.27)	1.65	90.15 (84.76; 82.79; 102.91)	12.30
Wheat germ	0.01	88.26 (92.47; 87.39; 84.91)	4.37	85.13 (96.20; 78.86; 80.33)	11.30
	0.1	88.93 (88.13; 89.85; 88.80)	0.97	74.61 (71.63; 77.69; 74.51)	4.06

Table 7.5.3.25-2: Summary of procedural recoveries for chlorothalonil, R182281 and R613636 in wheat grain and its processed matrices

Commodity	Fortification concentration (mg/kg)	Analyte	Recovery %
Grain	0.01	Chlorothalonil	99.47
	0.1		92.14
Grain	0.01	R182281	92.81
	0.1		96.34
Grain	0.01	R613636	138.97

Commodity	Fortification concentration (mg/kg)	Analyte	Recovery %
	0.1		82.26
Total bran	0.01	R613636	104.09
	0.1		127.10
Whole meal bread	0.01	R613636	87.74
	0.1		85.56

A summary of the measured residues from the various fractions for the processed fraction is given in Table 7.5.3.25-3.

Table 7.5.3.25-3: Summary of chlorothalonil, R182281 and R613636 residues in wheat and processed commodities from trials conducted in Italy and Poland

Trial:	Residues (mg/kg)					
	Italy			Poland		
Commodity	Chlorothalonil	R182281	R613636	Chlorothalonil	R182281	R613636
Grain (RAC)	0.02	<0.02	<0.01	0.01	<0.02	<0.01
Grain for processing	0.01	<0.02	<0.01	0.01	<0.02	<0.01
Bran	0.02	<0.02	<0.01	<0.01	<0.02	<0.01
Flour	<0.01	<0.02	<0.01	<0.01	<0.02	<0.01
Wholemeal flour	<0.01	<0.02	<0.01	<0.01	<0.02	<0.01
Wholemeal bread	0.02	<0.02	<0.01	<0.01	<0.02	<0.01
Wheat germ	<0.01	<0.02	<0.01	<0.01	<0.02	0.02

The mean transfer factors for each commodity for chlorothalonil were calculated and are presented in Table 7.5.3.25-4. Transfer factors for R182281 and R613636 were not calculated as residues in the grain before processing and in all processed commodities were below the LOQ (0.02 mg/kg and 0.01 mg/kg, respectively).

Table 7.5.3.25-4: Summary of transfer factors into processed wheat products

	Chlorothalonil	
Commodity	Transfer Factors	Median Transfer Factor
Bran	2.0, <1.0	1.5
Flour	<1.0, <1.0	<1.0
Wholemeal flour	<1.0, <1.0	<1.0
Wholemeal bread	2.0, <1.0	1.5
Wheat germ	<1.0, <1.0	<1.0

Transfer factor = residue in processed commodity/mean residue in grain for processing (e.g. for bran 0.02/0.01 = 2.0).

III. CONCLUSIONS

Residues in the pre-processed grain were very low; residues of R182281 and R613636 were below the LOQ in grain and all processed products meaning that transfer factors could not be determined. Transfer factors determined for the various process fractions were calculated for chlorothalonil only. Residues of chlorothalonil would not be expected to concentrate in white flour, wholemeal flour and wheat germs. Residues of chlorothalonil may be expected to concentrate in bran and wholemeal bread, although as residues in the pre-processed grain were very low the results of this study should be interpreted with caution.

However, the acceptability of the results is pending the demonstration of storage stability of chlorothalonil and SDS-3701 (R182281) in cereal grain.

B.7.6 Residues in rotational crops

During the initial peer review, a confined rotational crop study was conducted using [phenyl-U-^{14}C]-chlorothalonil to address the potential uptake and metabolism of chlorothalonil residues into succeeding or rotated crops following an application to the primary crop. Additional confined crop rotation studies have also been submitted. These studies have been evaluated within the framework of the renewal of chlorothalonil.

During the initial peer review, also field studies were conducted in the USA to measure levels of chlorothalonil residues in succeeding or rotated crops following an application to the primary crop. ~~An~~ Two additional field crop rotation ~~study has~~ studies have been conducted for the renewal of chlorothalonil, which ha~~s~~ves been evaluated in this RAR.

B.7.6.1 Metabolism in rotational crops

B.7.6.1.1 Metabolism in rotational crops, study 1

Previous evaluation	In DAR
RMS remark	Acceptable

Characteristics

reference	:	Zeneca, Nelsen et al., 1995	treatment	:	fortified
type of study	:	rotational crop study	rate	:	9.5 mg a.i/kg
year of execution	:	1982	formulation	:	methylene chloride
test substance	:	^{14}C-uniformly ring labelled chlorothalonil (specific activity 25.5 dpm/µg, radiochemical purity 99.9%)	GLP statement	:	no
crop/commodity	:	lettuce, carrots and spring wheat	guidelines	:	not applicable

Study design

^{14}C-Uniformly ring labelled chlorothalonil (specific activity 25.5 dpm/µg; radiochemical purity 99.9%) was formulated and applied in the form of a solution (vehicle: methylene chloride). The solution was uniformly incorporated at an exaggerated rate of 9.5 mg a.i/ kg, in a sieved (600 micron screen) sandy loam soil. The soil was aged under aerobic conditions in the dark with a 24 hour temperature cycle of 14 hrs at 27-29°C and 10 hours at 16-24 °C. After 30 and 88 days of aerobic aging, 4.0 kg subsamples of the amended soil were transferred to 6 fibre planting pots (20.3 cm in diameter and 25.2 deep). Duplicate pots were planted with spring wheat, carrots and lettuce. After germination, the pots were transferred into a greenhouse. The mature crops were harvested, separated into appropriate fractions (i.e wheat to grain, chaff and straw; carrots to tops and roots), frozen and ground in an analytical mill. After combusting/LSC to determine total ^{14}C-residue levels, crops were processed. In addition, soil samples were analysed for total residue levels and for quantification of specific components of the residue. The study was performed in Ohio (USA) in 1982-1983.

Results

The total soil residue levels at 30 and 88 days after treatment were not reported. At 30 days after soil treatment, somewhat more than 11% of the total soil residue was identified as the parent compound, which declined to 5% at 88 days. SDS 46851 accounted for almost 25% of the total soil residue at both preplant intervals. Other soil metabolites (SDS 3701, SDS 19221, SDS 47523/4 and SDS 47525) each accounted for less than 10% of the total soil residue.

The total residue levels in crops are summarized in the table below.

Table 7.9.1.1 Residue levels in lettuce, carrots and spring wheat (in mg a.i eq/kg)

Part of plant	Rotation interval (DAT)	Residues (mg a.i. eq/kg)		
		Lettuce	Carrot	Spring Wheat
Leaves	30 88	3.3 1.0		
Roots	30 88		1.0 0.9	
Tops	30 88		2.2 3.2	
Grain	30 88			3.3 21.6
Chaff	30 88			7.8 43.9
Straw	30 88			51.9 63.8

Residues present in lettuce, carrot and wheat samples from the rotational crop study were further characterized and identified. For all crops and crop parts, between approximately 45 and 80% of the extractable residue was organo extractable and 20-55% was aqueous soluble. The major identified metabolite was SDS-46851 (up to approximately 2.0 mg chlorothalonil equivalents/kg lettuce at a preplant interval of 30 days and 0.4 mg equivalents at a replant interval of 88 days; up to 0.63 and 1.1 mg equivalents/kg in carrot roots and tops, respectively, and up to 16.5 and 33.1 mg equivalents/kg in wheat grain and straw respectively; SDS-46851 was partly present in conjugated form). In addition, small amounts of SDS-3701 were detected (below 0.1 mg equivalents/kg lettuce, around or below 0.05 mg equivalents/kg carrot root, up to 0.4 mg equivalents/kg carrot top, up to 6.3 mg equivalents in wheat straw and below 0.5 mg equivalents in wheat grain; SDS-3701 was mainly present in conjugated form). The parent compound chlorothalonil was not detected in any plant part. Only 2.8 to 22.6 % of the total residue remained unextracted.

Conclusions

At 30 and 88 days after soil treatment with ^{14}C-uniformly ring labelled chlorothalonil, the major soil residue compound was SDS-46851 (almost 25% of the total soil residues), followed by the parent compound accounting for 11 and 5% of the total residue at the respective treatment days. SDS-46851 was the major residue compound identified in rotational crop samples. SDS-3701 was present at low levels. Parent compound was not detected in crop samples.

Limitations

Total soil residue levels at 30 and 88 days after treatment were not reported. As such, the present study cannot be fully evaluated.

B.7.6.1.2 Metabolism in rotational crops, study 2

Previous evaluation	Submitted for the purpose of renewal
RMS remark	Acceptable

Report:	K-CA 6.6.1/01. Rizzo F. and Ferrario F. (2005), Uptake, translocation and metabolism of 14C-Chlorothalonil in rotated crops of spring wheat, carrots and lettuce. Isagro Ricerca Srl, Novara, Italy. Study number MEF.03.03. Syngenta File No. R044686_11201.

Guidelines

None

GLP

The study was carried out according to the principles of Good Laboratory Practice.

EXECUTIVE SUMMARY

A confined rotational crop metabolism study was conducted to provide information on the magnitude and nature of residues of chlorothalonil in following crops. [Phenyl-U-^{14}C]-chlorothalonil in acetonitrile/water was applied drop-wise at 7.5 kg a.s/ha to separate containers of soil prior to sowing. At each rotational interval of 30, 120 and 365 days after application (DAT), a representative cereal (spring wheat), leafy vegetable (lettuce) and root vegetable (carrot) were sown into the soil. All crops were grown under field conditions and harvested at maturity. Harvested crops were separated into commodities of representative food and feed items (wheat forage, straw and grain; mature lettuce; carrot foliage and roots). The total radioactive residue concentration (TRR, mg a.s. equivalents per kg of commodity, mg/kg) was quantified and characterised

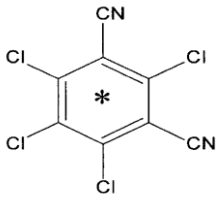
Radioactivity was measured by combustion and LSC. Samples were extracted using various solvents. Sample extracts were analysed by normal and reverse-phase TLC, using two solvent systems for each stationary phase. Selected aqueous phases were submitted to enzymatic (β-glucosidase) hydrolysis. Post-extraction solids were refluxed successively with water, alkali (NaOH) and acid (HCl). Metabolite identification was performed using co-chromatography with certified reference compounds.

The TRR (mg/kg chlorothalonil equivalents) by combustion analysis in all rotational commodities were ≥0.05 mg/kg. The highest TRR were observed in 30 and 120 DAT mature lettuces (0.241 mg/kg), 120 DAT carrot leaves (1.516 mg/kg), carrot roots (0.431 mg/kg) and wheat straw (25.12 mg/kg), and 365 DAT wheat forage (1.407 mg/kg) and wheat grain (2.23 mg/kg). In general, TRR tended to remain at similar levels across all plant-back intervals. The majority of the radioactive residues were extractable, accounting for 65 to 95% TRR.

In all crops the majority of the radioactive residue was assigned to the metabolites R611965 and R417888, coeluting together. Up to 40% TRR was assigned as conjugated material. Enzyme hydrolysis of the extracts released radioactivity indicating that the conjugated material was made up of glucosyl conjugates of R611968, R613636 and “compound C15”. Other metabolites identified, included R611553, R182281 and R612636, and represented minor percentages of the TRR. R611968 accounted for up to 10% TRR in grain.

The post-extraction solids (PES) were 8.9 - 23% TRR in carrot root, 21.4 – 32.7% TRR in carrot leaves and 22.4% TRR in lettuce. Post extraction solids for cereal samples were 7.8 – 10.5% TRR and 4.6 – 15.7% TRR for grain and straw, respectively. A large proportion of this radioactivity was released by aqueous reflux (ranging between 3.62 and 24.67% of TRR), with further radioactivity incorporated into the sodium hydroxide reflux fraction representing cellulose incorporation (ranging between 0.45 and 3.96% TRR) and into the acid reflux fraction, representing lignin incorporation (0.49 to 2.69% TRR).

A1. Test Materials

Structure/Label	[Phenyl-U-^{14}C]-chlorothalonil	
	 (* = ^{14}C position)	
Common name	Chlorothalonil	
Syngenta code	-	
CAS Number	1897-45-6	
Batch number	Lot #218	Lot #220
Specific Activity	2.087 MBq/mg 56.4122 µCi/mg 125235 dpm/µg	5.162 MBq/mg 139.5261 µCi/mg 309748 dpm/µg
Radiochemical Purity	>99%	>99%

A2. Test System

The crops used were:

Carrot (*Daucus carota*), variety Mezzalunga Nantese 3.

Lettuce (*Lactuca sativa*), variety Kagraner Sommer 2.

Spring Wheat (*Triticum spp.)*, variety Pandas.

A3. Test Soil

Soil texture	Sandy loam
Soil composition	51.75% sand, 68.50% silt, 4.25% clay
pH	6.41
Organic carbon	0.90%
Cation exchange capacity	12.65 meq/100 g

A4. Test Facilities

The study was performed at Isagro Ricerca Srl, Environmental Chemistry Department, Metabolism and Environmental Fate, Unit 1, Via Fauser 4, Novara, Italy.

B. STUDY DESIGN AND METHODS

B1. Field Phase

22 pots were filled with sandy loam soil and treated with a 2g/L solution of a mixture of ^{14}C-labelled and non- labelled chlorothalonil in acetonitrile/water (30/70 v/v) at an equivalent rate of 7.5 kg a.s./ha by drop-wise addition to the soil. After aging of soil for 30, 120 and 365 days crops of carrot, lettuce and spring wheat were sown. Non-treated soils were included. The crops were grown outdoors in accordance with usual agricultural practice and irrigated as necessary.

Test Samples

Samples of crops at harvest maturity were taken for each plant-back interval. Carrots were separated into tops (leaves) and roots after excess soil was removed. The roots were washed gently with water and the washing returned to the growing pots. Lettuce samples were cut and rinsed gently, with the washing returned to the growing pots. Immature and mature wheat samples were cut close to the soil surface. Mature wheat was separated into straw and grain. Duplicate soil cores (5 cm diameter and 25 cm deep) were taken at treatment, at planting and at harvest for each plant-back interval.
All samples were stored frozen (approximately -20°C) for a maximum of 9 months until extraction and analysis.

B2. Analytical Phase

Homogenised samples were combusted and the total radioactive residue (TRR) was determined by liquid scintillation counting (LSC).
An aliquot of the homogenised plant material was extracted with acetone/water (50/50, v/v) using an Ultraturrax Homogenizer and concentrated to remove the acetone. The extracts were then partitioned with hexane or ethyl acetate. Selected aqueous phases were submitted to enzymatic (β-glucosidase) hydrolysis.
The remaining solids were further extracted with acetone by shaking, followed by centrifugation. After drying, the solids were refluxed successively with water, followed by 5% sodium hydroxide solution and then 6N hydrochloric acid to release any bound radioactivity.

The un-extracted residues (post-extraction solids, PES) were determined by combustion and LSC. Sample extracts were analysed by normal and reverse-phase TLC, using two solvent systems for each stationary phase, and metabolites identified by co-chromatography against reference standards. Soil samples were extracted by shaking with successive aliquots of acetone, acetone / water (50/50 v/v) then acetone/ 0.1N HCl (50/50 v/v) followed by filtration. The extracts were concentrated to remove the acetone and analysed by co-chromatography against reference standards. Un-extracted residues were determined by combustion and LSC.

II. RESULTS AND DISCUSSION

Distribution of residues

All residue values quoted in this section are expressed as mg chlorothalonil equivalents/kg.

The TRR and extractability of residues in following crops are summarised in Table 7.6.1.2-1 and Table 7.6.1.2-2.

The highest TRR were observed in 30 and 120 DAT mature lettuces (0.24 mg/kg), 120 DAT carrot leaves (1.5 mg/kg), carrot roots (0.43 mg/kg) and wheat straw (25 mg/kg) and 365 DAT wheat forage (1.4 mg/kg) and wheat grain (2.2 mg/kg). In general, TRR tended to remain at similar levels across all plant-back intervals.

The majority of the radioactive residues were extractable, accounting for 65 to 95% TRR. For all plant-back intervals the majority of the extractable residue was partitioned into the aqueous phase with the exception of wheat grain where the majority of extractable radioactivity was associated with the organic ethyl acetate phase (50%, 65% and 73% TRR associated with ethyl acetate for the 30, 120 and 365 day plant-back intervals, respectively).

The radioactivity in soil was quantified at treatment, at sowing and at harvest of each crop; total residues ranged from 0.68 to 6.9 mg/kg. TLC analysis of soil extracts showed that chlorothalonil was present only in the shorter soil ageing periods; levels decreased over time. Chlorothalonil was extensively metabolised in soil and at least eight known metabolites were identified.

Table 7.6.1.2-1: Summary of total radioactive residues by combustion in rotational crop samples grown in soil treated with [Phenyl-U-^{14}C]-chlorothalonil

Crop	Crop Commodity	mg/kg chlorothalonil equivalents Plant Back Interval		
		30 DAT	120 DAT	365 DAT
Lettuce	Mature	0.24	0.24	0.12
Carrot	Leaves	0.78	1.52	0.82
	Roots	0.28	0.43	0.18
Wheat	Forage	1.4	1.4	1.4
	Straw	23	25	24
	Grain	1.3	1.5	2.2

Table 7.6.1.2-2: Summary of total radioactive residues and extractability in rotational crop samples grown in soil treated with [Phenyl-U-^{14}C]-chlorothalonil

Crop	Days after Treatment	Crop Commodity	Extractable Radioactivity*		Non-extractable Radioactivity*		TRR
			%TRR	mg/kg	%TRR	mg/kg	mg/kg
Lettuce	30	Mature	73	0.18	22	0.05	0.24
	120	Mature	92	0.22	8.7	0.02	0.24
	365	Mature	85	0.10	16	0.02	0.12
Carrot	30	Root	90	0.28	8.9	0.03	0.28
		Leaves	75	0.59	21	0.17	0.78
	120	Root	91	0.43	11	0.05	0.43
		Leaves	65	0.99	33	0.50	1.5
	365	Root	78	0.18	23	0.04	0.18
		Leaves	72	0.59	28	0.23	0.82
Wheat	30	Forage	89	1.2	14	0.19	1.4
		Straw	86	20	12	2.7	23
		Grain	90	1.2	10	0.13	1.3
	120	Forage	88	1.2	14	0.20	1.4
		Straw	85	21	16	3.9	25
		Grain	90	1.5	9.3	0.14	1.5
	365	Forage	95	1.3	8.7	0.12	1.4
		Straw	95	23	4.6	1.1	24
		Grain	91	2.03	7.8	0.17	2.23

*percentages may differ as values in mg/kg were rounded off and the total of the measured extracts and residues is not always exactly equivalent to the TRR.

Characterisation of residues

Tables 7.6.1.2-3 – 7.6.1.2-8 summarise the results of the characterisation of residues and metabolite levels found.

Chlorothalonil was not detected in any of the plant samples (limit of detection 0.02 mg/kg).

In carrot roots the majority of the radioactive residue was assigned to the metabolites R611965 and R417888, accounting for 59%, 66% and 51% TRR for the 30, 120 and 365 plant-back intervals, respectively. These metabolites co-eluted on the TLC systems used, however further TLC analysis of the sample extract from the 30 day plant-back interval indicated that the majority of the residue was due to metabolite R611965 (51% TRR, 0.14 mg/kg). 24 - 31% TRR was assigned as conjugated material. Enzyme hydrolysis of this extract released radioactivity indicating that the conjugated material was made up of glucosyl conjugates of R611968, R613636 and “compound C15” (11%, 13% and 6.4% TRR, respectively, for the 30 day plant-back interval).

The metabolic profile for carrot leaves and lettuce was similar, with the majority of the radioactive residue assigned to metabolites R611965, R417888 and to conjugated material. Other metabolites identified at lower levels in carrot leaves and lettuce included R611553 (1.1 – 3.3% TRR), R182281

(0.41 – 0.98% TRR), R611968 (0.41-1.8% TRR), and R613636 (0.66 -5.4% TRR). These metabolites were identified for all plant-back intervals.

Table 7.6.1.2-3: Summary of identification and characterisation of residues in mature carrot roots grown in soil treated with [pheny-U-^{14}C]-chlorothalonil

	30 DAT plant-back		120 DAT plant-back		365 DAT plant-back	
TRR (mg/kg)	0.28		0.43		0.18	
Component	%TRR	mg/kg	%TRR	mg/kg	%TRR	mg/kg
Chlorothalonil	N/D	N/D	N/D	N/D	N/D	N/D
R611965 + R417888	59	0.16	66	0.29	51	0.09
R611968	N/D	N/D	N/D	N/D	N/D	N/D
Compound C15	N/D	N/D	N/D	N/D	N/D	N/D
R182281	0.71	0.002	0.93	0.004	0.56	0.001
R611553	N/D	N/D	N/D	N/D	N/D	N/D
Compound VIS02	N/D	N/D	N/D	N/D	1.69	0.003
R613636	N/D	N/D	N/D	N/D	N/D	N/D
Conjugates	31	0.09	24	0.10	25	0.04
Total identified	90	0.25	91	0.39	78	0.14
Non-extractable	8.9	0.03	11	0.05	23	0.04

The figures in the table are the total radioactive residue for each metabolite reported expressed as mg/kg chlorothalonil equivalents.
N/D Not detected

Table 7.6.1.2-4: Summary of identification and characterisation of residues in mature carrot leaves grown in soil treated with [^{14}C]-chlorothalonil

	30 DAT plant-back		120 DAT plant-back		365 DAT plant-back	
TRR (mg/kg)	0.78		1.5		0.82	
Component	%TRR	mg/kg	%TRR	mg/kg	%TRR	mg/kg
Chlorothalonil	N/D	N/D	N/D	N/D	N/D	N/D
R611965 + R417888	41	0.32	35	0.52	30	0.25
R611968	1.5	0.01	1.8	0.03	1.2	0.01
Compound C15	N/D	N/D	N/D	N/D	N/D	N/D
R182281	0.90	0.007	0.92	0.01	0.98	0.008
R611553	2.2	0.02	1.4	0.02	1.1	0.009
Compound VIS02	0.38	0.003	0.20	0.003	1.6	0.01
R613636	0.77	0.006	0.66	0.01	1.4	0.01
Conjugates	29	0.22	26	0.39	36	0.30
Total identified	75	0.59	65	0.99	72	0.59
Non-extractable	21	0.17	33	0.50	28	0.23

The figures in the table are the total radioactive residue for each metabolite reported expressed as mg/kg chlorothalonil equivalents.
N/D Not detected

Table 7.6.1.2-5: Summary of identification and characterisation of residues in mature lettuce grown in soil treated with [^{14}C]-chlorothalonil

	30 DAT plant-back		120 DAT plant-back		365 DAT plant-back	
TRR (mg/kg)	0.24		0.24		0.12	
Component	%TRR	mg/kg	%TRR	mg/kg	%TRR	mg/kg
Chlorothalonil	N/D	N/D	N/D	N/D	N/D	N/D
R611965 + R417888	34	0.08	44	0.11	39	0.05
R611968	0.41	0.001	1.2	0.003	1.7	0.002
Compound C15	0.83	0.002	0.83	0.002	0.83	0.001
R182281	0.41	0.001	1.2	0.003	--	--
R611553	3.3	0.008	3.3	0.008	2.5	0.003
Compound VIS02	2.5	0.006	1.7	0.004	6.6	0.008
R613636	4.2	0.010	5.4	0.01	5.0	0.006
Conjugates	25	0.06	32	0.08	30	0.04
Unknown[1]	3.7	0.009	2.9	0.007	--	--
Total identified	73	0.18	92	0.22	85	0.10
Non-extractable	22	0.05	8.7	0.02	16	0.02

The figures in the table are the total radioactive residue for each metabolite reported expressed as mg/kg chlorothalonil eq.
N/D Not detected.
[1] Unassigned radiocomponents which chromatographed away from the origin using 2D-TLC.

In wheat forage, straw and grain the metabolite profile was similar for all three crop parts over the three plant-back intervals. The majority of the radioactive residue was assigned to metabolites R611965 and R417888 with further TLC analysis of the sample extracts from the 30 day plant-back interval indicating that the majority of the residue was due to metabolite R611965 (76% TRR, 0.085 mg/kg for forage, 68% TRR, 0.87 mg/kg for grain and 26% TRR, 5.8 mg/kg for straw). Other identified metabolites represented minor percentages, including R182281 (1.2-1.5% TRR in forage, 0.84 – 7.7 % in straw and not found in grain) and R611968 (1.7-2.5% TRR in forage, 0.68-4.0 %TRR in straw and 4.3-9.8% TRR in grain). 11 - 41% TRR was assigned as conjugated material. Enzyme hydrolysis of this extract released radioactivity indicating that, as for carrot roots the conjugated material was made up of glucosyl conjugates of R611968, R613636 and “compound C15”

Table 7.6.1.2-6: Summary of identification and characterisation of residues in wheat forage grown in soil treated with [^{14}C]-chlorothalonil

	30 DAT plant-back		120 DAT plant-back		365 DAT plant-back	
TRR (mg/kg)	1.4		1.4		1.4	
Component	%TRR	mg/kg	%TRR	mg/kg	%TRR	mg/kg
Chlorothalonil	N/D	N/D	N/D	N/D	N/D	N/D
R611965 + R417888	61	0.84	62	0.87	67	0.94
R611968	2.2	0.03	1.7	0.02	2.5	0.04
Compound C15	1.2	0.02	0.85	0.01	1.2	0.02
R182281	1.4	0.02	1.2	0.02	1.5	0.02
R611553	N/D	N/D	N/D	N/D	N/D	N/D
Compound VIS02	N/D	N/D	N/D	N/D	N/D	N/D
R613636	N/D	N/D	N/D	N/D	N/D	N/D
Conjugates	23	0.31	22	0.31	23	0.33
Total identified	89	1.2	88	1.2	95	1.3
Non-extractable	14	0.19	14	0.20	8.	0.12

The figures in the table are the total radioactive residue for each metabolite reported expressed as mg/kg chlorothalonil eq.
N/D Not detected

Table 7.6.1.2-7: Summary of identification and characterisation of residues in wheat straw grown in soil treated with [^{14}C]-chlorothalonil

	30 DAT plant-back		120 DAT plant-back		365 DAT plant-back	
TRR (mg/kg)	23		25		24	
Component	%TRR	mg/kg	%TRR	mg/kg	%TRR	mg/kg
Chlorothalonil	N/D	N/D	N/D	N/D	N/D	N/D
R611965 + R417888	49	11	50	12	49	12
R611968	0.68	0.16	1.1	0.28	4.0	0.96
Compound C15	0.80	0.18	0.79	0.20	--	--
R182281	7.7	1.8	4.7	1.2	0.84	0.20
R611553	N/D	N/D	1.1	0.28	0.94	0.22
Compound VIS02	N/D	N/D	N/D	N/D	0.42	0.10
R613636	0.93	0.21	1.5	0.37	N/D	N/D
Conjugates	27	6.2	26	6.6	40	9.5
Total identified	86	20	85	21	95	23
Non-extractable	12	2.7	16	3.9	4.6	1.1

The figures in the table are the total radioactive residue for each metabolite reported expressed as mg/kg chlorothalonil equivalents.
N/D Not detected

Table 7.6.1.2-8: Summary of Identification and Characterisation of Residues in Wheat Grain Grown in Soil Treated with [^{14}C]-chlorothalonil

	30 DAT plant-back		120 DAT plant-back		365 DAT plant-back	
TRR (mg/kg)	1.3		1.5		2.2	
Component	%TRR	mg/kg	%TRR	mg/kg	%TRR	mg/kg
Chlorothalonil	N/D	N/D	N/D	N/D	N/D	N/D
R611965 + R417888	71	0.91	73	1.1	40	0.89
R611968	4.3	0.06	6.0	0.09	9.8	0.22
Compound C15	2.9	0.04	N/D	N/D	N/D	N/D
R182281	N/D	N/D	N/D	N/D	N/D	N/D
R611553	N/D	N/D	N/D	N/D	N/D	N/D
Compound VIS02	N/D	N/D	N/D	N/D	N/D	N/D
R613636	N/D	N/D	N/D	N/D	N/D	N/D
Conjugates	12	0.15	11	0.17	41	0.92
Total identified	90	1.2	90	1.4	91	2.0
Non-extractable	10	0.13	9.3	0.14	7.8	0.17

The figures in the table are the total radioactive residue for each metabolite reported expressed as mg/kg chlorothalonil equivalents.
N/D Not detected

The post-extraction solids (PES) residues ranged from 0.03 to 0.05 mg/kg (8.9 - 23% TRR) in carrot root, 0.17 to 0.50 mg/kg (21 – 33% TRR) in carrot leaves and 0.02 to 0.05 mg/kg (8.7 – 22% TRR) in lettuce. Post extraction solids for cereal samples ranged from 0.12 – 0.2 mg/kg (8.7-14.1% TRR), 0.13 – 0.17 mg/kg (7.8 – 10% TRR) and 1.1 – 3.9 mg/kg (4.6 – 16% TRR) for grain and straw, respectively. Considerable amounts of this radioactivity were released by aqueous reflux (ranging between 3.6 and 25% of TRR); with further radioactivity incorporated into the sodium hydroxide reflux fraction representing cellulose incorporation (ranging between 0.45 and 4.0% TRR) and into the acid reflux fraction, representing lignin incorporation (0.49 to 2.7% TRR).

Proposed metabolic pathway for chlorothalonil in following crops

The proposed metabolic pathway for chlorothalonil in following crops is given in Figure 7.6.1.2-1.

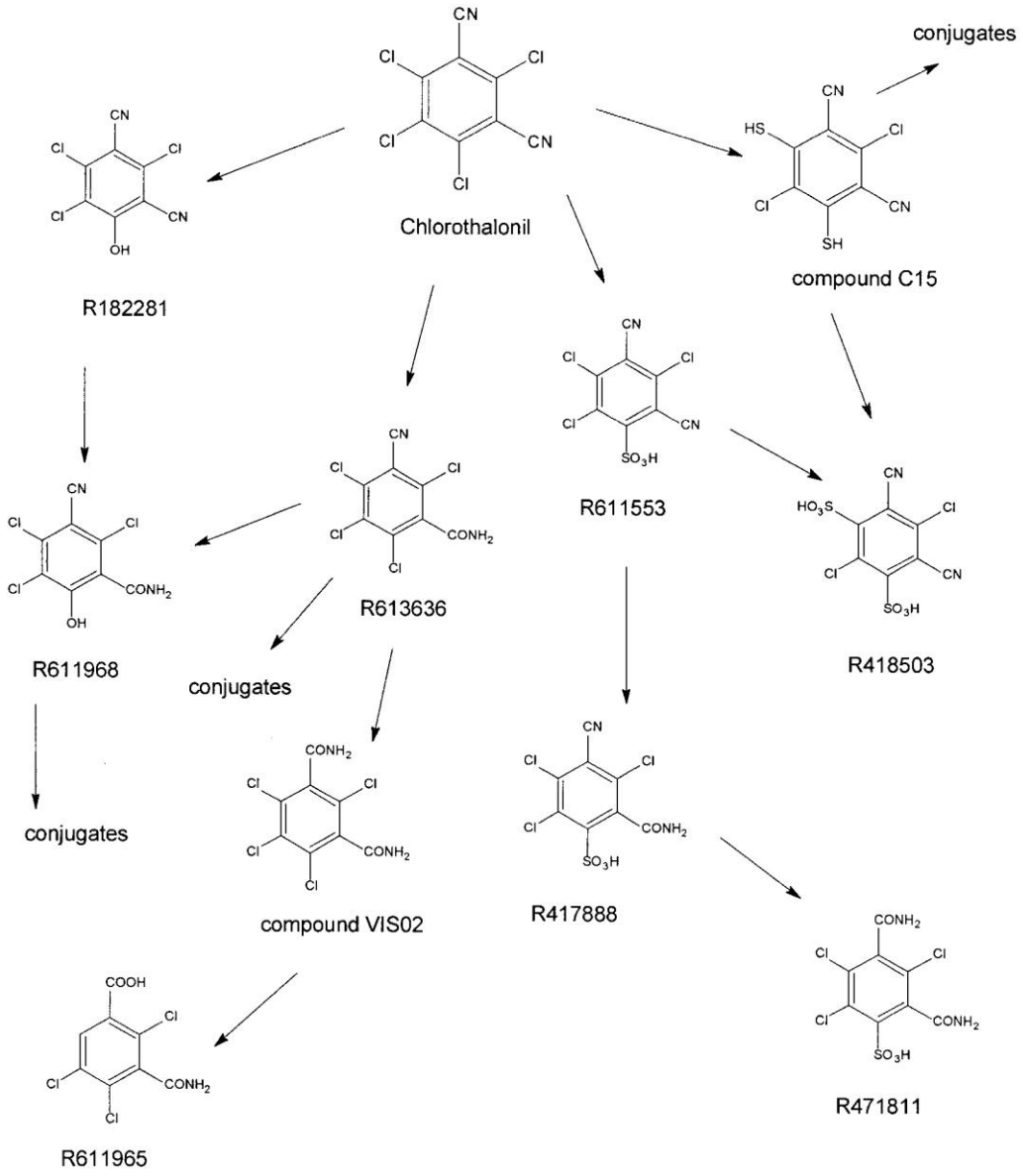
III. CONCLUSIONS

Following application of [phenyl-U-^{14}C]-chlorothalonil to bare soil at 7.5 kg a.s/ha, lettuce, carrot, and wheat were sown in the treated soil after periods of 30, 120 and 365 days. Samples of mature lettuce, carrot root and leaves and wheat forage, straw and grain were taken after each ageing period.
The highest total radioactive residues (TRR, mg/kg chlorothalonil equivalents), were observed in 30 and 120 DAT mature lettuces (0.24 mg/kg), 120 DAT carrot leaves (1.5 mg/kg), carrot roots (0.43 mg/kg) and wheat straw (25 mg/kg), and 365 DAT wheat forage (1.4 mg/kg) and wheat grain (2.2 mg/kg). The majority of the radioactive residues were extractable, accounting for 65 to 95% TRR.
The results show that:

- In general, TRR remained at similar levels across all plant-back intervals.
- Levels of residues in crops grown in soil treated with chlorothalonil were ≥0.12 mg/kg for all plant-back intervals.
- Parent chlorothalonil is a minor residue (not detected) in rotational crops.
- R611965 and R417888 were significant metabolites in all crops. These metabolites are known soil metabolites with long DT_{50} values.
- Levels of R182281 were low (<5%TRR) in carrots, lettuce and wheat forage, and were not detected in wheat grain. Levels in wheat straw decreased from a maximum of 7% TRR with longer plant-back intervals.

Chlorothalonil is metabolised in soil initially to R613636 and R182281 and then to other multiple components which are available for uptake by crops. The metabolism in following crops is similar to that in primary crops.

Figure 7.6.1.2-1: Proposed metabolic pathway for chlorothalonil in following crops



B.7.6.1.3 Metabolism in rotational crops, study 3

Previous evaluation	Submitted for the purpose of renewal
RMS remark	Acceptable

Report:	K-CA 6.6.1/02. Mamouni A. (2009), ^{14}C-Chlorothalonil – Confined accumulation in rotational crops, Harlan Laboratories Ltd. Switzerland, Report number B34931. Syngenta File No. R044686_11194.

Guidelines

The study was performed according to the EU Commission Working Document 7524/VI/95 rev. 2: Appendix C - Testing of Plant Protection Products in Rotational Crops and under consideration of OPPTS Guideline 860.1850

GLP

The study was carried out according to the principles of Good Laboratory Practice.

EXECUTIVE SUMMARY

[Phenyl-U-^{14}C]-chlorothalonil formulated as a SC was applied as a single spray application at 1 kg a.s./ha to separate containers of soil. The soil was aged for 30 days and representative crops of cereals (barley), leafy vegetable (spinach) and root vegetable (radish) were sown into the soil and grown under field conditions. Samples were taken soon after emergence and at immature and mature growth stages. Harvested crops were separated into commodities of representative food and feed items (barley forage, straw and grain; spinach leaves; radish leaves and roots). Samples were homogenised and the total radioactive residue concentration (TRR, mg chlorothalonil equivalents per kg of commodity, mg/kg) was measured in each commodity. Commodities were extracted with acetonitrile: water and the extracts analysed by HPLC and TLC to determine the nature of the residues.

In spinach leaves, the total radioactive residues (TRR) were 0.067, 0.031 and 0.039 mg/kg for the immature (emergence), immature (pre-harvest) and mature crop samples, respectively. The corresponding values for radish leaves were 0.014, 0.019 and 0.026 mg/kg. For radish roots, the TRR values were 0.022, 0.019 and 0.021 mg/kg. In barley forage, the TRR for immature (emergence) and forage (immature plant) samples were 0.018 and 0.019 mg/kg, respectively. The barley harvested at maturity was separated into three parts straw, chaff and grains. The residues in these samples were 0.120, 0.059 and 0.012 mg/kg respectively.

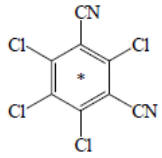
The majority of the radioactive residues were extractable, accounting for 50 to 73% TRR. Due to the very low level of residues in the plant parts remaining after extraction, no additional hydrolysis of the non-extractable residues was performed.

Parent chlorothalonil was detected only in the radish root samples, and at very low levels (0.001 mg/kg).

The major identified metabolite was R611965, which represented 13% TRR (0.009 mg/kg), 19% TRR (0.006 mg/kg) and 14% TRR (0.006 mg/kg) in immature (emergence), immature (pre-harvest) and mature spinach respectively. R611965 was detected in all barley samples with the highest level found in mature straw, representing 25% TRR (0.030 mg/kg). In the radish root R611965 accounted for 11.5% TRR (0.003 mg/kg) and 8.3% TRR (0.002 mg/kg) in the immature (emergence) roots and at maturity, respectively. The metabolite R182281 was also identified in all spinach and radish root samples, and the mature barley samples, however at levels less than 0.01 mg/kg.

The remaining radioactive fractions corresponded mainly to conjugates and did not exceed 10% TRR, 0.01 mg/kg for edible plant material (human food) or 0.05 mg/kg for animal feed items.

A1 Test Materials

Structure/Label	[Phenyl-U-^{14}C]-Chlorothalonil
	 (* = ^{14}C position)
CAS Number	1897-45-6
Batch Number	6BLY041 and 8BLY035
Specific Activity	8.49 MBq/mg and 6.96 MBq/mg
Radiochemical Purity	>98 %

A2. Test System

The crops selected to represent the three different groupings (leafy, cereal and root) were spinach (variety Butterblatt), radish (variety Cherry Belle) and barley (variety Mandolin/CEBADA), respectively. These crops and were grown from commercially available seed, outdoors under conditions representative of those used for commercial production.

A3. Test Soil

Soil texture	Silty clay
Soil composition	10% sand, 44.7% silt, 45.3% clay
pH	7.33
Organic carbon	3.15%
Cation exchange capacity	13.3 meq/100 g

B. STUDY DESIGN AND METHODS

B1. Field Phase

Nominal application rate	1 kg a.s/ ha
Number of applications	1
Target seasonal application rate	1 kg a.s./ ha
Achieved seasonal application rate	99.5%
Formulation type	Suspension concentrate (SC)
Formulation code	209583/A
Spray rate	12 mL/plant pot (equivalent to51 mL/1m^2)
Method of application	manual sprayer
Plant-back intervals	30 days after application

Test Samples

The following crop samples were taken:

Emergence spinach leaves, 20 days after sowing
Pre-harvest spinach leaves, 29 days after sowing
Mature spinach leaves, 40 days after sowing
Emergence radish leaves and roots, 20 days after sowing
Pre-harvest radish leaves and roots, 29 days after sowing
Mature radish leaves and roots, 40 days after sowing
Emergence barley forage, 20 days after sowing
Pre-harvest barley forage, 29 days after sowing
Mature barley, straw, chaff and grain, 111 days after sowing

Sample Preparation

Plant tissue and soil samples were frozen and then milled to form a powder. Total radioactivity was determined by combustion followed by LSC.

B2. Analytical Phase

The following procedure was used for the extraction of all samples except radish leaves which were not extracted. Samples were extracted up to 3 times with acetonitrile: water (1:1, v: v; adjusted to pH 3 for soil) on a shaker for 30 minutes. The samples were centrifuged to remove the solids after each extraction. Extracts were pooled and concentrated for analysis. Soil was additionally extracted with the same solvent acidified further to pH 1 under reflux conditions for 4 hours. The PES were allowed to dry, and then analysed by combustion LSC.

Identification of the radioactive components in the sample extracts was carried out by 2 dimensional TLC with phosphor imaging and HPLC-UV using co-chromatography with reference standards.

II. RESULTS AND DISCUSSION

Total Radioactive Residues and Extractability

Total radioactive residues are summarised in Table 7.6.1.3-1 and the extractability for all commodities are summarised in Table 7.6.1.3-2.

Significant TRR were observed in all samples with the lowest TRR in mature grain (0.012 mg/kg) and the highest TRR in mature straw samples (0.120 mg/kg). The majority of the radioactive residues were extractable, accounting for 50 to 73% TRR.

Chlorothalonil was detected only in the radish root samples, and at very low levels (0.001 mg/kg).

The major identified metabolite in all crops was R611965 with residues ranging from 0.002 mg/kg (8.3% TRR, mature radish roots) to 0.030 mg/kg (25% TRR, mature straw samples). R182281 was identified in spinach, radish root and mature barley samples, however at levels less than 0.01 mg/kg.

Table 7.6.1.3-1: Summary of total radioactive residues by combustion in rotational crop samples grown in soil treated with [phenyl-U-^{14}C]-chlorothalonil and aged for 30 days

Crop	Days after treatment	Crop Commodity	mg/kg Chlorothalonil Equivalents
Spinach	50	Immature leaves (emergence)	0.067
	59	Immature leaves (pre-harvest)	0.031
	70	Mature leaves	0.039
Radish	50	Immature roots (emergence)	0.022
	59	Immature roots (pre-harvest)	0.019
	70	Mature roots	0.021
	50	Immature leaves (emergence)	0.014
	59	Immature leaves (pre-harvest)	0.019
	70	Mature leaves	0.026
Wheat	50	Forage (emergence)	0.018
	59	Immature plant forage (pre-harvest)	0.019
	141	Mature straw	0.120
	141	Mature chaff	0.059
	141	Mature grain	0.012

Table 7.6.1.3-2: Summary of total radioactive residues and extractability in rotational crop samples grown in soil treated with [phenyl-U-^{14}C]-chlorothalonil and aged for 30 days

Crop	Days after treatment	Crop Commodity	Extractable Radioactivity		Non-extractable Radioactivity		TRR
			%TRR	mg/kg	%TRR	mg/kg	mg/kg
Spinach	50	Immature leaves (emergence)	66.4	0.045	33.6	0.023	0.067
	59	Immature leaves (pre-harvest)	69.4	0.021	30.6	0.009	0.031
	70	Mature leaves	65.2	0.027	34.8	0.014	0.041
Radish	50	Immature roots (emergence)	49.6	0.011	50.4	0.011	0.022
	59	Immature roots (pre-harvest)	63.5	0.012	36.5	0.007	0.019
	70	Mature roots	50.6	0.011	49.4	0.011	0.022
Wheat	50	Forage (emergence)	56.0	0.010	44.0	0.008	0.018
	59	Immature plant forage (pre-harvest)	52.9	0.010	47.1	0.009	0.019
	141	Mature straw	67.4	0.081	32.6	0.039	0.120
	141	Mature chaff	73.4	0.043	26.6	0.016	0.059
	141	Mature grain	50.3	0.006	49.7	0.006	0.012

Characterisation and Identification of Residues

The extracts were analysed as summarised in the previous section. The identified components for each commodity are summarised in Tables 7.6.1.3-3 to 7.6.1.3-5.

Table 7.6.1.3-3: Summary of identification and characterisation of residues in spinach grown in soil treated with [phenyl-U-^{14}C]-chlorothalonil and aged for 30 days

	Immature leaves (emergence)		Immature leaves (pre-harvest)		Mature leaves	
TRR mg/kg	0.067		0.031		0.041	
Component	%TRR	mg/kg	%TRR	mg/kg	%TRR	mg/kg
Chlorothalonil	N/D	N/D	N/D	N/D	N/D	N/D
R611965	13.3	0.009	18.7	0.006	13.5	0.006
R182281	13.9	0.009	12.1	0.004	11.4	0.005
M3	6.2	0.004	2.7	0.001	1.6	0.001
M4	N/D	N/D	N/D	N/D	1.7	0.001
M5	N/D	N/D	N/D	N/D	3.6	0.001
M6	N/D	N/D	N/D	N/D	1.8	0.001
M7	N/D	N/D	N/D	N/D	1.7	0.001
M8	N/D	N/D	N/D	N/D	1.3	0.001
M9	N/D	N/D	N/D	N/D	1.7	0.001
M10	N/D	N/D	N/D	N/D	4.0	0.002
M11	N/D	N/D	N/D	N/D	3.3	0.001
M12	9.3	0.006	10.0	0.003	1.9	0.001
M13	N/D	N/D	9.0	0.003	4.5	0.002
M14	8.0	0.005	6.0	0.002	1.9	0.001
M15	6.4	0.004	3.7	0.001	4.2	0.002
M16	9.3	0.006	7.1	0.002	7.1	0.003
Total identified	66.4	0.044	69.3	0.021	65.2	0.025
Un-extracted	33.6	0.023	30.6	0.009	34.8	0.014

N/D not detected

Table 7.6.1.3-4: Summary of identification and characterisation of residues in radish roots grown in soil treated with [phenyl-U-^{14}C]-chlorothalonil and aged for 30 days

	Immature roots (emergence)		Immature roots (pre-harvest)		Mature roots	
TRR mg/kg	0.022		0.019		0.021	
Component	%TRR	mg/kg	%TRR	mg/kg	%TRR	mg/kg
Chlorothalonil	4.1	0.001	N/R	N/R	3.4	0.001
R611965	11.5	0.003			8.3	0.002
R182281	22.0	0.005			36.0	0.008
M3	N/D	N/D			N/D	N/D
M10	4.6	0.001			2.0	<0.001
M11	5.0	0.001			0.8	<0.001
M12	2.3	0.001			N/D	N/D
Total identified	49.5	0.011			50.5	0.011
Un-extracted	50.4	0.011			49.4	0.010

N/D not detected

N/R Could not be separated

Table 7.6.1.3-5: Summary of identification and characterisation of residues in cereals grown in soil treated with [phenyl-U-^{14}C]-chlorothalonil and aged for 30 days

	Forage (emergence)		Immature plant forage (pre-harvest)		Mature straw		Mature chaff		Mature grain	
TRR mg/kg	0.018		0.019		0.120		0.059		0.012	
Component	%TRR	mg/kg	%TRR	mg/kg	%TRR	mg/kg	%TRR	mg/kg	%TRR	mg/kg
Chlorothalonil	N/D	N/D	N/D	N/D	N/D	N/D	N/D	N/D	N/D	N/D
R611965	56.0	0.010	52.9	0.010	24.8	0.03	22.2	0.013	31.8	0.004
R182281	N/D	N/D	N/D	N/D	5.5	0.007	2.3	0.001	18.5	0.002
M3	N/D	N/D	N/D	N/D	N/D	N/D	2.1	0.001	N/D	N/D
M8	N/D	N/D	N/D	N/D	N/D	N/D	1.7	0.001	N/D	N/D
M9	N/D	N/D	N/D	N/D	7.0	0.008	3.2	0.002	N/D	N/D
M10	N/D	N/D	N/D	N/D	7.3	0.009	5.9	0.003	N/D	N/D
M11	N/D	N/D	N/D	N/D	5.5	0.007	8.4	0.005	N/D	N/D
M12	N/D	N/D	N/D	N/D	5.2	0.006	2.1	0.001	N/D	N/D
M13	N/D	N/D	N/D	N/D	N/D	N/D	5.5	0.003	N/D	N/D
M14	N/D	N/D	N/D	N/D	N/D	N/D	1.3	0.001	N/D	N/D
M15	N/D	N/D	N/D	N/D	6.5	0.008	9.4	0.006	N/D	N/D
M16	N/D	N/D	N/D	N/D	5.5	0.007	5.0	0.003	N/D	N/D
M17	N/D	N/D	N/D	N/D	N/D	N/D	4.4	0.003	N/D	N/D
Total identified	56.0	0.01	52.9	0.010	67.3	0.081	73.2	0.043	50.3	0.006
Un-extracted	44.0	0.008	47.1	0.009	32.6	0.039	26.6	0.016	49.7	0.006

N/D not detected

III. CONCLUSIONS

In spinach leaves, the total radioactive residues (TRR) were 0.067, 0.031 and 0.039 mg/kg for the immature (emergence), immature (pre-harvest) and mature crop samples, respectively. The corresponding values for radish leaves were 0.014, 0.019 and 0.026 mg/kg. For radish roots, the TRR values were 0.022, 0.019 and 0.021 mg/kg. In barley forage, the TRR for immature (emergence) and forage (immature plant) samples were 0.018 and 0.019 mg/kg, respectively. The barley harvested at maturity was separated into three parts straw, chaff and grains. The residues in these samples were 0.120, 0.059 and 0.012 mg/kg respectively.

The majority of the radioactive residues were extractable, accounting for 50 to 73% TRR. Due to the very low level of residues in the plant parts remaining after extraction, no additional hydrolysis of the non-extractable residues was performed.

Parent chlorothalonil was detected only in the radish root samples, and at very low levels (0.001 mg/kg).

The major identified metabolite was R611965, which represented 13% TRR (0.009 mg/kg), 19% TRR (0.006 mg/kg) and 14% TRR (0.006 mg/kg) in immature (emergence), immature (pre-harvest) and mature spinach respectively. R611965 was detected in all barley samples with the highest level found in mature straw, representing 25% TRR (0.030 mg/kg). In the radish root R611965 accounted for 11.5% TRR (0.003 mg/kg) and 8.3% TRR (0.002 mg/kg) in the immature (emergence) roots and at maturity, respectively. The metabolite R182281 was also identified in all spinach and radish root samples, and the mature barley samples, however at levels less than 0.01 mg/kg.

The remaining radioactive fractions corresponded mainly to conjugates and did not exceed 10% TRR, 0.01 mg/kg for edible plant material (human food) or 0.05 mg/kg for animal feed items.

B.7.6.2 Magnitude of residues in rotational crops

B.7.6.2.1 Magnitude of residues in rotational crops, study 1

Previous evaluation	In DAR
RMS remark	Acceptable, however, studies were performed in the USA. No storage time of the samples has been reported.

Characteristics

reference	:	Zeneca, Dillon et al., 1984.	treatment	:	field study
type of study	:	rotational crop study	rate	:	8 applications of 2.5 kg a.i/ha
year of execution	:	1981	formulation	:	Bravo 500 (40.4% chlorothalonil)
test substance	:	chlorothalonil	GLP statement	:	no
crop/commodity	:	wheat, carrots, snapbeans and spinach	guidelines	:	not applicable

Study design

Plots in Georgia, Texas and California were sprayed with Bravo 500, 8 applications at a rate of 2.5 kg a.i/ha at 7 day intervals. The water rate ranged from 190 to 380 l/ha. Wheat, carrots, snapbeans and spinach were planted 14, 30, 60 and 90 days and about 1 year after the last application. The crops were harvested at harvest-ripeness and crop samples were processed, extracted and partitioned into organic solvent. Soil samples were taken 0, 14, 30, 60, 90, 120 and 150 days and at 1 year after the last application, at depths of 0-8, 8-15, 15-23 and 23-30 cm. Additional samples were taken at each wheat harvest, after the 4th application in the Georgia vegetable plot after selected vegetable harvests. Chlorothalonil and its metabolites were extracted from soil, partitioned into organic solvent and assayed.

Results

a. Georgia

Most of the soil residues of chlorothalonil were found in the top soil segment (0-8 cm). The SDS-2787 (parent compound) residues ranged from 5.48 mg/kg on the day of the last application to 0.10 mg/kg 372 days after the last application. Chlorothalonil degraded in soil to produce the soil metabolites SDS-3701, SDS-19221, SDS-46851, SDS-47523/4 and SDS-47525. Most of the metabolite residues remained in the top 8 cm of soil.

The major identified metabolite in soil was SDS-3701; 0.22 mg/kg, 0.78 mg/kg and 0.25 mg/kg in the top layer at pre-sampling intervals (PSI) of 0, 30 and 372 days respectively. The metabolites SDS-19221 and SDS-47523/SDS-47524 were present in the first 8 cm of soil at levels of 0.16 mg/kg at a PSI of 0 days, 0.24 mg/kg at a PSI of 30 days and approximately 0.025 mg/kg at a PSI of 372 days.

The maximum residue level of the metabolite SDS-46851 in the top layer of soil, 0.15 mg/kg, was reached at a PSI of 30 days. The SDS-46851 residue levels in this layer ranged from 0.03 to 0.07 mg/kg at the other PSI's. SDS-47525 residues remained at or below 0.05 mg/kg in the top layer.

No residues of the parent compound were detected in the rotational crops. A summary of the residues of the metabolites SDS-3701 and SDS-46851, resulting in crops grown on chlorothalonil-treated soil, are given in Table 7.9.2.1.

Table 7.9.2.1 Mean residue levels (in mg/kg) of the metabolites SDS-3701 and SDS-46851 in crops.

Rotation interval (days)	Spinach	Snap Beans	Carrots		Wheat	
			Roots	Tops	Grain	Straw
SDS-3701						
14	0.02	<0.01	<0.01	<0.01	<0.01	0.01
30	0.05	<0.01	0.02	<0.01	<0.01	<0.01
60	0.05	<0.01	0.03	0.02	<0.01	0.04
90	0.19	<0.01	0.02	0.02	<0.01	0.02
372	<0.01	-[1]	<0.01	0.03	<0.01	<0.01
SDS-46851						
14	2.15	0.19	0.13	0.23	0.17	3.15
30	1.05	0.15	0.10	0.26	0.23	5.03
60	1.82	0.22	0.38	0.36	0.68	10.35
90	2.20	1.00	0.59	0.65	0.58	6.74
372	<0.05	-[1]	<0.01	<0.01	0.01	<0.05

1 no sample received

Conclusions

At all pre-sampling intervals, the major residue component in soil was the parent compound, followed by the metabolite SDS-3701. SDS-46851 was the major residue compound identified in rotational crop samples. SDS-3701 was present at lower levels. The parent compound was not detected in crop samples.

b.Texas

Most of the soil residues of chlorothalonil were found in the top soil segment (0-8 cm). The SDS-2787 (parent compound) residues ranged from 5.18 mg/kg on the day of the last application to <0.01 mg/kg

257 days after the last application. Chlorothalonil degraded in soil to produce the soil metabolites SDS-3701, SDS-19221, SDS-46851, SDS-47523/4 and SDS-47525. Most of the metabolite residues remained in the top 8 cm of soil, but SDS-3701, SDS-46851 and SDS-47525 were also detected in lower layers of soil.

The major identified metabolite in soil was SDS-3701; 1.03 mg/kg, 2.46 mg/kg and 0.80 mg/kg in the top layer at pre-sampling intervals (PSI) of 0, 30 and 257 days respectively. The metabolite SDS-19221 was present at levels of 0.22 mg/kg, 0.40 mg/kg and 0.03 mg/kg in the top layer at PSI's of 0, 30 and 257 days respectively. The SDS-47523/SDS-47524 residue levels reached a maximum of 0.08 mg/kg in the top soil layer at a 14 day pre-sampling period and the SDS-47525 residue levels reached a maximum of 0.06 mg/kg at a 30 day pre-sampling period. The maximum residue level of the metabolite SDS-46851 in the top layer of soil, 0.10 mg/kg, was reached at a PSI of 10 days. The SDS-46851 residue levels in this layer ranged from <0.01 to 0.08 mg/kg at the other PSI's.

No parent compound was detected in the rotational crops. A summary of the levels of the metabolites SDS-3701 and SDS-4685, resulting in crops grown on chlorothalonil-treated soil, are given in Table 7.9.2.2.

Table 7.9.2.2 Mean residue levels (in mg/kg) of the metabolites SDS-3701 and SDS-46851 in crops.

Rotation interval (days)	Spinach	Snap Beans	Carrots		Wheat	
			Roots	Tops	Grain	Straw
SDS-3701						
14	0.01	<0.01	0.01	0.02	-[2]	-[2]
30	0.02	<0.01	0.02	0.04	-[2]	-[2]
60	0.04	<0.01	0.03	0.02	-[2]	-[2]
90	0.03	-[1]	-[1]	-[1]	-[2]	-[2]
372	<0.01	-[1]	<0.01	<0.01	<0.01	0.02
SDS-46851						
14	<0.05	0.03	0.02	0.03	-[2]	-[2]
30	<0.05	0.15	0.02	0.04	-[2]	-[2]
60	<0.05	0.74	0.02	0.02	-[2]	-[2]
90	<0.05	-[1]	-[1]	-[1]	-[2]	-[2]
372	<0.05	-[1]	<0.01	<0.01	<0.01	<0.05

1 no sample received

2 wheat too immature to analyse

Conclusions

On the day of the last application, the major soil residue compound was the parent compound, followed by the metabolite SDS-3701. At all other PSI's, the metabolite SDS-3701 prevailed in the top soil layer. SDS-3701 was the major residue compound identified in spinach and wheat straw samples and SDS-46851 was the major residue compound in snap beans, carrot roots and and carrot tops. No residues were detected in wheat grain (only the one-year samples were mature enough to analyse). The parent compound was not detected in crop samples.

c.California

Most of the soil residues of chlorothalonil were found in the top soil segment (0-8 cm). The SDS-2787 (parent compound) residues amounted 17.46 mg/kg on the day of the last application, 45.17 mg/kg at a 14 days pre-sampling period and 0.04 mg/kg at a PSI of 417 days. Chlorothalonil degraded in soil to produce the soil metabolites SDS-3701, SDS-19221, SDS-46851, SDS-47523/4 and SDS-47525. Most of the residues remained in the top 8 cm of soil.

The major identified metabolite in soil was SDS-3701 and levels ranged from 1.83 mg/kg on the day of the last application to 0.80 mg/kg 417 days pre-sampling, in the top layer of soil. The maximum residue level of the metabolite SDS-19221 in the top layer of soil, 0.27 mg/kg, was reached at a PSI of 150 days. The SDS-19221 residue levels in this layer ranged from 0.02 to 0.15 mg/kg at the remaining PSI's. SDS-46851, SDS-47523/4 and SDS-47525 residues remained at or below 0.04 mg/kg, 0.08 mg/kg and 0.06 mg/kg respectively in the top layer.

No parent compound was detected in the rotational crops. A summary of the residues of the metabolites SDS-3701 and SDS-4685, resulting in crops grown on chlorothalonil-treated soil, are given in Table 7.9.2.3.

Table 7.9.2.3 Mean residue levels (in mg/kg) of the metabolites SDS-3701 and SDS-46851 in crops.

Rotation interval (days)	Spinach	Snap Beans	Carrots		Wheat	
			Roots	Tops	Grain	Straw
SDS-3701						
14	0.03	-[1]	0.02	0.02	<0.01	0.08
30	0.01	<0.01	0.01	0.02	<0.01	0.03
60	0.05	<0.01	0.03	0.03	-[3]	-[3]
90	0.02	<0.01	0.02	0.04	-[3]	-[3]
372	<0.01	-[2]	<0.01	-[1]	<0.01	0.01
SDS-46851						

Rotation interval (days)	Spinach	Snap Beans	Carrots		Wheat	
			Roots	Tops	Grain	Straw
14	<0.05	-[1]	<0.01	0.03	<0.01	0.07
30	<0.05	0.02	<0.01	0.03	0.03	0.18
60	<0.05	0.02	0.02	0.02	-[3]	-[3]
90	<0.05	0.03	0.01	0.02	-[3]	-[3]
372	<0.05	-[2]	0.02	-[1]	0.06	0.37

1 insufficient sample for analysis

2 no sample received

3 wheat too immature to analyse

Conclusions

At all pre-sampling intervals, the major soil residue compound was the parent compound, followed by the metabolite SDS-3701. SDS-46851 was the major residue compound identified in wheat grain and straw and in snap beans. SDS-3701 was the major residue compound in spinach. Comparable amounts of both metabolites were detected in carrot roots and tops. The parent compound was not detected in crop samples.

Limitations

The studies did not reveal any significant limitations. However, it should be noted that the studies were all performed in the USA. The conditions under which growth and cultivation take place including weather can significantly influence the residual behaviour of a plant protection product. The studies should therefore be representative of the areas where Community authorization is granted or envisaged. In this case, the extensive data from several climatic zones in the USA seem to provide an adequate basis on which to evaluate the residue situation in the Member States of the EC.

B.7.6.2.2 Magnitude of residues in rotational crops, study 2

Previous evaluation	In DAR
RMS remark	Acceptable, however, studies were performed in the USA. No storage time of the samples has been reported.

Characteristics

reference	:	Zeneca, Rose et al., 1991	treatment	:	field study
type of study	:	rotational crop study	rate	:	range 1.3-2.6 kg a.i/ha (see table x)
year of execution	:	1985	formulation	:	Bravo 500 (40.4% chlorothalonil) and Bravo 720 (54.0 % chlorothalonil)
test substance	:	chlorothalonil	GLP statement	:	yes
crop/commodity	:	a whole scala of crops (see Table 7.9.3.1)	guidelines	:	not applicable

Study design

Test sites were treated with Bravo 500/720 at maximum rates for the primary crop and at the most intensive schedule of applications allowed. Data concerning the treatment of the various primary crops are summarized in Table 7.9.3.1.

Table 7.9.3.1 Data concerning the treatment of the primary crops

Location	Primary crop	Formulation	Rate (kg a.i./ha)	No. of applications
Georgia (Donalsonville)	Peanuts	Bravo 720	1.3	10
Texas	Cucumbers	Bravo 720	2.6	8
Oklahoma	Peanuts	Bravo 720	1.3	8
California (Fresno)	Tomatoes	Bravo 500	2.4	8
North Dakota	Potatoes	Bravo 500	1.2	8
California (Greenfield)	Broccoli	Bravo 500	1.3	9
Idaho	Potatoes	Bravo 720	1.3	8
Georgia (Parrot)	Peanuts	Bravo 500	1.3	11
Georgia (Plains)	Peanuts	Bravo 500	1.3	6
New York	Potatoes	Bravo 500	1.3	12
Louisiana	Soybeans	Bravo 720	1.7	3
Maryland	Tomatoes	Bravo 720	2.6	8

Following treatment, the primary crops were harvested at normal maturity, and rotational crops were planted at varying intervals following harvest of the primary crops. The rotated crops were harvested at maturity. Soil samples were taken at several depths at pre-treatment, at last application, and at or near the planting and harvest of rotational crops.

Results

Most of the soil residues of chlorothalonil were found in the top soil segment (0-8 cm). The determined chlorothalonil residue levels in the top soil are presented in Table 7.9.3.2.

Table 7.9.3.2 Chlorothalonil residue ranges in the top soil segment

Location	PTI	Residues (mg/kg)			PTI	Residues (mg/kg)		
		SDS-2787	SDS-3701	SDS-46851		SDS-2787	SDS-3701	SDS-46851
Georgia (Donalsonville)	0	0.21	0.13	0.04	414	<0.01	<0.01	<0.03

Location	PTI	Residues (mg/kg)			PTI	Residues (mg/kg)		
		SDS-2787	SDS-3701	SDS-46851		SDS-2787	SDS-3701	SDS-46851
Texas	13	1.28	0.50	<0.03	277	0.05	2.02	<0.03
Oklahoma	0	0.10	0.05	<0.03	422	<0.01	0.01	<0.03
California (Fresno)	0	2.04	0.20	<0.03	360	0.20	0.24	<0.03
North Dakota	224	0.79	0.48	0.04	385	<0.01	0.19	<0.03
California (Greenfield)	0	2.21	0.28	<0.03	413	0.02	0.14	<0.03
Idaho	221	0.13	0.08	<0.03	374	<0.01	0.04	<0.03
Georgia (Parrot)	270	<0.01	0.02	<0.03	328	<0.01	0.01	<0.03
Georgia (Plains)	286	<0.01	0.03	<0.03	429	<0.01	0.01	<0.03
New York	0	4.66	0.33	<0.03	391	<0.01	0.10	<0.03
Louisiana	0	0.27	0.06	<0.03	322	<0.01	<0.01	<0.03
Maryland	32	0.34	0.11	<0.03	420	0.01	0.15	<0.03

A summary of the residues of the parent compound (SDS-2787) and the metabolites SDS-3701 and SDS-4685, resulting in crops grown on chlorothalonil-treated soil, are given in Table 7.9.3.3.

Table 7.9.3.3 Mean residues levels of SDS-2787 and the metabolites SDS-3701 and SDS-46851 in crops

Location	Rotational crop	Rotation interval (days)	PHI (days)	Residue (mg/kg)		
				SDS-2787	SDS-3701	SDS-46851
Georgia (Donalsonville)	Turnip tops	34	60	<0.01	<0.01	0.59
	Turnip roots	34	90	<0.01	<0.01	0.10
	Cabbage	34	180	<0.01	<0.01	<0.03
	Wheat grain	34	253	<0.01	<0.01	<0.03
	Wheat straw	34	253	0.02[1]	<0.01	0.05
	Corn	180	320	<0.01	<0.01	<0.03
	Summer squash	222	291	0.02	<0.01	<0.03
	Peanut vines	222	376	0.22[4]	<0.01	<0.03
	Sweet potato	222	390	<0.01	<0.01	<0.03
	Soybean	253	414	<0.01	<0.01	<0.03
	Cottonseed	253	414	<0.01	<0.01	<0.03
Texas	Spinach	13	100	<0.01	<0.01	<0.03
	Carrot roots	13	175	<0.01	0.02	0.03
	Carrot tops	13	175	0.02[1]	0.02	0.10
	Onions	13	175	0.02[1]	<0.01	<0.03
	Cucumbers	99	190	<0.01	<0.01	0.14
	Bell peppers	99	228	0.01	<0.01	<0.03
	Sorghum grain	99	228	<0.01	<0.01	<0.03
	Sorghum forage	99	228	<0.01	0.04	0.26
	Cottonseed	99	277	<0.01	<0.01	<0.03

Location	Rotational crop	Rotation interval (days)	PHI (days)	Residue (mg/kg)		
				SDS-2787	SDS-3701	SDS-46851
Oklahoma	Wheat grain	59	260	<0.01	<0.01	<0.03
	Wheat straw	59	260	<0.01	<0.02	<0.03
	Potatoes	179	282	<0.01	<0.01	<0.03
	Corn	220	290	<0.01	<0.01	<0.03
	Peanut nutmeats	260	380	<0.01	<0.01	<0.03
	Peanut hulls	260	380	0.02	<0.01	<0.03
	Sorghum	260	380	0.01	<0.01	<0.03
	Cottonseed	260	422	<0.01	<0.01	<0.03
	Cucumbers	262	330	<0.01	<0.01	<0.03
California (Fresno)	Lettuce	31	163	<0.01	<0.01	<0.03
	Wheat forage	31	177	0.01[2]	<0.02	0.08
	Broccoli	31	185	<0.01	<0.01	0.18
	Carrot roots	31	185	<0.01	<0.01	<0.03
	Carrot tops	31	185	<0.01	<0.01	<0.03
	Wheat grain	31	224	<0.01	<0.01	0.04
	Wheat straw	31	224	<0.01	<0.02	0.19
	Onions	31	224	<0.01	<0.01	<0.03
	Sugar beet roots	31	290	<0.01	<0.01	<0.03
	Sugar beet tops	31	290	<0.01	<0.01	0.07
	Tomatoes	177	318	0.01	<0.01	<0.03
	Cottonseed	177	360	0.03	0.01	0.04
North Dakota	Lettuce	257	315	<0.01	0.02	<0.03
	Soybean	259	385	<0.01	<0.01	<0.03
	Potatoes	n.a[3]	350	<0.01	<0.01	0.04
	Sugarbeet roots	n.a	385	0.03[8]	<0.01	<0.03
	Wheat grain	n.a	333	<0.01	<0.01	<0.03
	Wheat straw	n.a	333	<0.01	0.02	<0.03
	Cabbage	n.a	333	<0.01	<0.01	<0.03
California (Greenfield)	Radishes	197	238	<0.01	<0.01	0.10
	Spinach	197	248	<0.01	<0.01	0.11
	Broccoli	197	290	<0.01	<0.01	0.14
	Lettuce	197	290	<0.01	<0.01	<0.03
	Potatoes	197	302	<0.01	<0.01	0.33
	Carrot roots	197	309	<0.01	<0.01	<0.03
	Wheat grain	197	314	<0.01	<0.01	0.26
	Wheat straw	197	314	<0.01	<0.02	0.23
	Sugar beet roots	197	363	0.01	<0.01	0.13
	Celery	197	413	<0.01	<0.01	<0.03
	Fresh peas	223	297	<0.01	<0.01	0.08
	Dry peas	223	309	<0.01	<0.01	<0.03
Idaho	Wheat grain	221	370	<0.01	<0.01	<0.03
	Wheat straw	221	370	<0.01	<0.02	0.09
	Sugarbeet roots	221	374	<0.01	<0.01	<0.03
	Sugarbeet tops	221	374	<0.01	<0.01	<0.03
	Potatoes	245	376	<0.01	<0.01	<0.03
	Carrot roots	249	344	<0.01	<0.01	<0.03
	Carrot tops	249	344	<0.01	<0.01	<0.03
	Dry peas	249	351	<0.01	<0.01	<0.03
	Pea fodder	249	351	0.06[1]	0.07[1]	<0.03
	Rapeseed	249	370	<0.01	<0.01	<0.03
	Bean hay	269	374	0.09[1]	<0.02	0.11
	Dry beans	269	374	<0.01	<0.01	<0.03
Georgia (Parrot)	Wheat grain	83	270	<0.01	<0.01	<0.03
	Wheat straw	83	270	<0.01	<0.02	<0.03
	Corn	208	328	<0.01	<0.01	<0.03
	Sorgum	240	328	0.02[1]	<0.01	<0.03

Location	Rotational crop	Rotation interval (days)	PHI (days)	Residue (mg/kg)		
				SDS-2787	SDS-3701	SDS-46851
Georgia (Plains)	Wheat grain	90	279	<0.01	<0.01	<0.03
	Collards	204	286	0.01	<0.01	<0.03
	Corn	214	351	<0.01	<0.01	<0.03
	Sorghum	280	392	0.03[2]	<0.01	<0.03
	Cottonseed	280	429	<0.01	<0.01	<0.03
New York	Oat grain	179	282	<0.01	<0.01	0.40
	Oat straw	179	282	<0.01	<0.02	2.95
	Corn	199	348	<0.01	<0.01	0.13
	Spinach	229	320	<0.01	0.01	0.80
	Cabbage	229	320	<0.01	<0.01	<0.03
	Carrot tops	229	320	<0.01	<0.01	0.31
	Carrot roots	229	320	<0.01	<0.01	0.23
	Tomatoes	229	327	<0.01	<0.01	0.06
	Winter squash	229	341	<0.01	<0.01	1.05
	Soybean	229	362	<0.01	<0.01	0.09
	Onions	229	391	<0.01	<0.01	<0.03
	Potatoes	236	362	<0.01	<0.01	0.64
Louisiana	Rice	194	322	<0.01	<0.01	<0.03
Maryland	Wheat grain	61	300	<0.01	<0.01	<0.03
	Cantaloupe	284	341	<0.01	<0.01	<0.03
	Lima Beans	284	381	<0.01	<0.01	<0.03
	Tomatoes	284	381	<0.01	<0.01	<0.03
	Carrot tops	284	402	<0.01	<0.01	0.16[5]
	Carrot roots	284	402	<0.01	<0.01	<0.03
	Beet tops	284	402	<0.01	<0.01	0.33[6]
	Beet roots	284	402	<0.01	<0.01	0.20
	Corn	284	402	<0.01	<0.01	0.05
	Soybeans	284	420	<0.01	<0.01	<0.03
	Turnip tops	284	420	0.01	0.01	0.49[7]

1 0.02 mg/kg mean residue in untreated sample

2 0.01 mg/kg mean residue in untreated sample

3 n.a:data not available in field report

4 0.08 mg/kg mean residue in untreated sample

5 0.05 mg/kg mean residue in untreated sample

6 0.04 mg/kg mean residue in untreated sample

7 0.08 mg/kg mean residue in untreated sample

8 apparent residue of 0.05 mg/kg found in one of the two lab duplicates

Conclusions

At all pre-sampling intervals, the major soil residue compound was the parent compound (SDS-2787), followed by the metabolite SDS-3701. The metabolite SDS-46851 was hardly detected in the soil of the test plots. Rotational crops planted into areas previously treated with chlorothalonil formulations under normal label directions did not contain any residues of SDS-2787 (parent compound) larger than 0.03 mg/kg, except peanut vines in Georgia (Donalsonville), where an SDS-2787 level of 0.22 mg/kg was found, and pea fodder and bean hay in Idaho, where SDS-2787 levels amounted 0.06 and 0.09 mg/kg, respectively. The highest SDS-3701 level was found in pea fodder from Idaho and amounted 0.07 mg/kg. Further detected SDS-3701 levels were all at or below 0.04 mg/kg. SDS-46851 was the major metabolite detected in the rotational crops. In Georgia, SDS-46851 levels of 0.59 and 0.10 mg/kg were detected in turnip tops and roots respectively. In Texas, carrot tops, cucumbers and

sorghum forage contained 0.10, 0.14 and 0.26 mg/kg SDS-46851, respectively. In California, broccoli and wheat straw from Fresno contained approximately 0.18 mg/kg SDS-46851. The SDS-46851 levels in other crops remained below 0.08 mg/kg. Radishes, spinach, broccoli and sugar beet roots from California (Greenfield) contained SDS-46851 at levels between 0.10-0.14 mg/kg and potatoes, wheat grain and wheat straw at levels between 0.23-0.33 mg/kg. In Idaho, SDS-46851 was detected in wheat straw and bean hay at levels of 0.09 and 0.11 mg/kg, respectively. In New York, a maximum of SDS-46851 residue was found on oat straw: 2.95 mg/kg. An SDS-46851 level of 1.05 mg/kg was detected on winter squash. SDS-46851 levels on oat grain, corn, spinach, carrot tops and roots, soybean and potatoes ranged from 0.09-0.80 mg/kg. Finally, in Maryland SDS-46851 levels in carrot tops, beet tops and roots and turnip tops amounted 0.16, 0.33, 0.30 and 0.49 mg/kg, respectively.

Limitations

The studies did not reveal any significant limitations. However, it should be noted that the studies were all performed in the USA. The conditions under which growth and cultivation take place including weather can significantly influence the residual behaviour of a plant protection product. The studies should therefore be representative of the areas where Community authorization is granted or envisaged. In this case, the extensive data from several climatic zones in the USA seem to provide an adequate basis on which to evaluate the residue situation in the Member States of the EC.

B.7.6.2.3 Magnitude of residues in rotational crops, study 3

Previous evaluation	Submitted for the purpose of renewal
RMS remark	Acceptable, pendig the submission of storage stability data of chlorothalonil and metabolite SDS-3701 in cereal grain. ~~for results on SDS-46851, however, storage stability has not been fully covered for chlorothalonil and SDS-3701.~~

Report:	K-CA 6.6.2/01. Eversfield S, (~~2014~~ 2017) Chlorothalonil – Residue study on Rotational Crops in Germany and the United Kingdom in 2011 and 2012. Eurofins Agroscience Services Ltd, UK. Report Number S11-00508. (Syngenta File No. A7867A_11262)[a]

[a] -The final report S11-00508 has been amended to include the storage periods for each of the crop rotation samples.

Guidelines

FAO Guidelines on Producing Pesticide Residues Data from Supervised Trials (Rome, 1990).
Commission of the European Communities, General Recommendations for the Design, Preparation and Realization of Residue Trials; (SANCO 7029/V1/95 rev. 5 22/7/1997)
OECD Test Guideline 504. Residues in Rotational Crops (limited Field Studies).
Commission of the European Communities, Testing of plant protection products in rotational crops: (SANCO 7524/V1/95 rev. 2 22/7/1997)
European Commission Guidance Document on Residue Analytical Method, SANCO/825/00 revision 8.1 (16 Nov 2010).

European Commission Guidance for Generating and Reporting Methods of Analysis in Support of Pre-registration Requirements for Annex II (Part A, Section 4) of Directive 91/414, SANCO/3029/99 revision 4 (11 Jul 2000).

GLP

The study was carried out according to the principles of Good Laboratory Practice.

EXECUTIVE SUMMARY

Two field trials were conducted during 2011 and 2012, one in Germany and one in the United Kingdom. Chlorothalonil was applied as A7867A, a suspension concentrate (SC) formulation containing 500 g chlorothalonil per litre to bare soil at a rate of 2000 g a.s./ha. At each rotational interval of 30, 60 and 365 days after application (DAT), a representative cereal (spring wheat or barley), leafy vegetable (spinach) and root vegetable (carrot) were sown into the soil. All crops were grown under field conditions and harvested at immature and mature growth stages. Commodities of representative food and feed items (cereal immature whole plant, mature straw and mature grain; immature and mature spinach; mature carrot foliage and roots) were samples at intervals after sowing and analysed for residues of chlorothalonil, R182281 and R611965. The LOQ was 0.01 mg/kg for all compounds.

At all plant-back intervals no residues of chlorothalonil or R182281 were found at or above the LOQ (0.01 mg/kg) in any of the treated samples.

Residues of R611965 were found in samples taken after the 30 and 60 day plant-back intervals (PBI). For spinach these ranged from 0.02 – 0.06 mg/kg at the 30 day PBI and from 0.01-0.03 mg/kg for the 60 day PBI. In cereals residues of R611965 in immature plant, grain and straw were in the range 0.08 – 0.16 mg/kg, 0.01 – 0.11 mg/kg and 0.09 – 0.29 mg/kg respectively for the 30 day PBI. Residues in cereals for the 60 day PBI were 0.11 mg/kg, 0.02 – 0.09 mg/kg and 0.08 – 0.25 mg/kg in immature plant, grain and straw respectively. For carrots residues in roots were in the range <0.01 – 0.01 mg/kg for the 30 day PBI and were < LOQ for the 60 day PBI. Residues in carrot leaves were in the range <0.01 – 0.04 mg/kg and <0.01 – 0.02 mg/kg for the 30 and 60 day PBIs respectively. No residues of R611965 were found at or above the LOQ in any of the samples from the 365 day plant-back interval.

A1. Test Materials

Test Material	A7867A
Description	Suspension concentrate formulation containing chlorothalonil
Purity	495 g/L
Batch number	SAV0L00018
Stability of test compound	The test substance is assumed to be stable for the period of use in the study

A2. Test System

Trial site	01, Niedersachsen, Germany	02, Oxfordshire, UK
Soil	Loamy sand	Clay loam
Leafy vegetable	Spinach (variety: Tornado)	Spinach (variety: Renegade)

Cereal	Spring wheat (variety: Shasin)	Spring barley (variety: Doyen/Westminster)
Root vegetable	Carrot (variety: Laguna F1)	Carrot (variety: Napoli)

A3. Test Facilities

Field trials	Niedersachsen, Germany	Oxfordshire, UK
Analytical phase	Eurofins Agroscience Services Ltd., Slade Lane, Wilson, Melbourne, Derbyshire DE73 8AG, UK	

B. STUDY DESIGN AND METHODS

B1. Field Phase

Plots were treated with chlorothalonil formulated as a SC at a rate of 2000 g a.s./ha (actual rates were 1956-1988 g a.s./ha) in a spray volume of approximately 300 L/ha to bare soil. The soil was aged for 30, 60/63 and 365 days (trial 1) and 27, 60 and 365 days (trial 2) after which the plots were lightly cultivated before drilling of representative crops of carrot, spinach and spring wheat or barley. The crops were grown outdoors in accordance with usual agricultural practice.

Test Samples

Samples of spinach (immature plant and mature leaves), carrot (roots and tops with leaves) and spring wheat / spring barley (immature whole plant, grain and straw) were taken by hand (or with a combine harvester for mature grain and straw) and the samples were stored deep frozen at <-18 °C before analysis.

Spinach samples were stored for up to 226 days (8 months) for chlorothalonil analysis, which is covered by storage stability studies for chlorothalonil. These samples were stored for maximally 229 days (8 months) for the analysis of R182281, also being covered by storage studies. For R611965 analysis, these samples were maximally stored for 954 days (32 months), while storage stability of R611965 has been demonstrated for 30 months. However, since R611965 is considered stable after 30 months, it is not expected that after 32 months it suddenly is not stable anymore. Therefore, this 2 months difference is considered acceptable.

Cereal samples were stored for up to 237 days (8 months) for chlorothalonil and R182281 analysis. During the expert Peer Review Meeting (#164) storage stability of chlorothalonil and its metabolites has been discussed. It was concluded that chlorothalonil is stable in straw for 9 months and in cereal grain in 62 days. Hence, it can be concluded based on storage time in the study, that results for wheat straw are acceptable, however, for wheat grain, storage stability of chlorothalonil still has to be demontstrated (data gap). For metabolite R182281 (SDS-3701) the expert concluded that stability extrapolation from other high starch content matrix is not acceptable and storage stability in grain still needs to be demonstrated (data gap). For straw, stability is demonstrated up to 12 months, hence the results for straw are considered acceptable.

~~Storage stability for chlorothalonil has been demonstrated in grain, straw and whole plant for at least 12 months. Storage stability for R182281 has been demonstrated for 24 months in high starch crops (covering grain), 12 months in straw, and 24 months in watery crops (covering whole plant), thus~~

~~covering the storage period in this study.~~ For the analysis of R611965, cereal samples were maximally stored for 901 days (30 months), which is covered by storage stability studies of R611965.
Carrot samples have been stored for up to 177 days (6 months) for chlorothalonil and R182281 analysis, which is covered by storage stability studies. These samples have been stored for up to 840 days (28 months) for R611965 analysis, which is covered by storage stability studies.
Some control samples have been analysed in duplo; the duplo samples have been stored for longer periods before their analysis, however, since the first analysis was within the period of demonstrated storage stability, this is considered acceptable.
~~Samples were stored for up to 901 days (30 months) before analysis. Storage stability studies (see Volume 1, 2.7.1) have demonstrated storage stability of chlorothalonil for this time period, except for cereal straw. Stability of chlorothalonil in straw has only been demonstrated for 12 months. SDS-3701 has been shown stable for 24 months in watery crops and high starch crops. This period does not cover the storage of all the samples in this study (30 months). Stability of SDS-46851 has been demonstrated for 30 months.~~

B2. Analytical Phase

Samples were analysed for chlorothalonil and R182281 using method GRM005.01A, and for R611965 using method GRM005.06A. The LOQ was 0.01 mg/kg for all analytes in all commodities. Method GRM005.01A involved extraction with acetone/5M sulphuric acid (95:5 v/v), dilution with water followed by SPE clean up for chlorothalonil or taking up in acetonitrile:water (50:50 v/v) for R182281. Subsequently, analysis was performed by gas chromatography with mass selective detection (GD-MSD) for chlorothalonil and LC-MS/MS for R182281. Method GRM005.06A for R611965 involved extraction with acetone/5M sulphuric acid (95:5 v/v), dilution with water and partitioning with dichloromethane (wheat whole plant only) and tert-butyl ether, evaporation of the organic layer to dryness, and dissolution in methanol. Subsequently, R611965 was analysed by LC-MS/MS. Full method descriptions and validation data are presented in B.5.2.1 (K-CA 4.2/01-K-CA 4.2/07) and B.5.1.2.5 (K-CA 4.12/26).

II. RESULTS AND DISCUSSION

Method Validation

Procedural recoveries were determined for each compound for each commodity. Individual and mean recoveries are summarised in Table 7.6.2.3-1.

Table 7.6.2.3-1: Summary of procedural recoveries for chlorothalonil, R182281 and R611965 in following crops

Commodity	Fortification level (mg/kg)	Chlorothalonil		R182281		R611965	
		Recovery (%)	Mean recovery (%)	Recovery (%)	Mean recovery (%)	Recovery (%)	Mean recovery (%)
Spinach leaves	0.01	84, 87, 84	85	103, 99, 93	98	95	-
	0.10	81, 84, 91	85	85, 83, 85	84	96	-
Cereal whole plant	0.01	97, 93	95	115, 102	109	77	-
	0.10	89, 99	94	100, 99	100	77	-
Cereal grain	0.01	91, 85	88	102, 105	104	97	-
	0.10	95, 90	93	84, 96	90	95	-
Cereal straw	0.01	92, 93	93	101, 99	100	70	-
	0.10	98, 97	98	85, 100	93	75	-
Carrot root	0.01	85, 85	85	101, 101	101	100	-
	0.10	92, 86	89	86, 101	94	100	-
Carrot leaves	0.01	88, 85	87	91, 102	97	105	-
	0.10	93, 88	91	92, 96	94	99	-

Residues in following crops

The results of the rotational crop trials for chlorothalonil, R182281 and R611965 are presented in Tables 7.6.2.3-2 to 7.6.2.3-4. The results are not corrected for recoveries.

Table 7.6.2.3-2: Residues in rotational spinach grown in soil treated with chlorothalonil at 2000 g a.s/ha

Spinach commodity	Interval: treatment to sampling (days)	Trial 01, Germany			Interval: treatment to sampling (days)	Trial 02, UK		
		Chlorothalonil	R182281	R611965		Chlorothalonil	R182281	R611965
Plant-back interval:		**30 days**				**27 days**		
Immature leaves	77	<0.01	<0.01	0.06	77	<0.01	<0.01	0.02
Mature leaves	87	<0.01	<0.01	0.04	88	<0.01	<0.01	0.02
Plant-back interval:		**63 days**				**60 days**		
Immature leaves	98	<0.01	<0.01	0.02	105	<0.01	<0.01	0.01
Mature leaves	104	<0.01	<0.01	0.03	112	<0.01	<0.01	0.01
Plant-back interval:		**365 days**				**365 days**		
Immature leaves	419	<0.01	<0.01	<0.01	440	<0.01	<0.01	<0.01
Mature leaves	437	<0.01	<0.01	<0.01	449	<0.01	<0.01	<0.01

Table 7.6.2.3-3: Residues in rotational cereals grown in soil treated with chlorothalonil at 2000 g a.s/ha

Wheat (trial 01) / barley (trial 02) commodity	Interval: treatment to sampling (days)	Trial 01, Germany			Interval: treatment to sampling (days)	Trial 02, UK		
		Chlorothalonil	R182281	R611965		Chlorothalonil	R182281	R611965
Plant-back interval:		**30 days**				**27 days**		
Immature plant	73	<0.01	<0.01	0.16	85	<0.01	<0.01	0.08
Grain	140	<0.01	<0.01	0.11	145	<0.01	<0.01	0.01
Straw	140	<0.01	<0.01	0.29	145	<0.01	<0.01	0.09
Plant-back interval:		**63 days**				**60 days**		
Immature plant	104	<0.01	<0.01	0.11	112	<0.01	<0.01	0.11
Grain	174	<0.01	<0.01	0.09	173	<0.01	<0.01	0.02
Straw	174	<0.01	<0.01	0.25	173	<0.01	<0.01	0.08
Plant-back interval:		**365 days**				**365 days**		
Immature plant	440	<0.01	<0.01	<0.01	449	<0.01	<0.01	<0.01
Grain	500	<0.01	<0.01	-	529	<0.01	<0.01	<0.01
Straw	500	<0.01	<0.01	-	529	<0.01	<0.01	<0.01

- not analysed.

Table 7.6.2.3-4: Residues in rotational carrot grown in soil treated with chlorothalonil at 2000 g a.s/ha

Carrot commodity	Interval: treatment to sampling (days)	Trial 01, Germany			Interval: treatment to sampling (days)	Trial 02, UK		
		Chlorothalonil	R182281	R611965		Chlorothalonil	R182281	R611965
Plant-back interval:		**30 days**				**27 days**		
Mature roots	126	<0.01	<0.01	0.01	167	<0.01	<0.01	<0.01
Mature tops	126	<0.01	<0.01	0.04	167	<0.01	<0.01	<0.01
Plant-back interval:		**63 days**				**60 days**		
Mature roots	156	<0.01	<0.01	<0.01	176	<0.01	<0.01	<0.01
Mature tops	156	<0.01	<0.01	0.02	176	<0.01	<0.01	<0.01
Plant-back interval:		**365 days**				**365 days**		
Mature roots	483	<0.01	<0.01	<0.01	543	<0.01	<0.01	<0.01
Mature tops	483	<0.01	<0.01	<0.01	543	<0.01	<0.01	<0.01

No residues of chlorothalonil, R182281 or R611965 were found at or above the LOQ (0.01 mg/kg) in any of the untreated samples.

After all plant-back intervals (PBIs) no residues of chlorothalonil or R182281 were found at or above the LOQ (0.01 mg/kg) in any of the treated samples.

No residues of R611965 were found above the LOQ in any of the samples from the 365 day plant-back interval. Residues of R611965 were found in samples taken after the 30 and 60 day PBI. For spinach these were 0.02 – 0.06 mg/kg at the 30 day PBI and 0.01-0.03 mg/kg for the 60 day PBI. In cereals residues of R611965 in immature plant, grain and straw were 0.08 – 0.16 mg/kg, 0.01 – 0.11 mg/kg and 0.09 – 0.29 mg/kg respectively, for the 30 day PBI. Residues in cereals for the 60 day PBI were 0.11 mg/kg, 0.02 -0.09 mg/kg and 0.08-10.25 mg/kg in immature plant, grain and straw, respectively. For carrots residues in roots were <0.01 -0.01 mg/kg for the 30 day PBI and < LOQ for the 60 day PBI. Residues in carrot leaves were <0.01 – 0.04 mg/kg and <0.01 – 0.02 mg/kg for the 30 and 60 day PBIs, respectively.

III. CONCLUSIONS

Residues of chlorothalonil and R182281 were not found above the LOQ in following crops of spinach, cereals and carrot planted at nominal intervals of 30, 60 and 365 days after treatment of bare soil with chlorothalonil at a nominal rate of 2000 g a.s./ha. Residues of R611965 were not found in crops from the 365 day plant-back interval; residues above the LOQ were found in all crops in the 30 and 60 day plant-back interval.

B.7.6.2.4 Magnitude of residues in rotational crops, study 4

Previous evaluation	Submitted for the purpose of renewal
RMS remark	Acceptable, pendig the submission of storage stability data of chlorothalonil and in cereal grain and straw and metabolite SDS-3701 in cereal grain.

Report:	K-CA 6.6.2/02. Eversfield S, (2017a) Chlorothalonil – Residue study on Rotational Crops in Spain and the Italy in 2011 and 2012. Eurofins Agroscience Services Ltd, Report Number S11-00509. (Syngenta File No. A7867A_11264)[a]

[a] The final report S11-00509 has been amended to include the storage periods for each of the crop rotation samples.

Guidelines

FAO Guidelines on Producing Pesticide Residues Data from Supervised Trials (Rome, 1990).
Commission of the European Communities, General Recommendations for the Design, Preparation and Realization of Residue Trials; (SANCO 7029/V1/95 rev. 5 22/7/1997)
OECD Test Guideline 504. Residues in Rotational Crops (limited Field Studies).
Commission of the European Communities, Testing of plant protection products in rotational crops: (SANCO 7524/V1/95 rev. 2 22/7/1997)
European Commission Guidance Document on Residue Analytical Method, SANCO/825/00 revision 8.1 (16 Nov 2010).
European Commission Guidance for Generating and Reporting Methods of Analysis in Support of Pre-registration Requirements for Annex II (Part A, Section 4) of Directive 91/414, SANCO/3029/99 revision 4 (11 Jul 2000).

GLP

The study was carried out according to the principles of Good Laboratory Practice.

EXECUTIVE SUMMARY

Two field trials were conducted during 2011 and 2012, one in Spain and one in the Italy. Chlorothalonil was applied as A7867A, a suspension concentrate (SC) formulation containing 500 g chlorothalonil per litre to bare soil at a rate of 2000 g a.s./ha. At each rotational interval of 30, 60 and 358 days after application (DAT), a representative cereal (spring wheat), leafy vegetable (spinach) and root vegetable (carrot) were sown into the soil. All crops were grown under field conditions and harvested at immature and mature growth stages. Commodities of representative food and feed items (cereal immature whole plant, mature straw and mature grain; immature and mature spinach; mature carrot foliage and roots) were samples at intervals after sowing and analysed for residues of chlorothalonil, R182281 and R611965. The LOQ was 0.01 mg/kg for all compounds.

At all plant-back intervals no residues of chlorothalonil or R182281 were found at or above the LOQ (0.01 mg/kg) in any of the treated samples.

No residues of R611965 were found above the LOQ in the samples from the nominal 30, 60 and 360 day plant-back intervals for carrot and the nominal 60 and 360 day plant-back interval for spinach. No residues of R611965 were found above the LOQ in the nominal 30 and 60 day plant-back intervals for cereal grain and all treated wheat samples for the nominal 360 day plant back interval.

Residues of R611965 were found in some samples taken after the 30 and 60 day plant-back intervals (PBI). For spinach these ranged from <0.01 – 0.02 mg/kg at the 30 day PBI. In cereals residues of R611965 in immature plant, and straw were in the range <0.01 – 0.05 mg/kg and <0.01 – 0.05 mg/kg respectively for the 30 day PBI. Residues in cereals for the 60 day PBI were <0.01 – 0.02 mg/kg and <0.01 – 0.02 mg/kg in immature plant, and straw respectively.

I. MATERIALS AND METHODS

A1. Test Materials

Test Material	A7867A
Description	Suspension concentrate formulation containing chlorothalonil
Purity	495 g/L
Batch number	SAV0L00018
Stability of test compound	The test substance is assumed to be stable for the period of use in the study

A2. Test System

Trial site	01, Maro, Spain	02, Piedmont, Italy
Soil	Sandy loam	Sandy loam
Leafy vegetable	Spinach (variety: Emilia)	Spinach (variety: Virids/Spargo F1)
Cereal	Spring wheat (variety: Glucol)	Spring wheat (variety: Valbona)

Root vegetable	Carrot (variety: Nantesa)	Carrot (variety: Nantes 2)

A3. Test Facilities

Field trials	Maro, Spain	Piedmont, Italy
Analytical phase	Eurofins Agroscience Services Ltd., Slade Lane, Wilson, Melbourne, Derbyshire DE73 8AG, UK	

B. STUDY DESIGN AND METHODS

B1. Field Phase

Plots were treated with chlorothalonil formulated as a SC at a rate of 2000 g a.s./ha (actual rates were 1963-2095 g a.s./ha) in a spray volume of approximately 260 L/ha to bare soil. The soil was aged for 30, 61 and 366 days (trial 1) and 30, 60 and 358 days (trial 2) after which the plots were lightly cultivated before drilling of representative crops of carrot, spinach and spring wheat. The crops were grown outdoors in accordance with usual agricultural practice.

Test Samples

Samples of spinach (immature plant, BBCH 43 in trial 1/BBCH 41 in trial 2; and mature leaves, BBCH 49), carrot (roots and tops with leaves) and spring wheat (immature whole plant, BBCH 39, grain and straw) were taken by hand (or with a combine harvester for mature grain and straw) and the samples were stored deep frozen at <-18 °C before analysis.

Spinach samples were stored for up to 599 days (20 months) for chlorothalonil analysis, which is covered by storage stability studies for chlorothalonil. These samples were stored for maximally 259 days (9 months) for the analysis of R182281, also being covered by storage studies. For R611965 analysis, these samples were maximally stored for 822 days (28 months), which is covered by storage stability studies.

Cereal samples were stored for up to 359 days (12 months) for chlorothalonil and R182281 analysis. During the expert Peer Review Meeting (#164) storage stability of chlorothalonil and its metabolites has been discussed. It was concluded that chlorothalonil is stable in straw for 9 months and in cereal grain in 62 days. Hence, for wheat grain and straw, storage stability of chlorothalonil still has to be demontstrated (data gap). For metabolite R182281 (SDS-3701) the expert concluded that stability extrapolation from other high starch content matrix is not acceptable and storage stability in grain still needs to be demonstrated (data gap). For straw, stability is demonstrated up to 12 months, hence the results for straw are considered acceptable.

~~Storage stability for chlorothalonil has been demonstrated in grain, straw and whole plant for at least 12 months.~~ Storage stability for R182281 has been demonstrated for 24 months in high starch crops (covering grain), 12 months in straw, and 24 months in watery crops (covering whole plant), thus covering the storage period in this study. For the analysis of R611965, cereal samples were maximally stored for 934 days (31 months), while storage stability of R611965 has been demonstrated for 30 months. However, since R611965 is considered stable after 30 months, it is not expected that after 31 months it suddenly is not stable anymore. Therefore, this 1 month difference is considered acceptable.

Carrot samples have been stored for up to 184 days (6 months) for chlorothalonil and R182281 analysis, which is covered by storage stability studies. These samples have been stored for up to 833 days (28 months) for R611965 analysis, which is covered by storage stability studies.

B2. Analytical Phase

Samples were analysed for chlorothalonil and R182281 using method GRM005.01A, and for R611965 using method GRM005.06A. The LOQ was 0.01 mg/kg for all analytes in all commodities. Full method descriptions and validation data are presented in B.5.2.1 (K-CA 4.2/01-K-CA 4.2/07) and B.5.1.2.5 (K-CA 4.12/26).

II. RESULTS AND DISCUSSION

Method Validation

Procedural recoveries were determined for each compound for each commodity. Individual and mean recoveries are summarised in Table 7.6.2.4-1. Linearity was acceptable.

Table 7.6.2.4-1: Summary of procedural recoveries for chlorothalonil, R182281 and R611965 in following crops

Commodity	Fortification level (mg/kg)	Chlorothalonil		R182281		R611965	
		Recovery (%)	Mean recovery (%)	Recovery (%)	Mean recovery (%)	Recovery (%)	Mean recovery (%)
Spinach leaves	0.01	94,86,93	91	93, 84, 92	90	110	-
	0.10	93, 90, 91	91	92, 79, 101	88	95	-
Cereal whole plant	0.01	91, 102	97	104, 111	108	87	-
	0.10	95, 104	100	106, 95	101	93	-
Cereal grain	0.01	103, 95	99	107, 107	107	97	-
	0.10	100, 98	99	97, 99	98	100	-
Cereal straw	0.01	95, 96	96	114, 114	114	109	-
	0.10	97, 93	95	97, 101	99	96	-
Carrot root	0.01	90, 95	93	95, 95	95	95	-
	0.10	92, 83	88	85, 97	91	96	-
Carrot leaves	0.01	93, 92	93	91, 102	97	87	-
	0.10	85, 101	93	95, 92	94	95	-

Residues in following crops

The results of the rotational crop trials for chlorothalonil, R182281 and R611965 are presented in Tables 7.6.2.4-2 to 7.6.2.4-4. The results are not corrected for recoveries.

Table 7.6.2.4-2: Residues in rotational spinach grown in soil treated with chlorothalonil at 2000 g a.s/ha

Spinach	Interval:	Trial 01, Spain	Interval:	Trial 02, Italy

commodity	treatment to sampling (days)	Chlorothalonil	R182281	R611965	treatment to sampling (days)	Chlorothalonil	R182281	R611965
Plant-back interval:		**30 days**				**30 days**		
Immature leaves	93	<0.01	<0.01	<0.01	60	<0.01	<0.01	0.02
Mature leaves	123	<0.01	<0.01	<0.01	82	<0.01	<0.01	-
Plant-back interval:		**61 days**				**60 days**		
Immature leaves	123	<0.01	<0.01	<0.01	89	<0.01	<0.01	<0.01
Mature leaves	155	<0.01	<0.01	<0.01	101	<0.01	<0.01	<0.01
Plant-back interval:		**366 days**				**358 days**		
Immature leaves	432	<0.01	<0.01	Not analysed	417	<0.01	<0.01	<0.01
Mature leaves	439	<0.01	<0.01	Not analysed	426	<0.01	<0.01	<0.01

Table 7.6.2.4-3: Residues in rotational cereals grown in soil treated with chlorothalonil at 2000 g a.s/ha

Wheat commodity	Interval: treatment to sampling (days)	Trial 01, Spain			Interval: treatment to sampling (days)	Trial 02, Italy		
		Chlorothalonil	R182281	R611965		Chlorothalonil	R182281	R611965
Plant-back interval:		**30 days**				**30 days**		
Immature plant	93	<0.01	<0.01	<0.01	68	<0.01	<0.01	0.05
Grain	221	<0.01	<0.01	<0.01	117	<0.01	<0.01	<0.01
Straw	221	<0.01	<0.01	<0.01	117	<0.01	<0.01	0.05
Plant-back interval:		**61 days**				**60 days**		
Immature plant	123	<0.01	<0.01	<0.01	92	<0.01	<0.01	0.02
Grain	230	<0.01	<0.01	<0.01	153	<0.01	<0.01	<0.01
Straw	230	<0.01	<0.01	<0.01	153	<0.01	<0.01	0.02
Plant-back interval:		**366 days**				**358 days**		
Immature plant	432	<0.01	<0.01	<0.01	426	<0.01	<0.01	<0.01
Grain	465	<0.01	<0.01	<0.01	483	<0.01	<0.01	<0.01
Straw	465	<0.01	<0.01	<0.01	483	<0.01	<0.01	<0.01

Table 7.6.2.4-4: Residues in rotational carrot grown in soil treated with chlorothalonil at 2000 g a.s/ha

Carrot commodity	Interval: treatment to sampling (days)	Trial 01, Spain			Interval: treatment to sampling (days)	Trial 02, Italy		
		Chlorothalonil	R182281	R611965		Chlorothalonil	R182281	R611965
Plant-back interval:		**30 days**				**30 days**		
Mature roots	141	<0.01	<0.01	<0.01	153	<0.01	<0.01	<0.01
Mature tops	141	<0.01	<0.01	<0.01	153	<0.01	<0.01	<0.01
Plant-back interval:		**61 days**				**60 days**		
Mature roots	155	<0.01	<0.01	<0.01	159	<0.01	<0.01	<0.01
Mature tops	155	<0.01	<0.01	<0.01	159	<0.01	<0.01	<0.01
Plant-back interval:		**366 days**				**358 days**		
Mature roots	439	<0.01	<0.01	<0.01	475	<0.01	<0.01	<0.01
Mature tops	439	<0.01	<0.01	Not analysed	475	<0.01	<0.01	<0.01

No residues of chlorothalonil, R182281 or R611965 were found at or above the LOQ (0.01 mg/kg) in any of the untreated samples.

After all plant-back intervals (PBIs) no residues of chlorothalonil or R182281 were found at or above the LOQ (0.01 mg/kg) in any of the treated samples.

No residues of R611965 were found at or above the LOQ in any of the samples from the nominal 30, 60 and 360 day plant-back intervals for carrot and the nominal 60 and 360 day plant-back interval for spinach. No residues of R611965 were found above the LOQ in the nominal 30 and 60 day plant-back intervals for cereal grain and in any of the treated wheat samples for the nominal 360 day plant back interval. Residues of R611965 were found in some samples taken after the 30 and 60 day plant-back intervals (PBI). For spinach these ranged from <0.01 – 0.02 mg/kg at the 30 day PBI. In cereals residues of R611965 in immature plant, and straw were in the range <0.01 – 0.05 mg/kg and <0.01 – 0.05 mg/kg respectively for the 30 day PBI. Residues in cereals for the 60 day PBI were <0.01 – 0.02 mg/kg and <0.01 – 0.02 mg/kg in immature plant, and straw respectively.

III. CONCLUSIONS

Residues of chlorothalonil and R182281 were not found above the LOQ in following crops of spinach, cereals and carrot planted at nominal intervals of 30, 60 and 365 days after treatment of bare soil with chlorothalonil at a nominal rate of 2000 g a.s./ha. Residues of R611965 were not found in treated crops from the 365 day plant-back interval, or all treated carrot and grain samples from the 30 and 60 day plant back interval or treated spinach samples from the 30 day plant back interval. Residues above the LOQ were found in some crops in the 30 and 60 day plant-back interval.

B.7.7 Other studies

B.7.7.1 Effect on the residue level in pollen and bee products

For the time being there are no agreed test guidelines for the estimation of the effect of actives on the residue level in pollen and bee products. Therefore, no studies have been conducted (see also SANCO/10181/2013 rev 2.1). Furthermore, potatoes, tomatoes and cereals are not attractive crops to honey-bees and so for the supported representative uses, there is a low likelihood of residues of chlorothalonil and SDS-3701 in pure blossom honey or other bee products from these uses.
The Applicant has not conducted an assessment on the effect on residue levels in pollen and other bee products as there is currently no guidance available for the conduct of such studies.
The data requirement objective of these studies is to determine the residue in pollen and bee products for human consumption resulting from residues taken up by honeybees from crops at blossom.
In the Guidance Document on the risk assessment of plant protection products on bees (EFSA Journal 2013;11(7):3295), information about the relative attractiveness of different crops to honey bees, bumble bees and solitary bees is presented. For honey bees (which is of relevance to the potential transfer of residues from treated crops into edible bee products), in relation to wheat, barley, potato and tomatoes, the crops are generally considered to be of low attractiveness to bees. Thus the potential for transfer of residues into honey is considered to be low for the representative crops and - given the levels of consumption of honey - of no concern for consumer safety.

B.7.8 References relied on

Literature search

A literature search was carried out. A relatively large number of databases were selected for the literature search and justification was provided to support the choice of selected search engines. The search was carried out with the active substance, and for all known metabolites of chlorothalonil. ~~, but it seems that no search has been done with its metabolites. Therefore, there is a data requirement for an additional systematic literature search for the relevant metabolites. However, it has been commented by the applicant that any relevant study on chlorothalonil metabolites would only be present in the public literature in combination with chlorothalonil itself. Accordingly, a separate search on the chlorothalonil metabolites would not retrieve any additional references of interest. This argumentation is being understood, however, the RMS cannot assess whether there are indeed no additional public studies of interest on chlorothalonil metabolites (which are not already retrieved by the existing literature search). Therefore, it seems reasonable to perform the additional systematic literature search.~~

All ~~1354~~ 1506 hits were evaluated for relevance by scanning the study titles and abstracts, and none of them were considered relevant based on a list of criteria. The relevance criteria were based on the following points: well defined test material; applicable test species; study conditions should not differ significantly from guidelines and recommended protocols; trial site/test system not previously exposed to the test material or other contaminants; sufficient experimental information is provided to substantiate and evaluate whether the study conclusions and endpoints are robust; validated analytical methodology employed; study conditions do not interfere with the interpretation of the study results.

Reference list

Data point	Author(s)	Year	Title Company Report No. Source (where different from company) GLP or GEP status Published or not	Vertebrate study Y/N	Data protection claimed Y/N	Justification if data protection is claimed	Owner
~~6.1/01~~	~~Anderson L., Chaggar S.~~	~~2007~~	~~Chlorothalonil (R44686) and R182281 (SDS-3701) - Storage Stability of Field-Incurred Residues in Homogenised Crops stored Deep Frozen for up to Two Years Syngenta Crop Protection AG, Basel, Switzerland Syngenta - Jealott's Hill International, Bracknell, Berkshire, United Kingdom, T000559-06-REG-04-S606 GLP not published Syngenta File No R182281/0023~~	~~N~~	~~Y~~	~~New data; eligible for data protection according to SANCO/12576/2012~~	~~SYN~~
6.1/02	Anderson	2008	Chlorothalonil (R44686) and	N	Y	New data; eligible for	SYN

Data point	Author(s)	Year	Title Company Report No. Source (where different from company) GLP or GEP status Published or not	Vertebrate study Y/N	Data protection claimed Y/N	Justification if data protection is claimed	Owner
	L.		R182281 (SDS-3701) : Storage Stability in Various Crops Prepared in Acid and stored Deep Frozen for up to Two Years Syngenta - Jealott's Hill International, Bracknell, Berkshire, United Kingdom Syngenta - Jealott's Hill International, Bracknell, Berkshire, United Kingdom, T005407-04-REG GLP not published Syngenta File No R44686/4298			data protection according to SANCO/12576/2012	
6.1/03	Brown D.	2014	Chlorothalonil - Storage Stability of residues of Chlorothalonil and R182281 in Cereal Straw for up to 12 months Syngenta Eurofins Agroscience Services Ltd, Wilson, UK, S12-01844 GLP not published Syngenta File No R044686_11076	N	Y	New data; eligible for data protection according to SANCO/12576/2012	Chlorothalonil Task Force
6.1/04	Gasso-Brown D.	2015	Chlorothalonil - Storage Stability of residues of R611965 in Crop Matrices Stored Frozen for up to 30 Months Syngenta Eurofins Agroscience Services Ltd, Wilson, UK, S12-04611 GLP not published Syngenta File No R611965_10041	N	Y	New data; eligible for data protection according to SANCO/12576/2012	Chlorothalonil Task Force
6.1/05	Krainz A.	2007	Chlorothalonil: Frozen Storage Stability in Wheat (Grain And Straw) ARYSTA LIFESCIENCE SAS, Noguères, France RCC Ltd., Itingen, Switzerland, A71256 GLP not published Syngenta File No	N	Y	New data; eligible for data protection according to SANCO/12576/2012	Arysta LifeScience

Data point	Author(s)	Year	Title Company Report No. Source (where different from company) GLP or GEP status Published or not	Vertebrate study Y/N	Data protection claimed Y/N	Justification if data protection is claimed	Owner
			R044686_11197				
6.1/06	Heillaut C., Anderson L.	2007	R613636- Storage stability of residues in processed crop commodities stored deep frozen for up to two years Syngenta - Jealott's Hill International, Bracknell, Berkshire, United Kingdom ADME - Bioanalyses, Vergeze, France, T007198-04-REG GLP not published Syngenta File No R613636/0003	N	Y	New data; eligible for data protection according to SANCO/12576/2012	SYN
6.1/07	Krainz A.	2007a	Frozen Storage Stability in Bovine Muscle, Fat, Liver, Kidney and Cow's Milk ARYSTA LIFESCIENCE SAS, Noguères, France RCC Ltd., Itingen, Switzerland, A71278 GLP not published Syngenta File No R182281_10018	N	Y	New data; eligible for data protection according to SANCO/12576/2012	Arysta LifeScience
6.1/08	Amic S.	2015	Chlorothalonil (R44686) - Storage Stability of Chlorothalonil Metabolite R182281 in Animal Matrices under Freezer Storage Conditions for up to Two Years Syngenta Eurofins Agroscience Services Chem SAS, Vergèze, France, S12-04421 GLP not published Syngenta File No R044686_10047	N	Y	New data; eligible for data protection according to SANCO/12576/2012	Chlorothalonil Task Force
6.1/09	Lister N	2000	Chlorothalonil: Validations of SOP RAM 320/01 for the Determination of Residues in Crops. Zeneca Agrochemicals, Jealott's Hill, United Kingdom , RJ2872B GLP	N	Y	New data; eligible for data protection according to SANCO/12576/2012	SYN

Data point	Author(s)	Year	Title Company Report No. Source (where different from company) GLP or GEP status Published or not	Vertebrate study Y/N	Data protection claimed Y/N	Justification if data protection is claimed	Owner
			not published Syngenta File No R44686/0099				
6.1/10	McGill C., Robinson N.	2002	Chlorothalonil Metabolite R182281 (SDS-3701) : Validation of Analytical Method 384/01 for the Determination of Residues in Bovine Muscle, Fat, Kidney, Liver, Milk and Hen's Eggs Syngenta Crop Protection AG, Basel, Switzerland Syngenta - Jealott's Hill International, Bracknell, Berkshire, United Kingdom, RJ3312B GLP not published Syngenta File No R44686/3317	N	Y	New data; eligible for data protection according to SANCO/12576/2012	SYN
6.2.2/01	Hardwick T.	2014	[14C]-Chlorothalonil - Metabolism of [14C]-Chlorothalonil in the Laying Hen Syngenta Covance Laboratories Limited, Harrogate, UK, 8243812 GLP not published Syngenta File No R044686_11082	Y	Y	New data; eligible for data protection according to SANCO/12576/2012	Chlorothalonil Task Force
6.3.1/01	Schulz D., Trumper C.	2016	Chlorothalonil - Residue study on Field Tomatoes in Southern France, Spain and Italy in 2014 Syngenta Eurofins Agroscience Services Chem GmbH, Hamburg, Germany, S14-02773 GLP not published Syngenta File No A7867A_11391	N	Y	New data; eligible for data protection according to SANCO/12576/2012	Chlorothalonil Task Force
6.3.1/02	Schulz D., Trumper C.	2015	Chlorothalonil - Residue study on Field Tomatoes in Northern France, Poland and Hungary in 2014 Syngenta Eurofins Agroscience	N	Y	New data; eligible for data protection according to SANCO/12576/2012	Chlorothalonil Task Force

Data point	Author(s)	Year	Title Company Report No. Source (where different from company) GLP or GEP status Published or not	Vertebrate study Y/N	Data protection claimed Y/N	Justification if data protection is claimed	Owner
			Services Chem GmbH, Hamburg, Germany, S14-02774 GLP not published Syngenta File No A7867A_11386				
6.3.1/03	Schulz D., Trumper C.	2016a	Chlorothalonil - Residue study on Field Tomatoes in Northern France and Germany in 2015 Syngenta Eurofins Agroscience Services Chem GmbH, Hamburg, Germany, S15-02003 GLP not published Syngenta File No A7867A_11403	N	Y	New data; eligible for data protection according to SANCO/12576/2012	Chlorothalonil Task Force
6.3.2/01	White T.	2013	Chlorothalonil - Residue Study on Barley in Germany, Northern France and the United Kingdom in 2011 Syngenta Eurofins Agroscience Services Ltd, Wilson, UK, S11-00522 GLP not published Syngenta File No A14111B_10905	N	Y	New data; eligible for data protection according to SANCO/12576/2012	SYN
6.3.2/02	White T.	2014	Chlorothalonil - Residue Study on Barley in Germany, Poland and the United Kingdom in 2012 Syngenta Eurofins Agroscience Services Ltd, Wilson, UK, S12-01274 GLP not published Syngenta File No A14111B_10908	N	Y	New data; eligible for data protection according to SANCO/12576/2012	Chlorothalonil Task Force
6.3.2/03	White T.	2014a	Chlorothalonil - Residue Study on Barley in Spain, Italy and Southern France in 2011 Syngenta Eurofins Agroscience Services Ltd, Wilson, UK,	N	Y	New data; eligible for data protection according to SANCO/12576/2012	SYN

Data point	Author(s)	Year	Title Company Report No. Source (where different from company) GLP or GEP status Published or not	Vertebrate study Y/N	Data protection claimed Y/N	Justification if data protection is claimed	Owner
			S11-00523 GLP not published Syngenta File No A14111B_11144				
6.3.2/04	White T.	2014b	Chlorothalonil - Residue Study on Barley in Southern France, Italy and Spain in 2012 Syngenta Eurofins Agroscience Services Ltd, Wilson, UK, S12-01275 GLP not published Syngenta File No A14111B_10899	N	N	New data; eligible for data protection according to SANCO/12576/2012	Chlorothalonil Task Force
6.3.2/05	White T.	2013a	Chlorothalonil - Residue Study on Barley in Southern France in 2013 Syngenta Eurofins Agroscience Services Ltd, Wilson, UK, S13-01041 GLP not published Syngenta File No A14111B_10861	N	Y	New data; eligible for data protection according to SANCO/12576/2012	Chlorothalonil Task Force
6.3.2/06	Sala A.	2014	Determination of Chlorothalonil and its metabolite SDS3701 residues in raw agricultural commodity barley (grain, straw) following two applications of Chlorothalonil 500 SC (2 Trials, Northern Europe, Year 2013) Oxon Italia S.p.A., Pero, Italy Research Centre Biospheres, Salerano sul Lambro, Italy, RAU-020-13 GLP not published Syngenta File No R044686_11190	N	Y	New data; eligible for data protection according to SANCO/12576/2012	Chlorothalonil Task Force
6.3.2/07	Sala A.	2014a	Determination of Chlorothalonil and its metabolite SDS3701 residues in raw agricultural commodity barley (grain,	N	Y	New data; eligible for data protection according to SANCO/12576/2012	Chlorothalonil Task Force

Data point	Author(s)	Year	Title Company Report No. Source (where different from company) GLP or GEP status Published or not	Vertebrate study Y/N	Data protection claimed Y/N	Justification if data protection is claimed	Owner
			straw) following two applications of Clortosip 500 SC (2 Trials, Southern Europe, Year 2013) Oxon Italia S.p.A., Pero, Italy Research Centre Biospheres, Salerano sul Lambro, Italy, RAU-018-13 GLP not published Syngenta File No R044686_11181				
6.3.2/08	Mazzi F.	2014	Determination of Chlorothalonil and its metabolite SDS3701 residues in raw agricultural commodity barley (plant, silage, hay, grain, straw) following two applications of Clortosip 500 SC (Northern Europe - 6 Trials Year 2014) Oxon Italia S.p.A., Pero, Italy Research Centre Biospheres, Salerano sul Lambro, Italy, BIU-017-14 GLP not published Syngenta File No R044686_11180	N	Y	New data; eligible for data protection according to SANCO/12576/2012	Chlorothalonil Task Force
6.3.2/09	Mazzi F.	2014a	Determination of Chlorothalonil and its metabolite SDS3701 residues in raw agricultural commodity barley (plant, silage, hay, grain, straw) following two applications of Clortosip 500 SC (South Europe - 6 Trials Year 2014) Oxon Italia S.p.A., Pero, Italy Research Centre Biospheres, Salerano sul Lambro, Italy, BIU-016-14 GLP not published Syngenta File No R044686_11182	N	Y	New data; eligible for data protection according to SANCO/12576/2012	Chlorothalonil Task Force
6.3.3/01	Lakaschus S., Gizler A.	~~2014~~ 2017	Chlorothalonil - Residue study on Wheat in Northern France, Germany, Poland	N	Y	New data; eligible for data protection according to	Chlorothalonil Task Force

Data point	Author(s)	Year	Title Company Report No. Source (where different from company) GLP or GEP status Published or not	Vertebrate study Y/N	Data protection claimed Y/N	Justification if data protection is claimed	Owner
			and the United Kingdom in 2012 Syngenta Eurofins Agroscience Services Chem GmbH, Hamburg, Germany, S12-01272 GLP not published Syngenta File No A14111B_11147 (including amendment)			SANCO/12576/2012	
6.3.3/02	Lakaschus S., Gizler A.	~~2014a~~ 2017	Chlorothalonil - Residue study on Wheat in Southern France, Italy and Spain in 2012 Syngenta Eurofins Agroscience Services Chem GmbH, Hamburg, Germany, S12-01273 GLP not published Syngenta File No A14111B_11149 (including amendment)	N	Y	New data; eligible for data protection according to SANCO/12576/2012	Chlorothalonil Task Force
6.3.3/03	Sala A.	2014b	Determination of Chlorothalonil and its metabolite SDS3701 residues in raw agricultural commodity winter wheat (grain, straw) following two applications of Chlorothalonil 500 SC (4 Trials, Northern Euope, Year 2013) Oxon Italia S.p.A., Pero, Italy Research Centre Biospheres, Salerano sul Lambro, Italy, RAU-019-13 GLP not published Syngenta File No R044686_11186	N	Y	New data; eligible for data protection according to SANCO/12576/2012	Chlorothalonil Task Force
6.3.3/04	Sala A.	2014c	Determination of Chlorothalonil and its metabolite SDS3701 residues in raw agricultural commodity winter wheat (grain, straw) following two applications of	N	Y	New data; eligible for data protection according to SANCO/12576/2012	Chlorothalonil Task Force

Data point	Author(s)	Year	Title Company Report No. Source (where different from company) GLP or GEP status Published or not	Vertebrate study Y/N	Data protection claimed Y/N	Justification if data protection is claimed	Owner
			Chlorothalonil 500 SC (4 Trials, Southern Euope, Year 2013) Oxon Italia S.p.A., Pero, Italy Research Centre Biospheres, Salerano sul Lambro, Italy, RAU-017-13 GLP not published Syngenta File No R044686_11188				
6.3.3/05	Mazzi F.	2014b	Determination of Chlorothalonil and its metabolite SDS3701 residues in raw agricultural commodity winter wheat (plant, silage, hay, grain, straw) following two applications of Clortosip 500 SC (Northern Euope - 4 Trials Year 2014) Oxon Italia S.p.A., Pero, Italy Research Centre Biospheres, Salerano sul Lambro, Italy, BIU-015-14 GLP not published Syngenta File No R044686_11187	N	Y	New data; eligible for data protection according to SANCO/12576/2012	Chlorothalonil Task Force
6.3.3/06	Mazzi F.	2014c	Determination of Chlorothalonil and its metabolite SDS3701 residues in raw agricultural commodity winter wheat (plant, silage, hay, grain, straw) following two applications of Chlorothalonil 500 SC (South Europe - 4 Trials Year 2014) Oxon Italia S.p.A., Pero, Italy Research Centre Biospheres, Salerano sul Lambro, Italy, BIU-014-14 GLP not published Syngenta File No R044686_11185	N	Y	New data; eligible for data protection according to SANCO/12576/2012	Chlorothalonil Task Force
6.3.4/01	Sala A.	2014d	Determination of	N	Y	New data; eligible for	Oxon Italia

Data point	Author(s)	Year	Title Company Report No. Source (where different from company) GLP or GEP status Published or not	Vertebrate study Y/N	Data protection claimed Y/N	Justification if data protection is claimed	Owner
			Chlorothalonil and its Metabolite SDS3701 Residues in Raw Agricultural Commodity Potato Following Three Applications of Chlorothalonil 500 SC Oxon Italia S.p.A., Pero, Italy Research Centre Biospheres, Salerano sul Lambro, Italy, RAU-022-13 GLP not published Syngenta File No R044686_11232			data protection according to SANCO/12576/2012	SpA*
6.3.4/02	Sala A.	2014e	Determination of Chlorothalonil and its Metabolite SDS3701 Residues in Raw Agricultural Commodity Potato Following Three Applications of Chlorothalonil 500 SC Oxon Italia S.p.A., Pero, Italy Research Centre Biospheres, Salerano sul Lambro, Italy, RAU-011-14 GLP not published Syngenta File No R044686_11234	N	Y	New data; eligible for data protection according to SANCO/12576/2012	Oxon Italia SpA*
6.3.4/03	Sala A.	2014f	Determination of Chlorothalonil and its Metabolite SDS3701 Residues in Raw Agricultural Commodity Potato Following Three Applications of Chlorothalonil 500 SC Oxon Italia S.p.A., Pero, Italy Research Centre Biospheres, Salerano sul Lambro, Italy, RAU-021-13 GLP not published Syngenta File No R044686_11231	N	Y	New data; eligible for data protection according to SANCO/12576/2012	Oxon Italia SpA*
6.3.4/04	Sala A.	2014g	Determination of Chlorothalonil and its	N	Y	New data; eligible for data protection	Oxon Italia SpA*

Data point	Author(s)	Year	Title Company Report No. Source (where different from company) GLP or GEP status Published or not	Vertebrate study Y/N	Data protection claimed Y/N	Justification if data protection is claimed	Owner
			Metabolite SDS3701 Residues in Raw Agricultural Commodity Potato Following Three Applications of Chlorothalonil 500 SC Oxon Italia S.p.A., Pero, Italy Research Centre Biospheres, Salerano sul Lambro, Italy, RAU-022-14 GLP not published Syngenta File No R044686_11233			according to SANCO/12576/2012	
6.4.2/02	Dever M.	2008	The determination of tissue residues as measured by the major metabolite 4-hydroxy-2,5,6-trichloro-l,3-dicyanobenzene in liver, kidney, muscle and fat following daily ingestion by cattle of chlorothalonil over a 28 day period. Vischim Srl, Milano, Italy Veterinary Health Research Pty Ltd, NSW, Australia, SICB1880 GLP not published Syngenta File No R044686_11199	N	Y	New data; eligible for data protection according to SANCO/12576/2012	Oxon Italia SpA*
6.4.2/03	Rogers G.	2008	The determination of the major metabolite, 4-hydroxy-2,5,6-trichloro- 1,3-dicyanobenzene residues in bovine milk following daily ingestion of chlorothalonil by lactating dairy cows over a 28 day period. Vischim Srl, Milano, Italy Veterinary Health Research Pty Ltd, NSW, Australia, SICB1859 GLP not published Syngenta File No R044686_11200	N	Y	New data; eligible for data protection according to SANCO/12576/2012	Oxon Italia SpA*
6.5.1/01	Grout S.J.	2002	Chlorothalonil: Aqueous Hydrolysis at 90, 100 & 120 degrees C Syngenta Crop Protection	N	Y	New data; eligible for data protection according to SANCO/12576/2012	SYN

Data point	Author(s)	Year	Title Company Report No. Source (where different from company) GLP or GEP status Published or not	Vertebrate study Y/N	Data protection claimed Y/N	Justification if data protection is claimed	Owner
			AG, Basel, Switzerland Syngenta - Jealott's Hill, Bracknell, United Kingdom, RJ3331B GLP not published Syngenta File No R44686/3564				
6.5.3/01	Gardinal P.	2007	Chlorothalonil (R44686) - Residue Study on Outdoor Tomatoes and Processed Tomato Products in Southern France Syngenta Crop Protection AG, Basel, Switzerland Syngenta - Jealott's Hill International, Bracknell, Berkshire, United Kingdom, 05-6039 GLP not published Syngenta File No R44686/4093	N	Y	New data; eligible for data protection according to SANCO/12576/2012	SYN
6.5.3/02	Simon P.	2007	Chlorothalonil - Residue study in or on barley and processed barley products in Germany 2004 (Test product A7867A) Syngenta Crop Protection AG, Basel, Switzerland Syngenta Agro GmbH, Maintal, Germany, gba243004 GLP not published Syngenta File No R44686/4112	N	N	New data; eligible for data protection according to SANCO/12576/2012	SYN
6.5.3/03	North L.	2014	Chlorothalonil - Residue Study on Barley and Processed Products in Germany and Southern France in 2011 Syngenta Eurofins Agroscience Services Ltd, Wilson, UK, S11-00524-REG GLP not published Syngenta File No A7867A_11251	N	Y	New data; eligible for data protection according to SANCO/12576/2012	SYN
6.5.3/04	Sala A.	2014h	Determination of Chlorothalonil and its	N	Y	New data; eligible for data protection	Chlorothalonil Task Force

Data point	Author(s)	Year	Title Company Report No. Source (where different from company) GLP or GEP status Published or not	Vertebrate study Y/N	Data protection claimed Y/N	Justification if data protection is claimed	Owner
			metabolite SDS3701 residues and R613636 in raw agricultural commodity barley and processed commodity (pot barley, brewing malt, beer) following two applications of Chlorothalonil 500 SC (Northern and Southern Europe - 2 Trials Year 2014) Oxon Italia S.p.A., Pero, Italy Research Centre Biospheres, Salerano sul Lambro, Italy, RAU-008-14 GLP not published Syngenta File No R044686_11189			according to SANCO/12576/2012	
6.5.3/05	Gill J., Sutra G.	2001	Chlorothalonil Residue Levels in Wheat and Processed Wheat Products from Trials carried out in France during 1999. Zeneca Agrochemicals, Jealott's Hill, United Kingdom Zeneca Agrochemicals, Jealott's Hill, United Kingdom, RJ3094B GLP not published Syngenta File No R44686/2186	N	Y	New data; eligible for data protection according to SANCO/12576/2012	SYN
6.5.3/06	Gill J., Myles P.	2001	Residue Levels in Wheat and Processed Wheat Products from Trials carried out in the UK during 1999 Syngenta Crop Protection AG, Basel, Switzerland Syngenta - Jealott's Hill International, Bracknell, Berkshire, United Kingdom, RJ3095B GLP not published Syngenta File No R44686/2187	N	Y	New data; eligible for data protection according to SANCO/12576/2012	SYN
6.5.3/07	North L.	2014a	Chlorothalonil - Residue Study on Wheat and Processed Products in Germany and Northern	N	Y	New data; eligible for data protection according to SANCO/12576/2012	SYN

Data point	Author(s)	Year	Title Company Report No. Source (where different from company) GLP or GEP status Published or not	Vertebrate study Y/N	Data protection claimed Y/N	Justification if data protection is claimed	Owner
			France in 2011 Syngenta Eurofins Agroscience Services Ltd, Wilson, UK, S11-00526-REG GLP not published Syngenta File No A7867A_11256				
6.5.3/08	Sala A.	2015	Determination of Chlorothalonil and its Metabolites in Winter Wheat Oxon Italia SpA Research Centre Biospheres, Salerano sul Lambro, Italy, RAU-007-14, AB2-14-19393 GLP not published Syngenta File No R044686_11359	N	Y	New data; eligible for data protection according to SANCO/12576/2012	Chlorothalonil Task Force
6.6.1/01	Rizzo F., Ferrario F.	2005	Uptake, translocation and metabolism of 14C-Chlorothalonil in rotated crops of spring wheat, carrots and lettuce. Isagro Ricerca Srl, Novara, Italy Caffaro SpA, Cesano Maderno, Italy, MEF.03.03 GLP not published Syngenta File No R044686_11201	N	Y	New data; eligible for data protection according to SANCO/12576/2012	Oxon Italia SpA*
6.6.1/02	Mamouni A.	2009	14C-Chlorothalonil Confined Accumulation in Rotational Crops ARYSTA LIFESCIENCE SAS, Noguères, France Harlan Laboratories Ltd., Itingen, Switzerland, B34931 GLP not published Syngenta File No R044686_11194	N	Y	New data; eligible for data protection according to SANCO/12576/2012	Arysta LifeScience
6.6.2/01	Eversfield S.	~~2014~~ 2017	Chlorothalonil - Residue Study on Rotational Crops in Germany and the United Kingdom in 2011 and 2012 Syngenta Eurofins Agroscience	N	Y	New data; eligible for data protection according to SANCO/12576/2012	SYN

Data point	Author(s)	Year	Title Company Report No. Source (where different from company) GLP or GEP status Published or not	Vertebrate study Y/N	Data protection claimed Y/N	Justification if data protection is claimed	Owner
			Services Ltd, Wilson, UK, S11-00508 GLP not published Syngenta File No A7867A_11262 (including amendment)				
KCA 6.6.2 / 02	Eversfield S.	2017a	Chlorothalonil - Residue Study on Rotational Crops in Spain and in Italy in 2011 and 2012 Syngenta Eurofins Agroscience Services Ltd, Wilson, UK, S11-00509 GLP not published Syngenta File No A7867A_11264 (including amendment)	N	Y	New data; eligible for data protection according to SANCO/12576/2012	SYN

*OXON Italia SpA acquired Vischim S.r.l. in November 2011.

Studies relied on for the first inclusion of Chlorpropham in Annex I to Directive 91/414/EEC and for renewal of approval under Regulation (EC) 1107/2009

Data point	Author(s)	Year	Title Company Report No. Source (where different from company) GLP or GEP status Published or not	Vertebrate study Y/N	Data protection claimed Y/N	Owner
~~6.1~~	~~King, C.~~	~~1993~~	~~Determination of residues of tetrachloroisophthalo-nitrile (chlorothalonil, SDS-2787), SDS-3701, SDS-46851, HCB, and PCBN in cherries from a stability study (field incurred) - 1988 - Four-year interim report.~~ ~~Generated by: Ricerca, Inc.~~ ~~Submitted by: ISK Biosciences Corp./Zeneca~~ ~~Report No.: 3064-88-0068-CR-002~~ ~~Date: June 18, 1993~~ ~~GLP, Unpublished~~	~~N~~	~~N~~	
~~6.1~~	~~Kenyon, R. G.~~	~~1993~~	~~Determination of residues of tetrachloroisophthalo-nitrile (chlorothalonil, SDS-2787), SDS-3701, SDS-46851, HCB, and PCBN in wheat grain from a stability study (field incurred) - 1988 - Four-year interim report.~~ ~~Generated by: Ricerca, Inc.~~ ~~Submitted by: ISK Biosciences Corp./Zeneca~~ ~~Report No.: 3064-88-0070-CR-002~~ ~~Date: June 9, 1993~~	~~N~~	~~N~~	
6.1	~~Kenyon, R. G.~~	~~1993~~	~~Determination of residues of tetrachloroisophthalo-nitrile (chlorothalonil, SDS-2787), SDS-3701,~~	~~N~~	~~N~~	

Data point	Author(s)	Year	Title Company Report No. Source (where different from company) GLP or GEP status Published or not	Vertebrate study Y/N	Data protection claimed Y/N	Owner
			SDS-46851, HCB, and PCBN in tomatoes from a stability study (field incurred) - 1988 - Four year interim report. Generated by: Ricerca, Inc. Submitted by: ISK Biosciences Corp./Zeneca Report No.: 3064-88-0083-CR-002 Date: June 21, 1993			
6.1	Wiedmann, J.L.	1993	Determination of residues of tetrachloroiso-phthalonitrile (chlorothalonil, SDS-2787), SDS-3701, SDS-46851, HCB, and PCBN in cucumbers from a stability study (field incurred) - 1988 - Four year interim report. Generated by: Ricerca, Inc. Submitted by: ISK Biosciences Corp./Zeneca Report No.: 3064-88-0093-CR-002 Date: June 23, 1993	N	N	
6.1	Rose, C.A.	1993	Determination of residues of tetrachloroisophthalo-nitrile (chlorothalonil, SDS-2787), SDS-3701, SDS-46851, HCB, and PCBN in potatoes from a stability study (field incurred) - 1988 - Four year interim report. Generated by: Ricerca, Inc. Submitted by: ISK Biosciences Corp./Zeneca Report No.: 3064-88-0095-CR-002 Date: July 30, 1993			
6.1	Rose, C.A.	1993	Determination of residues of tetrachloroisophthalo-nitrile (chlorothalonil, SDS-2787), SDS-3701, SDS-46851, HCB, and PCBN in carrots from a stability study (field incurred) - 1988 - Four year interim report. Generated by: Ricerca, Inc. Submitted by: ISK Biosciences Corp./Zeneca Report No.: 3064-88-0096-CR-002 Date: July 1, 1993	N	N	
6.1	Kenyon, R. G.	1993	Determination of residues of tetrachloroisophthalo-nitrile (chlorothalonil, SDS-2787), SDS-3701, SDS-46851, HCB, and PCBN in soybeans from a stability study (field incurred) - 1988 - Four year interim report. Generated by: Ricerca, Inc. Submitted by: ISK Biosciences Corp./Zeneca Report No.: 3064-88-0097-CR-002 Date: June 22, 1993	N	N	
6.1	King, C.	1993	Determination of residues of tetrachloroisophthalo-nitrile (chlorothalonil, SDS-2787), SDS-3701, SDS-46851, HCB, and PCBN in celery from a stability study (field incurred) - 1988 - Four year interim report. Generated by: Ricerca, Inc. Submitted by: ISK Biosciences Corp./Zeneca Report No.: 3064-88-0136-CR-002 Date: June 25, 1993	N	N	
6.1	Wiedmann,	1993	Determination of residues of tetrachloroiso-	N	N	

Data point	Author(s)	Year	Title Company Report No. Source (where different from company) GLP or GEP status Published or not	Vertebrate study Y/N	Data protection claimed Y/N	Owner
	~~J.L.~~		~~phthalonitrile (chlorothalonil, SDS-2787), SDS-3701, SDS-46851, HCB, and PCBN in almond hulls and nutmeats from a stability study (field incurred) - 1988 - Four year interim report.~~ ~~Generated by: Ricerca, Inc.~~ ~~Submitted by: ISK Biosciences Corp./Zeneca~~ ~~Report No.: 3064-88-0158-CR-002~~ ~~Date: May 30, 1993~~			
6.1	~~King, C.~~	~~1993~~	~~Determination of residues of tetrachloroisophthalo-nitrile (chlorothalonil, SDS-2787), SDS-3701, SDS-46851, HCB, and PCBN in peanuts from a stability study (field incurred) - 1988 - Four year interim report.~~ ~~Generated by: Ricerca, Inc.~~ ~~Submitted by: ISK Biosciences Corp./Zeneca~~ ~~Report No.: 3064-88-0160-CR-002~~ ~~Date: August 2, 1993~~	~~N~~	~~N~~	
6.1	King C. and Pince P.	1995	Freezer storage stability of SDS-3701 in milk and cow tissue Syngenta, UK Report no.: 5927-93-0329-CR-001 GLP, not published	N	N	
6.2	Nelsen, T. R., Marks, A. F.	1985	A plant metabolism study with 14C chlorothalonil (2,4,5,6 tetrachloroisophthalonitrile) on lettuce. Generated by: SDS Biotech Corporation Submitted by: ISK Biosciences Corp./ Zeneca Report No.: 672 3EF 84 0014 001 Date: January 7, 1985	N	N	
6.2	Nelsen, T.F.	1988	A plant metabolism study with 14C chlorothalonil (2,4,5,6 tetrachloroisophthalonitrile) on tomatoes. Generated by: Ricerca, Inc. Submitted by: ISK Biosciences Corp./Zeneca Company File No.: 1184 85 0052 EF 001 Date: June 7, 1988	N	N	
6.2	lsen, T. F. Marks, A. F.	1987	A plant metabolism study with 14C chlorothalonil (2,4,5,6 tetrachloroisophthalonitrile) on carrots. Generated by: Ricerca, Inc. Submitted by: ISK Biosciences Corp./Zeneca Company File No.: 1186 86 0026 EF 001 Date: April 14, 1987	N	N	
6.2	Huhtanen, K. L., Doran, T. J.	1992	A plant metabolism study with 14C-Chlorothalonil (2,4,5,6-tetrachloroisophthalonitrile) on celery. Generated by: Ricerca, Inc. Submitted by: ISK Biosciences Corp./Zeneca Compay File No.: 3503-90-0184-EF-001 Date: October 6, 1992	N	N	
6.2	Huhtanen, K. L., Doran, T. J.	1993	A plant metabolism study with 14C-chlorothalonil (2,4,5,6-tetrachloroisophthalonitrile) on snapbeans. Generated by: Ricerca, Inc. Submitted by: ISK Biosciences Corp./Zeneca Report No.: 5216-92-0063-EF-001 Date: September 23, 1993	N	N	

Data point	Author(s)	Year	Title Company Report No. Source (where different from company) GLP or GEP status Published or not	Vertebrate study Y/N	Data protection claimed Y/N	Owner
6.2	Mayo, B.	1996	Chlorothalonil - Metabolism in Tomato Generated by: Huntingdon Life Sciences Ltd Submitted by: Vischim S.R.L. Company file No.: VCM 44/950175 Date: January 23, 1996	N	N	
6.2	Mayo, B.	1996	Chlorothalonil - Metabolism in Wheat Generated by: Huntingdon Life Sciences Ltd Submitted by: Vischim S.R.L. Company file no.: VCM 38/950767 Date: November 29, 1996	N	N	
6.2	McEwen, A.	1997	Chlorothalonil - Metabolism in Peas Generated by: Huntingdon Life Sciences Ltd Submitted by: Vischim S.R.L. Company file No.: VCM 68/962010 Date: February 10, 1997	N	N	
6.2	Capps, T. M.	1983	Poultry and egg residue study with 2,4,5,6-tetrachloroisophthalonitrile (14C-DS-2787). Generated by: Diamond Shamrock Corp. and BioLife Associates, Ltd. Submitted by: ISK Biosciences Corp./Zeneca Report No.: 596-4AM-82-0122-002 Date: February 24, 1983	Y	N	
6.2	Capps, T. M., Marciniszyn, J. P.	1984	Poultry and egg residue study with 4-hydroxy-2,5,6-trichloroisophthalonile (14C-DS-3701). Generated by: Ricerca, Inc. and Bio-Life Associates, Ltd. Submitted by: ISK Biosciences Corp./Zeneca Report No.: 596-4AM-82-0123-002 & 596-4AM-82-0123-002-001 Date: February 24, 1983 and February 17, 1984	Y	N	
6.2	Duane, W. C., Doran, T. J.	1990	A study to determine the nature of the residues in meat, milk and tissues from lactating goats fed 14C-Chlorothalonil (2,4,5,6-tetrachloroisophthalonitrile). Generated by: Ricerca, Inc. and Analytical Bio-Chemistry Laboratories, Inc. Submitted by: ISK Biosciences Corp./Zeneca Report No.: 1067-85-0080-EF-001 Date: June 15, 1990	Y	N	
6.2	Ku, H. S.	1990	A study to determine the nature of the residue in meat, milk and tissues from lactating goats dosed with 14C-4-hydroxy-2,5,6-trichloroisophthalonitrile (SDS-3701). Generated by: Ricerca, Inc. and Analytical Bio-Chemistry Laboratories, Inc. Submitted by: ISK Biosciences Corp./Zeneca Report No.: 1183-87-0024-EF-001 Date: February 20, 1990	Y	N	
6.2	Shaw, D.	1997	14C-Chlorothalonil - Metabolism in the lactating goat	Y	N	

Data point	Author(s)	Year	Title Company Report No. Source (where different from company) GLP or GEP status Published or not	Vertebrate study Y/N	Data protection claimed Y/N	Owner
			Generated by: Huntingdon Life Sciences Ltd Submitted by: Vischim S.R.L. Company file No.: VCM 73/961389 Date: May 15, 1997			
6.4	Wiedmann and Kenyon	1995	Meat and milk magnitude of residue study in lactating dairy cows dosed with chlorothalonil and SDS-3701. Ricerca Inc, Syngenta Report No. 6007-94-0120-CR-003 (Syngenta File No. R44686/1598).	Y	N	
6.5	King, C., Prince, P. M.	1993	Magnitude of residues following applications of Bravo 720 to potatoes - processing study Generated by: Ricerca, Inc. and The National Food Laboratory Submitted by: ISK Biosciences Corp./Zeneca Report No.: 5232-92-0105-CR-001 Date: April 2, 1993	N	N	
6.5	Marks, A. F.	1983	The effect of cooking 2,4,5,6-tetrachloroiso-phthalonitrile (chlorothalonil DS-2787) with vegetables. Generated by: Diamond Shamrock Corporation Submitted by: ISK Biosciences Corp./Zeneca Company File No: 372-3EF-83-0004-001 Date: May 18, 1983	N	N	
6.5	Ballee, D. L., Szalkowski, M. B., Stallard, D. E.	1980	Distribution of 2,4,5,6-tetrachloroiosphthalonitrile (chlorothalonil, DS-2787) and 4-hydroxy-2,5,6-trichloroisophthalonitrile (DS-3701) among the products of snapbean processing. Generated by: Diamond Shamrock Corp. Submitted by: ISK Biosciences Corp./Zeneca Report No.: 117-3CR-79-0136-001 Date: March 21, 1980	N	N	
6.5	Stallard, D. E. Ballee, D. L.	1977	The fate of chlorothalonil in the processing of cherries. Generated by: Diamond Shamrock Corp. Submitted by: ISK Biosciences Corp./Zeneca Report No.: 1000-3CR-77-2108-001 Date: December 14, 1977	N	N	
6.5	Ballee, D. L., Stallard, D. E.	1977	The fate of chlorothalonil in the processing of peaches. Generated by: Diamond Shamrock Corp. Submitted by: ISK Biosciences Corp./Zeneca Report No.: 1000-3CR-77-2109-001 Date: July 14, 1977	N	N	
6.5	Prince, P. M., King, C., Wiedmann, J. L.	1994	Magnitude of chlorothalonil residues in grapes and processed fractions. Generated by: Ricerca, Inc. Submitted by: ISK Biosciences Corp./Zeneca Report No.: 5919-94-0017-CR-001 Date: December 27, 1994	N	N	
6.5	Kenyon, R. G., Ballee, D. L.	1983	Distribution of residues of tetrachloroisophthalo-isophthalonitrile (chlorothalonil, DS-2787) and 4-hydroxy-trichloroisophthalonitrile (DS-3701) among the products of apple processing.	N	N	

Data point	Author(s)	Year	Title Company Report No. Source (where different from company) GLP or GEP status Published or not	Vertebrate study Y/N	Data protection claimed Y/N	Owner
			Generated by: SDS Biotech Corporation Submitted by: ISK Biosciences Corp./Zeneca Report No.: 607-3CR-83-0005-001 Date: July 29, 1983			
6.5	Stallard, D. E., Marks, A. F.	1983	Distributions or residues of 2,4,5,6-tetrachloroisophthalonitrile (chlorothalonil, DS-2787), 4-hydroxy-2,5,6-tetrachloroisophthalonitrile (DS-3701), hexachlorobenzene (HCB) and pentachlorobenzonitrile (PCBN) among products obtained from the milling of wheat grain. Generated by: SDS Biotech Corporation Submitted by: ISK Biosciences Corp./Zeneca Report No.: 573-3CR-82-0036-002 Date: March 3, 1983	N	N	
6.5	Szalkowski, M. B., Ballee, D. L.	1980	The effect of commercial processing upon the residue of 2,4,5,6-tetrachloroisophthalonitrile (chlorothalonil, DS-2787) on tomatoes. Generated by: Diamond Shamrock Corp. Submitted by: ISK Biosciences Corp./Zeneca Report No.: 411-3CR-80-0054-001 Date: October 29, 1980	N	N	
6.5	King, C., Ballee, D. L.	1987	Residues of tetrachlorisophthalonitrile (chlorothalonil, SDS-2787), SDS-3701, SDS-46851, HCB and PCBN on cucumbers - Processing study - 1985 and 1986. Generated by: Ricerca., Inc. Submitted by: ISK Biosciences Corp./Zeneca Report No.: 1351-86-0059-CR-001 Date: October 9, 1987	N	N	
6.5	King, C., Prince, P. M.	1990	Residues of tetrachloroisophthalonitrile (chlorothalonil, SDS-2787), SDS-3701, SDS-46851, HCB and PCBN on carrots - Processing study - 1988 Generated by: Ricerca., Inc. Submitted by: ISK Biosciences Corp./Zeneca Report No.: 3186-89-0286-CR-001 Date: April 26, 1990	N	N	
6.5	Ballee, D. L., Szalkowski, M. B.	1980	Distribution of residues of 2,4,5,6-tetrachloroisophthalonitrile (chlorothalonil, DS-2787) and 4-hydroxy-2,5,6-trichloro-isophthalonitrile (DS-3701) among products obtained from the processing of citrus. Generated by: Diamond Shamrock Corp. Submitted by: ISK Biosciences Corp./Zeneca Report No.: 017-3CR-80-0047-001 Date: 1980	N	N	
6.5	Kenyon, R. G., Ballee, D. L.	1986	Residues of tetrachloroisophthalonitrile (chlorothalonil, SDS-2787), SDS-3701, SDS-46851 HCB and PCBN on peanuts - Processing study. Generated by: Ricerca, Inc. Submitted by: ISK Biosciences Corp./Zeneca Report No.: 1155-86-0006-CR-001 Date: October 14, 1996	N	N	

Data point	Author(s)	Year	Title Company Report No. Source (where different from company) GLP or GEP status Published or not	Vertebrate study Y/N	Data protection claimed Y/N	Owner
6.5	Prince, P. M., King, C.	1993	Determination of the magnitude of residues, tetrachloroisophthalonitrile (chlorothalonil, SDS-2787), SDS-3701 and HCB on prunes - Processing Study - 1992. Generated by: Ricerca, Inc. Submitted by: ISK Biosciences Corp./Zeneca Report No.: 5485-93-0022-CR-001 Date: July 23, 1993	N	N	
6.5	King, C., Prince, P. M.	1990	Residues of tetrachloroisophthalonitrile (chlorothalonil, SDS-2787), SDS-3701, SDS-46851 HCB and PCBN on winter squash - Processing study. Generated by: Ricerca, Inc. Submitted by: ISK Biosciences Corp./Zeneca Report No.: 3185-89-0287-CR-001 Date: May 11, 1990	N	N	
6.5	Kenyon, R.G. Ballee, D.L. Marks, A.F.	1987	Residues of tetrachloroisophthalonitrile (chlorothalonil, SDS-2787), SDS-3701, SDS-46851, HCB and PCBN on soybeans - processing study Generated by: Ricerca, Inc. Submitted by: ISK Biosciences Corp./Zeneca Report No.: 1350-86-0058-CR-001 Date: April 15, 1987	N	N	
6.5	**Fitzgerald, T. J., Kenyon, R. G.**	1993	Magnitude of residues of tetrachloroisophthalonitrile (chlorothalonil, SDS-2787), SDS-3701 and HCB on process corn fractions - 1992. Generated by: Ricerca, Inc. Report No.: 5528-93-0090-CR-001 Date: September 24, 1993	N	N	
6.6	Nelsen, T. R., Marks, A. F.	1995	An indoor crop rotation study with 14C-chlorothalonil (2,4,5,6-tetrachloroisophthalonitrile). Generated by: SDS Biotech Corporation Submitted by: ISK Biosciences Corp./Zeneca Report No.: 608-4EF-82-0169-001 Date: December 15, 1995	N	N	
6.6	Dillon K. A., Ballee, D. L.	1984	Field rotation crop study with Bravo ® 500. Generated by: SDS Biotech Corporation Submitted by: Biosciences Corp./Zeneca Report No.: 535-3CR-81-0199-001 and 535-3CR-81-0199-001-001 (Report Addendum) Date: February 7, 1984	N	N	
6.6	Rose, C. A., Kenyon, R. G., Dillon, K. A. Wiedmann, J. L., Ballee, D. L.	1991	Rotational crop study: Summary of residues of tetrachloroisophthalonitrile (chlorothalonil, SDS-2787), its metabolites and manufacturing impurities in first season rotated crops and corresponding soils from Bravo® treated areas - 1985 - 1987. Generated by: Ricerca, Inc. Submitted by: ISK Biosciences Corp./Zeneca Report No.: 1401-86-0084-CR-019 Date: August 9, 1991	N	N	