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Report prepared in the context of the application for first inclusion of dodine in Annex I of the Council Directive 91/414/EEC

DODINE

Volume 3-4 rev.1

Annex B

Section B8

Summary, evaluation and assessments of the data.

List of tests and studies relied upon

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B.8.1.1 Aerobic degradation in soil (Annex IIA 7.1.1.1.1)

^{14}C-dodine: Aerobic Soil metabolism in two soils at 25ºC

Cooper J. D. L. *et al.* (1996)

Guidelines and GLP

The study was conducted according to the EPA Guidelines, Subdivision N, Paragraph 162-1 (1982). Principles of GLP were complied with.

Materials and methods

The degradation of dodine, ^{14}C-guanidine labelled (≥96.5% radiochemical purity; 55 mCi/mole specific activity; batch nº950214) was studied in two sandy loam soils, one from American origin and other from UK. The soils characteristics are summarized in the Table B.8 1. Non-radiolabelled dodine (purity of 100%; batch EA840SD2) was used to dilute the radiolabelled compound prior to dosing.

Portions of soils (aprox. 100 g over dried equivalent) were placed in uniquely labelled soil flasks and treated with ^{14}C-dodine solution at a concentration equivalent to a rate of 4.48 kg dodine/ha (5x the maximum field application rate of 900 g dodine/ha assuming 100% soil interception which corresponds to 4.4 mg dodine/kg dry soil). The metabolism was followed for 100 days, at which time the pattern of degradation of the compound (and the formation and degradation of metabolites) was clearly established. The incubation was carried out under aerobic conditions, in the dark at 25°C with the soils at 75% of their 1/3 bar moisture holding capacity.

Table B.8 1 - Soils used to investigate degradation and metabolism of dodine under aerobic conditions

Soil designation **Origin** **Soil type**	**Soil 1** **Essex / UK** **Sandy loam**	**Soil 2** **Mississippi / USA** **Sandy loam**
Textural analysis (USDA) 2000 - 50 µg, sand < 50 - 2 µg, silt <2 µg, clay	 62.7 % 28.5 % 8.8%	 65 % 29 % 6%
pH value Water $CaCl_2$, 0.01 N	5.3 4.6	5.9 -
Organic C (%)	2.2 %	0.6 %
Cation exchange capacity (meq/100g)	6.2	9.3
Bulk density (g /ml)	1.19	1.18
% Moisture holding capacity (0.3 bar)	16	17
Microbial biomass (mg microbial carbon/kg soil)	366	29 / 186*

*recovered to 186 at the end of the study

The flasks were incorporated into a closed system in which moist, CO_2-free air was continuously drawn over the soils. Volatiles and CO_2 were trapped in ethylene glycol and potassium hydroxide respectively. At intervals up to and including 100 days after treatment the soils were extracted with solvent (basified methanol followed by acidified methanol). The extracts were analysed by thin layer chromatography (TLC) and high performance liquid chromatography (HPLC). The unextracted radioactivity was assayed using a combustion technique. Selected extracts were examined by liquid chromatography followed by mass spectrometry (LC- MS).

Duplicate samples were analysed at 0, 1,3, 5, 7, 10, 14, 21, 28, 56, 84 and 100 days incubation.

Findings

Recoveries of applied radioactivity ranged between 88.6-106.6%. The major metabolite detected was carbon dioxide which accounted for approx. 91 % of the applied amount in the UK sandy loam and 92% in the US sandy loam after 100 days. The bound residues remained at low levels with a maximum of approx. 11.4% AR during the first 10 days decreasing to values near 3% at the end of the study, for both soils. During HPLC and TLC analysis, the non-volatile compounds that were detected in the soil extracts were seen to comprise dodine plus a cluster of minor metabolites (called 'M1 cluster', up to eleven compounds). In concentrated extracts several other very minor metabolites were detected. A polar metabolite (M7) was detected in the extracts of the last two time-points. The metabolites combined never accounted for as much as 10% of applied material and at the end of the study accounted for less than 1 % AR (see Table B.8 2).

Table B.8 2 - Recovery of applied radioactivity in % and distribution of metabolites after application of 14C-dodine to soil under aerobic conditions at a rate equivalent to 4.4 kg dodine/ha (4.4 mg dodine/kg dry soil)

Soil	Days after application	Dodine (%)	$^{14}CO_2$ (%)	M1 "cluster"	M7 "polar"	Bound residues	Total (%)
Soil 1 Essex / UK Sandy loam	0	80.5	-	9.7	< 0.1	1.06	100.8
	1	69.7	7.8	5.3	< 0.1	9.3	92.1
	3	64.7	17.3	2.7	< 0.1	9.6	94.3
	5	56.9	32.2	3.2	< 0.1	8.7	101
	7	45.2	42.7	3.1	< 0.1	6.9	97.9
	10	34.5	41.9	1.4	< 0.1	10.8	88.6
	14	31.2	66.6	1.8	< 0.1	7.0	106.6
	21	18.8	73.1	2.7	< 0.1	4.7	99.3
	28	9.1	81.3	2.4	< 0.1	4.1	96.9
	56	2.6	87.7	0.9	< 0.1	2.9	94.1
	84	2.3	91.9	0.5	0.2	2.7	97.6
	100	1.9	91.8	0.6	0.3	2.7	97.3
Soil 2 Mississippi / USA Sandy loam	0	81.6	-	7.6	< 0.1	9.8	99
	1	70.1	8.1	5.0	< 0.1	9.3	92.5
	3	64.8	16.2	2.3	< 0.1	9.9	93.2
	5	59.9	28.7	2.5	< 0.1	8.8	99.9
	7	54.1	28.0	2.1	< 0.1	9.4	93.6
	10	34.9	53.9	3.7	< 0.1	11.4	103.9
	14	17.3	73.4	2.4	< 0.1	7.3	100.4
	21	10.2	83.8	2.5	< 0.1	4.8	101.3
	28	6.9	92.1	2.0	< 0.1	3.8	104.8
	56	4.8	93.6	1.0	< 0.1	3.2	102.6
	84	4.3	92.8	0.2	0.1	3.0	100.4
	100	3.8	91.2	0.4	0.1	2.9	98.4

The degradation rate of dodine (DT_{50} and DT_{90}) in the two soils was calculated from both TLC and HPLC results, using the KIM computer program. First order DT_{50} values of 8.5-9.5 days and DT_{90} values of 30 days were estimated in both sandy loam soils, respectively (Table B.8 3).

Table B.8 3 - Degradation rate of 14C-dodine in soil under aerobic conditions when applied at a rate equivalent to 4.4 kg dodine/ha (4.4 mg dodine/kg dry soil)

Soil type	DT (days)	DT_{90} (days)	correlation coefficient r^2
Sandy loam (soil 1)	9.5	30	0.9743
Sandy loam (soil 2)	8.5	30	0.9275

^{14}C-dodine: Rate and route of degradation in three soil types under aerobic conditions with ^{14}C- guanidine labelled dodine and in one soil under aerobic conditions with ^{14}C-chain labelled dodine

Lowden P. et al. (1997).

Guidelines and GLP

The study was conducted according to Dutch Guidelines (CTB), Part G.1.1 (1991) Principles of GLP were complied with.

Material and methods

This study was performed to examine the rate and route of the aerobic soil metabolism of a)^{14}C-guanidine labelled dodine: batch 960806 (supplied by ARC/USA), radiochemical purity: >99.0%, with specific activity of 28.2 mCi mmol^{-1}; b)^{14}C-chain labelled dodine: batch GXR427E (supplied by Rhone-Poulenc UK), radiochemical purity: 95.2% with specific activity of 14 mCi mmol^{-1} and c) cold material: batch EA840SD2 (supplied by Nova), purity of 100%, at 20 ± 1 °C, under dark conditions, in a sandy silt loam, a clay loam and in a sandy soils, the first two from Essex and the other one from Sulffolk, UK. The soils characteristics are summarized in Table B.8 4.

Samples of soil (approximately 100g oven dried equivalent) were placed in uniquely labelled soil flasks and were treated with ^{14}C-guanidine labelled dodine solution at a rate equivalent to 2.63 kg/ha (3x the maximum field application rate of 900 g dodine/ha assuming 100% soil interception which corresponds to 2.63 mg dodine/kg dry soil). The studies followed for 120 days at which time the route of degradation of the compound (and the formation and degradation of metabolites) was established. The moisture content was adjusted to a tension value of pF2.5 and maintained at that level throughout the study. In addition, the metabolism of ^{14}C-chain labelled dodine was followed in one soil (clay loam) under the conditions already described and for the time as the ^{14}C-guanidine labelled dodine experiments. The non-radiolablelled dodine was used to treat the soil from which the microbial biomass was determined at the end of the study.

The incubation was carried out under aerobic conditions, in the dark at 20ºC. Double samples were analysed at 0, 1, 3, 5, 7,10, 14 ± 1 day, 28 ± 1 day, 62 ± 4 days and 120 ± 4 days.

The flasks were incorporated into a closed system in which moist, CO_2 free air was continuously passed over the soils. The evolved CO_2 was trapped in potassium hydroxide. After treatment the soils were extracted with solvent (basified methanol followed by acidified methanol), at intervals up to and including 120 days, and were analysed by HPLC. The unextracted radioactivity was assayed using a combustion technique. Selected extracts were examined by LC-MS.

Table B.8 4 - Soils used to investigate degradation and metabolism of dodine under aerobic conditions

Soil designation **Origin** **Soil type**	**Soil 3** **Essex/ UK** **Sandy silt loam**	**Soil 4** **Essex/ UK** **Clay loam**	**Soil 5** **Suffolk/UK** **Sand**
Textural analysis (ADAS) 2000 – 63 µm sand < 63 - 2 µm, silt <2 µm, clay	 45.5% 45.1% 9.3%	 23.7% 52.7% 23.7%	 90% 5.4% 4.6%
pH value Water CaCl2, 0.01M KCl, 1M	 6.6 6.0 5.7	 7.4 7.0 7.4	 6.7 6.3 6.3
Organic C (%)	1.2	2.1	2.3
Organic matter (%)	2.1	3.6	4.0
Cation exange capacity (meq/100g)	5.1	14.5	18.4
% Moisture content at pF value of 2.5	22.2	33.7	18.3
Microbial biomass (mg microbial carbon/kg soil) at beginning/end	185/107	461/268	280/288

Findings

The results obtained with the ^{14}C-labelled dodine, in terms of distribution of radioactivity and metabolites at different sampling dates are summarized in Table B.8 5.

Table B.8 5 - Recovery of applied radioactivity in % and distribution of metabolites after application of ^{14}C-dodine to soil under aerobic conditions at a rate of 2.6 kg dodine/ha (2.6 mg dodine/kg dry soil)

Soil	Days after applic.	Dodine (%)	$^{14}CO_2$ (%)	Metab.1 (%)	Metab.2 (%)	Metab.3 (%)	Metab.4 (%)	Bound Residues (%)	Total (%)
Soil 3 Essex/UK Sandy silt loam	0	95.6	n/a	0.9	nd	nd	nd	4.4	100.9
	1	88.4	4.3	0.8	2.0	nd	nd	7.4	102.9
	3	68.8	19.1	0.9	4.4	nd	nd	7.3	100.5
	5	48.5	30.0	nd	3.5	nd	nd	7.6	89.6
	7	34.1	60.8	0.3	2.4	nd	nd	6.1	103.7
	10	15.0	81.1	nd	1.7	nd	nd	4.5	102.3
	14	7.0	49.4	nd	1.2	nd	nd	3.6	61.2
	28	5.4	52.3	nd	0.9	nd	nd	3.1	61.7
	61	6.7	88.5	nd	1.0	nd	nd	3.9	100.1
	120	4.3	92.1	nd	1.0	0.1	nd	4.4	101.9
Soil 4 Essex/UK Clay loam	0	97.1	n/a	1.0	nd	nd	nd	7.4	105.5
	1	71.3	4.0	nd	1.9	0.9	nd	21.8	99.9
	3	51.4	36.6	nd	3.3	0.6	nd	11.9	103.8
	5	26.7	61.3	nd	4.0	nd	nd	10.8	102.8
	7	18.2	70.2	nd	2.4	nd	nd	9.3	100.1
	10	10.5	83.7	nd	1.3	nd	nd	9.3	104.8
	14	8.8	83.2	nd	0.9	nd	nd	6.9	99.8
	28	5.8	93.0	nd	0.8	nd	nd	5.3	104.9
	61	2.7	91.5	nd	0.2	nd	nd	2.8	97.2
	120	2.3	94.7	nd	nd	nd	nd	4.2	101.2
Soil 4* Essex/UK Clay loam	0	74.2	n/a	nd	2.8	nd	nd	21.8	98.8
	1	78.1	3.9	nd	5.5	nd	nd	7.9	95.4
	3	49.9	23.6	nd	4.5	0.4	nd	22.2	100.6
	5	30.0	38.8	nd	3.7	nd	nd	27.0	99.5
	7	15.6	52.5	nd	2.6	nd	nd	25.9	96.6
	10	9.8	56.1	nd	3.4	nd	1.4	25.2	95.9
	14	6.7	60.8	nd	2.2	nd	1.4	25.7	96.8
	28	4.4	67.1	nd	1.6	nd	1.2	23.3	97.6
	61	4.0	71.4	nd	0.7	nd	0.9	10.2	87.2
	120	2.1	81.4	nd	0.2	nd	0.3	17.2	101.2
Soil 5 Suffolk/UK Sand	0	94.8	n/a	1.2	nd	nd	nd	4.7	100.7
	1	88.8	6.2	nd	1.7	nd	nd	5.1	101.8
	3	71.0	25.8	nd	1.9	nd	nd	4.0	102.7
	5	50.9	48.1	nd	2.3	0.9	nd	4.4	106.6
	7	29.0	70.4	nd	1.5	nd	nd	3.8	104.7
	10	14.7	84.7	nd	0.5	nd	nd	3.0	102.9
	14	7.0	92.8	nd	nd	nd	nd	2.3	102.1
	28	3.5	93.7	nd	nd	nd	nd	3.0	100.2
	61	2.2	98.0	nd	0.03	0.02	nd	1.7	101.9
	120	1.5	95.4	nd	nd	nd	nd	1.9	98.8

* ^{14}C-chain labelled dodine

Recoveries of applied radioactivity ranged between 90-110% in the study for both labelled dodine fractions.

In all experiments dodine was rapidly degraded and the major metabolite detected was CO_2 which accounted for approximately 98% of AR in the sand, 95% in sandy silt loam and 92% in the clay loam soils treated with [^{14}C]-guanidine labelled dodine and 81% in the clay loam soil treated with [^{14}C]-chain labelled dodine, at the end of the study. The percentage of material in the solvent extract

fell with time from over 90% at day 0 down to less than 5% at day 120. This fall was not concomitant with the increase of unextractable residues, but matched the decrease in radioactivity associated with the soil as the compound was mineralised to carbon dioxide. The behaviour of [^{14}C]-guanidine labelled and [^{14}C]-chain labelled dodine in the clay loam soil was slightly different, since for guanidine labelled a significant decrees of unextractables during the study was observed, in which the maximum of 21.8% was observed in the first day with decrease to levels below 5%. For [^{14}C]-chain labelled dodine the decrease of unextractable was slower, the maximum of 27.0% was observed at day 5 and at the end of the study the level was 17.2%.

During HPLC analysis the non-volatile compounds detected in the soil extracts comprised dodine plus some minor metabolites. Spectrometric examination of the extracts confirmed the presence of parent material. A polar metabolite was detected by HPLC in the extracts at day 10 through to the final sampling point in the chain labelled experiment. The metabolites combined never accounted for as much as 10% of applied material and at the end of the study accounted for less than 1.5%.

The first order DT_{50} for dodine was calculated from HPLC results. It was found to be 4 days for the sand and sandy silt loam and 3 days for the clay loam in the [^{14}C]-guanidine labelled dodine experiment and 4 days for the clay loam in the [^{14}C]-chain labelled dodine experiment. The DT_{90} was found to be 15 days in the sand, 16 days in the sandy silt loam and 11 and 12 days in the clay loam in the [14C]-guanidine and [^{14}C]-chain labelled dodine experiments respectively (see Table B.8 6).

Table B.8 6 - Degradation rate of 14C-dodine in soil under aerobic conditions when applied at a rate of 2.6 kg dodine/ha (2.6 mg dodine/kg dry soil)

Soil type	DT_{50} (days)	DT_{90} (days)	Correlation coefficient (r^2)
^{14}C-guanidine labelled dodine in:			
Sandy silt loam soil (soil 3)	4.3	13.1	0.987
Clay loam soil (soil 4)	2.7	9.9	0.990
Sand (soil 5)	4.4	12.9	0.985
^{14}C-chain labelled dodine in:			
Clay loam soil (soil 4)	2.8	9.5	0.987

Degradation of dodine in Soil at 10ºC

Not available but since degradation occurs very rapidly in soils at 20ºC in the lab, it is not expected that the degradation of dodine will be of concern at 10ºC.

B.8.1.2 Supplementary studies

B.8.1.2.1 Anaerobic degradation in Soil

Anaerobic aquatic metabolism of Metasol DGH

Cady C. (1993)

Guidelines and GLP

The study was conducted according to US EPA FIFRA N-162-3, 40 CFR, section 158.130. Principles of GLP were complied with.

Materials and methods

Test radiolabelled material, ^{14}C-guanidine dodine (batch 2729-104 (supplied by Dupont/USA), radiochemical purity: 98.5%, specific activity of 27.69 mCi/mmol) was applied to a sandy loam soil Nebraska (USA). The soil characteristics are summarized in Table B.8 7. The metabolism of ^{14}C-dodine hydrochloride (dodine HCl) applied at a rate equivalent to 11.6 kg/ha (13x the maximum field application rate of 900 g dodine/ha assuming 100% soil interception which corresponds to 11.6 mg dodine HCI/kg dry soil) was followed for 1 year in flooded soil with well water. The incubation was carried out under anaerobic conditions, in the dark and 25°C. Prior to application the soil samples were incubated for 46 days in the dark under anaerobic conditions (confirmed by determination of the redox potential). The metabolism flasks were dosed by injecting the required amount of labelled dodine HCI into the culture tubes which were then shaken manually for a few seconds to homogenize.

Soil and water samples were analysed at 0, 1, 3, 7, 14 and 30 days and at 2, 3, 4, 6, 9 and 12 months after application of ^{14}C-DGH (dodine hydrochloride). Additionally, volatile degradates were sampled at 5, 7, 8, 10 and 11 months. After treatment the soils were extracted with solvent (basified methanol followed by acidified methanol) and the extracts were analysed by TLC (as confirmatory analytical technique) and HPLC. The unextracted radioactivity was assayed using a LSC technique

Table B.8 7 - Soil used to investigate degradation and metabolism of dodine under anaerobic conditions

Soil designation **Origin** **Soil type**	**Soil 6** **Nebraska/ UK** **Sandy loam**
Textural analysis (%) Sand silt clay	 56 26 18
pH value	6.8
Organic matter (%)	1.6
Cation exange capacity (meq/100cc)	11.6
Bulk density (g /ml)	1.51
% Moisture at field capacity (at 1/3 bar)	17.37
Microbial activity (colony-forming units/g) day 0 month 12	 1.1×10^6 3.5×10^5

Findings

The results obtained with the ^{14}C-labelled dodine HCl, in terms of distribution of radioactivity and metabolites at different sampling dates are summarized in Table B.8 8.

Table B.8 8 - Recovery of applied radioactivity in % after application of 14C-dodine HCl to soil under anaerobic conditions at a rate of 11.6 kg dodine HCl/ha (11.6 mg dodine HCl/kg dry soil)

Soil	Days (d)/ Months (m) After application	Water phase* (%)	$^{14}CO_2$ (%)	Extractable* (%)	Bound residues (%)	Total (%)
Soil 6 Nebraska/UK Sandy loam	0d	6.79	n/a	91.44	1.78	100.0
	1d	4.87	0	94.36	3.09	102.3
	3d	2.25	0	97.08	3.18	102.5
	7d	2.05	0.01	95.46	3.18	100.7
	14d	7.09	0.01	90.55	3.71	101.3
	30d	2.62	0.02	91.48	10.75	94.8
	2m	0.85	0.02	94.92	8.94	104.7
	3m	1.23	0.03	97.2	9.01	107.5
	4m	1.06	0.02	89.2	9.37	99.6
	6m	0.82	0.05	91.4	9.73	102.1
	9m	0.83	0.08	88.27	11.22	100.6
	12m	0.56	0.09	82.79	11.74	95.7

* 97% parent compound

Recoveries of applied radioactivity were generally good throughout the study with all of the individual results at each time point falling within the range 95-108%.

The sum of residues in the soil extracts and water layer dropped from 98.2 % AR at day 0 to 83.4 % after 12 months. The non-extractable ^{14}C-residues rose steadily from 1.78% AR at day 0 to 11.7% at 12 months.

The population of anaerobic microbes was indicative of an active soil/water system. The non-extractable ^{14}C-residues increased steadily from 1.78% AR at day 0 to 11.7% (0.88% in the humic acid fraction, 0.3% in the fulvic acid fraction and 8.69% in the humin phase) at 12 months, indicating that DGH residues are strongly bound into the soil matrix during anaerobic ageing. Cumulative volatiles reached 0.09% AR after 12 months and were identified as $^{14}CO_2$, HPLC analysis of the soil and water extracts showed that approx. 97% of the radioactivity after 12 months was the parent compound.

A polar metabolite (hydroxylated derivative of DGH) was present in the water layer at a concentration of approximately 0. 07µg/g (0.43%), and accounted for 75% of the activity in the aqueous phase at 12 months. For the whole system, the maximum concentration of this metabolite was 2.89% AR at 12 months.

The first order DT_{50} of DGH in the Nebraska sandy loam soil, under anaerobic conditions, calculated from the HPLC analysis of extractable ^{14}C-residues, was 2492 days (although the slope of the degradation curve was very low, it was statistically determined to be significantly different from zero). Therefore, ^{14}C-DGH is expected to be stable and resistant to breakdown in anaerobic conditions.

Table B.8 9 - Degradation rate of 14C-dodine in soil under anaerobic conditions

Soil type	DT50 (days)
^{14}C-guanidine labelled dodine in:	
Sandy loam soil (soil 6)	2492

Note: correlation coefficient not calculated in the original report

B.8.1.2.2 Soil photolysis (Annex IIA 7.1.1.1.2)

Dodine: photodegradation on soil

Mislankar S.G. (2001)

Guidelines and GLP

The study was conducted according to US EPA N 161-3 and EC Directive 95/36/EC 7.1.1.1.2. Principles of GLP were complied with.

Materials and methods

The test materials ^{14}C-gaunidine labelled dodine (batch nº of CFQ11834, radiochemical purity: >95.0%with specific activity of 118.8uCi/mg) and cold material (batch EA840SD2 (supplied by Nova), purity 99.5%) was applied to a sandy loam soil from California / USA. The soil characteristics are summarized in Table B.8 10. Soil samples (20 g dry weight) were treated by distributing the test substance over the surface at a rate of 4 ppm (4 mg dodine/kg dry soil) corresponding to 4 kg as/ha.

Table B.8 10 - Soil used to investigate photodegradation of dodine

Soil designation **Origin** **Soil type**	**Soil 7** **California/ US** **Sandy loam**
Textural analysis (%) sand silt clay	 59.6 29.2 11.2
pH value Water CaCl2, 0.01M KCl, 1M	 6.2 5.8 5.4
Organic matter (%)	0.44
Cation exange capacity (meq/100cc)	3.44
Bulk density (g /ml)	1.58
WHC at 1/3 bar (%) WHC at 15 bar (%) WHC saturation (%)	7.46 2.45 15.57

The samples were exposed to simulated sunlight irradiation (λ > 290 nm; Suntest CPS+ with Xenon burner and appropriate filters) for 12 hours (followed by 12 hours darkness) at 25 ± 1°C. The control units were incubated in the dark at 25 ± 1 °C. The intensity of the light source was recorded every hour for 30 days. An average intensity of the light received per day was 5850 w/m^2 which was equivalent to the maximum sunlight intensity on a hot summer day at latitude 50 ° N.

Samples from dark and irradiated units were analysed at 0 hours, 1, 3, 7, 14 and 30 days of exposure samples in duplicate were taken.

After extraction and reconstitution upon evaporation, the samples were chromatographed (HPLC/RAD), extracted soil samples underwent combustion (Harvey Oxidiser); radioactivity was determined by LSC and the confirmation of identity of parent compound was conducted by HPLC/MS.

Findings

The results obtained with the ^{14}C-labelled dodine, in terms of distribution of radioactivity and metabolites at different sampling dates are summarized in Table B.8 11.

Table B.8 11 - Recovery of applied radioactivity (%) in irradiated soil samples after application of 14C-dodine at a rate of 4 kg dodine/ha (4 mg dodine/kg dry soil)

Unit	Days after applic.	Dodine (% of extracted)	Met 1* (% of extracted)	Met 2 (% of extracted)	Met 3 (% of extracted)	Extracted	$^{14}CO_2$	Bound residues (% of IMD)	Total (% of IMD)
Irradiated	0	88.47	8.78	1.42	1.32	101.66	n/a	3.46	105.12
	1	86.63	10.60	1.61	2.15	95.19	0.01	4.78	99.98
	3	87.16	9.36	1.76	2.69	99.21	1.13	3.89	104.23
	7	87.28	8.04	1.45	3.22	91.30	2.06	4.23	97.59
	14	94.17	-	1.62	3.20	90.77	4.50	4.84	100.11
	30	85.68	6.09	1.72	4.40	83.30	14.39	5.65	103.32
Dark	0	88.90	8.73	1.28	1.09	99.28	n/a	3.37	102.65
	1	90.99	6.96	-	2.62	94.18	1.11	5.69	100.98
	3	90.37	4.96	0.98	3.81	91.92	1.94	6.58	97.44
	7	92.14	0.54	4.61	4.73	89.36	6.07	3.79	99.22
	14	93.11	-	-	5.52	85.68	7.81	3.97	97.46
	30	92.02	1.00	-	5.74	86.52	11.68	4.92	103.12

* contained multiple pecks

Overall recovery was in the range of 97 to 105% AR for both irradiated and dark control samples/units. In the soil extracts, recovery went down from initially 101.6% to 83.3 % for irradiated samples, and from 99.28 to 86.5% AR in dark samples over the period of 30 days of exposure. By the same time, $^{14}CO_2$ production increased to 14% in irradiated and to 12% in dark samples. The recovery in unextracted soil residue remained stable at approx. 3 to 6% for both incubation units.

The levels of parent compound recovered from the irradiated samples declined very slightly over the 30-days exposure period from 88% to 86% whereas it increased slightly in the control sample from 89% to 92%.

There was one fraction (metabolite 1) which accounted for approx. 10% of the applied radioactivity on day 1 for the irradiated sample. This fraction appeared to be a major metabolite, however, after isolation and reanalysis, under different HPLC conditions, it was shown that the fraction contained multiple peaks.

The first order kinetic model was used to estimate the half-life of dodine. The following half-life values were determined for both unit groups:

Table B.8 12 - Degradation rate of 14C-dodine in soil under irradiated and dark conditions

Soil type	Treatment Samples	DT50 (days)	correlation coefficient
^{14}C-guanidine labelled dodine in:			
Sandy loam (soil 7)	Irradiated	96	0.9975
	Dark control	130	0.9978

The degradation of dodine was similar in both the irradiated and dark control samples (see Table B.8 12, it was presumed that the relatively slow degradation of dodine under photolytic conditions compared to aerobic soil metabolism (with median DT_{50} of 4.3 days) was due to the low water holding capacity of the soil used and also the design of the photolysis study.

B.8.1.3 Field studies (Annex IIA 7.1.1.2.2; Annex IIIA 9.1.1.2)

B.8.1.3.1 Soil dissipation testing

Terrestrial Soil Dissipation after application of SYLLIT BRAND 65W fruit fungicide to bare ground plots simulating an orchard or grove.

Norris F.A. (1999)

Guidelines and GLP

The study was conducted according to US EPA FIFRA 164-1. Principles of GLP were complied with.

Materials and methods

The active substance, formulated as a 65% WP (batch 09JK0107-3, analysed content: 63.4%) was applied in four different soils, located in USA (California, Georgia, New Jersey and Washington). The soils characteristics are summarized in Table B.8-13. The degradation of dodine was studied following 6 applications of 2.18 kg dodine/ha (2.5x the maximum field application rate of 900 g dodine/ha assuming 100% soil interception which corresponds to 2.18 mg dodine/ka dry soil) with 7 days intervals between applications, for 1.5 year after the last treatment.

Dodine, formulated as a 65% WP, was applied to fallow (bare) soil (4 subplots per location). Soils were irrigated in order to maintain at least the historical average rainfall for each site. Effective precipitation between sampling intervals was recorded and verified that sufficient soil moisture was present to aid in microbial degradation, as an indicator of the vertical movement in the soil. Soil samples were collected to a depth of 0.9 m in 0.15 m increments, frozen at the field site and sent to the laboratory for storage (up to 1-17 months before analysis). A 12-month storage stability study showed that there were no appreciable losses of dodine during the freezer storage. Analysis occurred with GC-MSD after derivatization with hexafluoroacetylacetone. Since soil metabolism studies performed in the laboratory showed that the parent compound dodine dissipates without significant metabolites being formed, the analysis of soil samples for dodine (with a LOQ of 0.01 ppm) was deemed adequate to determine the magnitude and distribution of residues of dodine in the field.

Table B.8 13 - Soils used to investigate the degradation of dodine under field conditions

Soil designation Origin Soi Type (SCS soil classification)	Soil 8 Washinton /USA Sand (Quincy loamy fine sand)	Soil 9 New Jersey/USA Loam (Penn)	Soil 10 Georgia/USA (Dothan Loamy sand)	Soil 11 California/USA (Atwater loamy sand)
Climate, Summer	Warm/dry	Warm/ humid	Hot/humid	Hot/dry
Climate, Winter	Cool/dry	Cool / wet	Mild/ wet	Cool /dry
Irrigation	Necessary	As needed	As needed	Necessary
US Textural Analysis (%) sand silt clay	 89-93 4-8 3	 29-55 23-47 20-26	 65-87 7-12 3-26	 85-89 4-10 5-7
pH value (depth)	6.7 (15 cm) 6.6 (30 cm) 6.3 (45cm) 6.6 (60 cm) 6.7 (75cm) 6.7 (90cm)	6.0 (15 cm) 6.1 (30 cm) 6.2 (45cm) 6.3 (60 cm) 6.2 (75cm) 5.6 (90cm)	7.1-7.4 (15 cm) 6.4-6.6 (30 cm) 5.4-5.6 (45cm) 5.0 (60 cm) 5.4 (75cm) 5.4-5.5 (90cm)	7.1 (15 cm) 6.8 (30 cm) 7.2 (45cm) 7.9 (60 cm) 8.2 (75cm) 8.2 (90cm)
Organic Matter % (depth)	1.2 (15 cm) 0.9 (30 cm) 0.6 (45cm) 0.4 (60 cm) 0.4 (75cm) 0.2 (90cm)	2.3 (15 cm) 1.3 (30 cm) 0.4 (45cm) 0.2 (60 cm) 0.2 (75cm) 0.2 (90cm)	1.0-1.5 (15 cm) 0.7-0.9 (30 cm) 0.3-0.4 (45cm) 0.2-0.6 (60 cm) 0.3-0.4 (75cm) 0.1-0.5 (90cm)	0.5 (15 cm) 0.4 (30 cm) 0.2 (45cm) 0.2 (60 cm) 0.1 (75cm) 0.1 (90cm)
Cation exchange capacity (meq/100 g)	8.7-9.7	8.6-9.8	3.4-6.4	5.5-6.9
Bulk density (g/ml)	1.34-1.54	1.1-1.24	1.11-1.32	1.37-1.50
Field capacity (depth)	17.1 (15 cm) 16.2 (30 cm) 18.2 (45cm) 16.7 (60 cm) 15.4 (75cm) 13.3 (90cm)	25.3 (15 cm) 24.4 (30 cm) 24.1 (45cm) 21.9 (60 cm) 20.5 (75cm) 20.3 (90cm)	5.4-5.5 (15 cm) 5.3-5.7 (30 cm) 5.7-7.2 (45cm) 8.8-9.7 (60 cm) 10.7-11.8 (75cm) 12.4-14.4 (90cm)	6.3 (15 cm) 5.2 (30 cm) 4.9 (45cm) 6.1 (60 cm) 5.9 (75cm) 6.8 (90cm)

Findings

There were no significant residues (>0.01 ppm) of dodine below 15 cm in the soil profile at any location despite precipitation / irrigation averaging at least 95 mm a month. There were three sporadic trace levels (0.02 - 0.03 ppm) in deeper segments which were not supported by companion samples and were not considered significant as indicators of soil mobility. In the 4 soils, the residue declined rapidly during the first month after the last application, then dissipated more slowly (see Table B.8 14).

Table B.8 14 - Distribution of dodine residues after 6 applications, each of 2.18 kg/ha, of dodine formulated as 65% WP (ppm)

Soil designation	Depth (cm)	Months post application 6										
		0.25	0.5	1	2	3	6	8	10	12	15	18

		Months post application 6										
Soil 8 Washington/USA Sand	15	0.94	0.98	0.83	0.44	0.37	0.54	0.02	na	0.01	<LOQ	<LOQ
	30	<LOQ	<LOQ	<LOQ	<LOQ	nd	nd	<LOQ	na	<LOQ	<LOQ	<LOQ
	45	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	na	<LOQ	<LOQ	<LOQ
	60	na	na	na	na	na	<LOQ	<LOQ	na	<LOQ	na	na
	75	na	na	na	na	na	na	na	na	na	na	na
	90	na	na	na	na	na	na	na	na	na	na	na
Soil 9 New Jersey/USA Loam	15	0.53	0.31	0.16	0.16	0.13	0.18	0.09	0.04	na	0.02	0.02
	30	0.03*	<LOQ	<LOQ	<LOQ	nd	nd	nd	<LOQ	na	nd	<LOQ
	45	<LOQ	<LOQ	<LOQ	<LOQ	nd	nd	nd	nd	na	nd	nd
	60	na	na	na	<LOQ	nd	nd	nd	na	<LOQ	na	na
	75	na	na	na	na	na	na	na	na	na	na	na
	90	na	na	na	na	na	na	na	na	na	na	na
Soil 10 Georgia/USA Loamy sand	15	1.15	0.47	0.47	0.20	0.17	0.06	0.06	0.09	na	0.07	0.13
	30	nd	<LOQ	nd	nd	nd	nd	nd	nd	na	nd	<LOQ
	45	nd	nd	nd	nd	nd	nd	nd	nd	na	nd	nd
	60	na	na	na	na	nd	nd	nd	nd	<LOQ	na	na
	75	na	na	na	na	na	0.03*	na	na	na	na	na
	90	na	na	na	na	na	na	na	na	na	na	na
Soil 11 California/USA Loamy sand	15	2.53	1.63	0.54	0.09	0.08	0.04	0.02	0.02	na	<LOQ	<LOQ
	30	<LOQ	0.02*	nd	nd	nd	nd	nd	nd	na	nd	<LOQ
	45	nd	nd	nd	nd	nd	nd	nd	nd	na	<LOQ	<LOQ
	60	nd	na	na	na	nd	nd	nd	na	nd	na	na
	75	nd	na	na	na	na	na	na	na	na	na	na
	90	nd	na	na	na	na	na	na	na	na	na	na

na: not analyzed

nd: not detected

The determination of the half-life of a.s. at each test site was based on first order exponential curves of the results of the analyses after each application. Dissipation times are presented in the Table below.

Table B.8 15 - Dissipation time of dodine in soil under field conditions after 6 applications of each 2.18 kg as/ha.

Soil type	DT_{50}	DT_{90}	r^2 (1st order)
Sand (soil 8)	13.0	108.3	0.81
Loam (Soil 9)	6.7	22.1	0.75
Loamy sand (soil 10)	18.6	61.2	0.81
loamy sand (soil 11)	14.7	48.7	0.91

B.8.1.3.2 Soil residue testing

Not applicable since the degradation in soil is rapid and no succeeding crop is planted after application. Orchards stay in place for many years.

B.8.1.3.3 Soil accumulation testing

No data submitted. Since the estimated DT_{90} in the field < 1 year. Degradation in soil is rapid.

B.8.1.4 Summary of the route and rate of degradation in soil

Aerobic route and rate of degradation

The route of degradation of dodine in the laboratory, under aerobic conditions was assessed in a sandy loam soil at 25 ºC (Cooper. *et al.*, 1996), and in a sandy silt loam, clay loam and sand soil at 20 ºC (Lowden P *et al*, 1997).

The results of the experiment conducted with the guanidine labelled dodine show that the a.s. is quickly metabolised in soil and its degradation ultimately resulted in a formation of CO_2 without formation of any major metabolite or persistent unextractable residues.

Degradation of dodine occurred by fragmentation of the molecule in three parts: dodecane, guanidine and acetic acid. The guanidine and the dodecane chain should be rapidly used by soil microflora. Minor metabolites were detected, but not identified, one of the metabolite should be guanidine that ultimately degrades to CO_2. The minor metabolites should be molecules in which the chain length had been reduced (by β-oxidation) or to which additions have been made (e.g. hydroxidation). Both dodine+OH and dodine+CH2 were identified (by mass spectrometry) as minor synthetic impurities in the test material.

The results of the experiment conducted with chain labelled dodine show that the dodecyl moiety of the molecule was also ultimately degraded to carbon dioxide. Incorporation of the partially degraded chain resulted in disappearance of parent material at a similar rate to that seen in the guanidine labelled experiments but with a slightly delayed evolution of radiolabelled carbon dioxide. Intermediate metabolites were produced in very small quantities and were themselves degraded without a build up of residues remaining associated with the soil.

A proposed metabolic pathway based on the results of both guanidine and chain experiments are presented as Figure B.8 - 1.

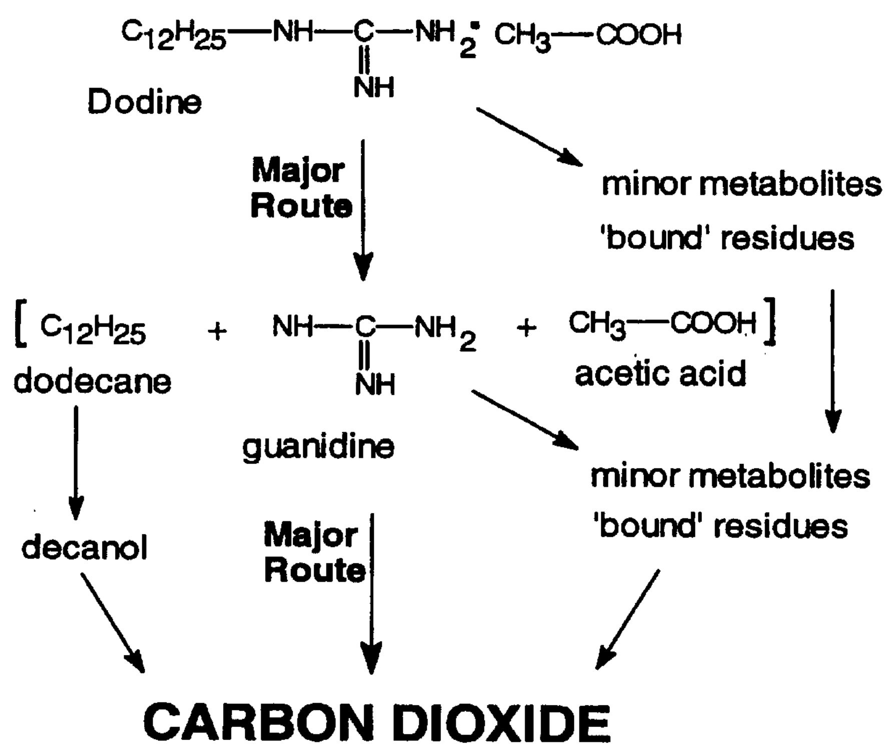

Figure B.8 1 – Proposed degradation pathway of dodine in aerobic soil

The major degradation product was CO_2 reaching 91.2 - 95.4% for guanidine labelled dodine at the end of the study and 81.4% AR for labelled chain dodine.

The bound residues remained at low level, with a maximum of 11.4% of AR during the first 10 days and values lower than 4.2% were registered at the end of the study, for all soils and for guanidine labelled dodine. Higher values of bound residues were registered in chain labelled dodine, the maximum value was 27% of AR at day 5, decreasing to 17.2% at the end of the study.

No major metabolites were identified in both studies. During HPLC and TLC (Cooper *et al*, 1996), dodine plus a cluster of minor metabolites were detected in soil extracts. More than 11 compounds were detected, the sum of those metabolites decreased from 9.7% AR at the beginning to 0.6% AR at the end of the study. A polar metabolite (M7) was detected in the extracts at the last two time-points, with max. of 3% AR. Four metabolites were detected by HPLC in the study conducted by Lowden (1997), these metabolites (all minor) could not be positively identified by use of mass spectrometry but free guanidine was tentatively assigned as one of them.

In the guanidine labelled experiments there was no metabolite that reached 5% AR at any time and the total of metabolites at the end of the study accounted for less than 1.5% of applied material. In the chain labelled experiment one metabolite reached a level of 5.5% of applied material after one day but thereafter the level decreased and it was not detectable at the end of the study at which time extractable metabolites accounted for only 0.3% of applied radioactivity.

The rate of dodine degradation in soil is slightly dependent on pH and moisture content on degradation is not influenced by the soil type or tested concentration.

The route of degradation of dodine in laboratory, under aerobic conditions was assessed in sandy loam soil at 25 ºC (Cooper. *et al.*, 1996), and in sandy silt loam, clay loam and sand soil at 20 ºC (Lowden P *et al*, 1997).

The DT_{50} (1[st] order) for dodine, at 20ºC, ranged between 2.8 and 6.02 days both C-guanidine and C-chain dodine. Detailed DT_{50} and DT_{90} in soils under aerobic conditions had shown Table B.8 16.

Table B.8 16 - Rates of degradation of dodine in soil under laboratory conditions

Soil	Texture class	pH in water	Aerobic or anaerobic	Temp. (ºC)	DT50 (days)	DT90 (days)	DT_{50}(d) 20ºC pF2.5/10kPa	r^2 1st order	Ref.
^{14}C-guanidine labelled dodine in:									
UK Essex (soil 1)	Sandy loam	5.3	aerobic	25*	6.2	28.0	6.0	0.988	Cooper (1996)
US Mississippi (soil 2)	Sandy loam	5.9	aerobic	25*	5.6	22.6	5.7	0.972	Lowden (1997)
UK Essex (soil 3)	Sandy silt loam	6.6	aerobic	20	4.3	13.1	4.3	0.987	
UK Essex (soil 4)	Clay loam	7.4	aerobic	20	2.7	9.9	2.7	0.990	
UK Suffolk (soil 5)	Sand	6.7	aerobic	20	4.4	12.9	4.4	0.985	
^{14}C-chain labelled dodine in:									
UK Essex (soil 4)	Clay loam soil	7.4	aerobic	20	2.8	9.5	2.8	0.987	Lowden (1997)
Geometric mean							4.3		

Water holding capacity of 75% of 1/3 bar

Anaerobic route of degradation

The anaerobic route of degradation of dodine HCl was studied by Caddy (1993), in a sandy loam soil from Nebraska (UK), under dark, at 25ºC, on anaerobic incubated flooded soil. The degradation of dodine was extremely slow with a calculated DT_{50} of 2492 days (see Table B.8 17).

The mineralisation of dodine under anaerobic conditions was very slow, reaching the maximum level of 0.09% AR after 12 months. An polar metabolite, identified as a hydroxylated, derivative of the parent compound reached a concentration of 2.89% AR after 12 months. The bound residues increased steadily from 2 % AR at day 0 to 12% at 12 month. Approximately 9% of the 12% where associated with the humin phase, indicating that dodine residues are strongly bound into soil matrix.

Table B.8 17 - Rates of degradation of dodine in soil under anaerobic conditions

Soil	Texture class	pH in water	Aerobic or anaerobic	Temp. (ºC)	DT50 (days)	DT90 (days)	DT_{50}(d) 20ºC pF2/10kPa	r^2 1st order	Ref.
^{14}C-guanidine labelled dodine in:									
UK Nebraska (soil 6)	Sandy loam	6.8	anaerobic	25	2492	--	2523	0.3	Cady (1993)

Soil photolysis

The photolysis of ^{14}C-guanidine labelled dodine was studied, by Mislankar S.G. (2001), in sandy loam soil from California/US, under artificial light during 12.12 LOD, at 25ºC. The control was incubated at the same temperature, but in dark. The average intensity of light source per day (5850 w/m^2) was considered equivalent to the maximum sunlight intensity on hot summer day at latitude 50ºN.

The DT_{50} was calculated to be 96 days in irradiated conditions and 130 days in the darkness. Three minor photodegradation products were detected, but not identified, they were named as Met 1, 2 and 3. The bound residues never exceeded more than 6% of the applied radioactivity. The values of CO_2 at the end of the study in light and dark samples were similar (max. of 14% AR). Photolysis may play a very small role in the dissipation of the active substance.

Field studies – soil dissipation

The degradation of dodine was studied in five USA soils, in one sandy (Washington), other loam (new Jersey) and two sandy loam soils (Georgia and California). At each site dodine was applied to bare soil, 6 times, at a rate of 2.18 kg a.s./ha (2.5 x the maximum field application rate of 900 g a.s./ha) with 7 day intervals between the applications of a 65% WP formulation These field evidenced characterize dodine to be a fungicide with a short half-life, with a DT_{50} range between 6.7 and 18.6 days and DT_{90} lower than 365 days, indication low potential to accumulate in soil. That consideration is supported by the determination of soil residues during the 18 months of field study.

In four soils, in top horizon, the residue declined rapidly during the first month after the last application, then dissipated more slowly. No significant residues of dodine were detected below 15 cm in the soil profile, indicating low potential of dodine to leach. No degradation products were detected in the soil horizons (Norris F.A., 1999). Detailed DT50 (1st order) and DT90 in aerobic soils is shown in the Table B.8 18.

Table B.8 18 - Rates of degradation of dodine in soil under field conditions

Location	Soil type	pH	Formulation rate	DT_{50} (days)	DT_{90} (days)	r^2 1st order	Ref.
USA Washington (soil 8)	Sand (bare soil)	6.7	6x 2.18 kg a.s./ha (7 days interval)	13.0	108.3	0.81	Norris (1999)
USA New Jersey (soil 9)	Loam (bare soil)	6.0		6.7	22.1	0.75	
USA Georgia (soil 10)	Sandy loam (bare soil)	7.1-7.4		18.6	61.2	0.81	
USA California (soil 11)	Sandy loam (bare soil)	7.1		14.7	48.7	0.91	
Geometric mean				13.25	60.08		

B.8.1.5 Assessment of the route and rate of degradation in soil

The degradation of dodine was studied in aerobic and anaerobic conditions, in laboratory studies. The influence of irradiation in the degradation process of dodine was also investigated. Dodine was rapidly degradated under aerobic conditions, with DT_{50} (1^{st} order) values between 3 and 6 days. Under anaerobic conditions the degradation was significantly slow, with an estimated DT_{50} far greater than 1 year. Considering the results of dodine photodegradation, it can be considered that photolysis does not play a significant role for dodine degradation, since a DT_{50} of 96 days was estimated in the irradiated sample and a DT_{50} of 103 days was determined in the dark soil sample. These results are consistent with the conclusion that dodine degrades in the environment by microbial degradation.

Under field conditions a DT_{50} between 7 and 19 days and a DT_{90} lower than 90 days were estimated, with exception of the DT90 estimated in soil 8. Dodine was found to be not persistent in soil. Degradation of dodine should occur via microbial activity, mediated by oxidation to CO_2, with formation of multiple and smaller minor metabolites. No major metabolites were found in the laboratory or field studies. Indeed no metabolites reached more than 6% of RA. The major degradation product was CO_2, with a maximum of 95.4% for guanidine labelled dodine at the end of the study and 81.4 for labelled chain dodine. The bound residues remained at low levels with a maximum of 11.4% of AR during the first 10 days and values lower than 4.2% were registered at the end of the study, for all soils and for guanidine labelled dodine. Higher values of bound residues were registered in chain labelled dodine, the maximum value was 27% of AR at day 5, decreasing to 17.2% at the end of the study.

B.8.2 Adsorption, desorption and mobility in soil (Annex IIA 7.1.2 and 7.1.3; Annex IIIA 9.1.2)

B.8.2.1 Adsorption and desorption of the active substance and relevant metabolites (Annex IIA 7.1.2)

Soil/sediment adsorption-desorption of DGH.

Williams M, Hargadine S. (1991)

Guidelines and GLP

The study was conducted according to US EPA FIFRA N-163-1. Principles of GLP were complied with.

Materials and methods

The adsorption and desorption of radiolabelled dodine (DGH) (batch 2633-116 (supplied by Dupont/USA), radiochemical purity: 96.1 % and specific activity of 27.69 mCi/mmol), and cold material (batch B0051202, formulated as a solution in ethanol/water: purity: 35% DGH), was studied in four different soils in the USA. The soils characteristics are summarized in Table B.8 19.

A preliminary study was conducted at a soil to water ratio of 1:10 at a nominal concentration of 10 ppm. The mean percent of ^{14}C-test material adsorbed into the 4 test soils was higher than the expected range for the Freundlich model i.e. 20% - 80%. The soil and water ratio was then adjusted to 1:250 during the definitive study to fit the Freundlich model with a nominal concentration of 1.0,

0.75, 0.50 and 0.10 ppm. Aqueous solutions of the test substance were prepared with sterile 0.01 N $CaCl_2$. The study was run at 25°C in the dark according to the batch equilibrium method.

The samples were shaken during 72 hours (equilibrium time) after the test substance administration. Mass balance was performed in all soils. The stability of the test item was shown by means of TLC and the degree of adsorption of the test item to the containers was also checked.

Table B.8 19 - Soils used to investigate adsorption/desorption of dodine

Soil designation **Origin** **Soil type**	**# 87** **Sand**	**# 79** **Sandy loam**	**#86** **Clay loam**	**#90** **Silt loam**
US Textural analysis (%) sand silt clay	 92 4% 4%	 54% 36% 10%	 22% 40% 38%	 24% 52% 24%
pH value	7.6	6.5	6.4	7.4
Organic matter %	0.1	0.8	1.3	4.2
Cation exange capacity (meq/100g)	5.0	4.7	19.6	22.7
Bulk density (g/ml)	1.53	1.15	1.38	1.08
Field capacity at 1/3 bar (%)	1.92	9.49	28.33	35.92

Findings

The test solution (at 1 or 10 mg as/l in 0.01 N $CaCl_2$) was shown to have a radiochemical purity of 95.3% (TLC) and the stability of dodine HCl at this concentration was shown by means of TLC during at least 48 hours. The LSC analysis of the control (no soil) samples showed that no adsorption of DGH to the container (Silanized Pyrex and Nalgene bottle) occurred. The measured concentrations (n=3, LSC) were 77%, 74%, 73% and 67% of nominal for the 1, 0.75, 0.5 and 0.1 ppm dose level respectively. Mass balance ranged for the 4 soils between 98.5 and 103%. The mean percent ^{14}C-DGH adsorbed to the test soils after 72 hours for all concentrations was 62.1 %, 56.4%, 98.7% and 96.1 % for the #87 sand, #79 sandy loam, #86 clay loam and #90 silt loam, respectively. However, it was observed at the low concentration (0.1 ppm nominal) that DGH exhibited different sorption characteristics in #87 sand and #79 sandy loam soils with an adsorption of 25.7% and 28.6% respectively, this occurred only in the two soils having an organic matter content of less than 1 %.

After the desorption step, the mean percent ^{14}C-DGH desorbed from the test soils for all concentrations was 70.6%, 50.3%, 4.8% and 12.47% of the absorbed quantity for the #87 sand, #79 sandy loam, #86 clay loam and #90 silt loam, respectively. In line with the observations made during the adsorption step, it was observed at the low concentration (0.1 ppm nominal) that DGH exhibited different desorption characteristics in soils sand (#87) and sandy loam (#79) with a desorption of 116% and 97.6% respectively. Again, this occurred only in the two soils having an organic matter content of less than 1 %. The mean percent adsorbed for soils clay loam (#86) and silt loam (#90) was not in the 20 - 80% range.

However, the determined adsorption and desorption isotherms revealed correlation coefficients (of 0.8770, 0.9735, 0.9955 and 0.9962 for #87, #79, #86 and #90 soils, respectively) implying that the data adequately fits the Freundlich model. The results obtained are summarised in the Table below.

Table B.8 20 - Adsorption/desorption parameters of dodine HCl in soil.

Soil	Organic carbon (%)	Soil pH	Adsorption		Desorption		Mobility class
			Kd	Koc	Kd	Koc	
#87 sand	0.05	7.6	6440	$1.29x10^7$	294	$5.88x10^5$	Immobile
#79 sandy loam	0.40	6.5	2202	$5.51x10^5$	3095	$7.74x10^5$	Immobile
#86 clay loam	0.65	6.4	18019	$2.77x10^6$	2028	$3.12x10^5$	Immobile
#90 silt loam	2.10	7.4	15228	$7.25x10^5$	760	$3.62x10^4$	Immobile

B.8.2.2 Column leaching studies with the active substance and relevant metabolites (Annex IIA 7.1.3.1; Annex IIIA 9.1.2.1)

Column leaching studies with the active substance using 4 soils

No data submitted, Since reliable adsorption coefficients were obtained in the study under point 7.1.2/1 and field study performed under section B.8.2.1 revealed a very low potential for leaching.

Column leaching studies with the metabolites using 4 soils

No data submitted. Not applicable since no major metabolites or degradation products were detected at levels > 10% in soil studies.

B.8.2.3 Aged residue column leaching (Annex IIA 7.1.3.2; Annex IIIA 9.1.2.1)

Aged leaching of dodine

Van Noorloos B, Slangen P.J. (2002)

Guidelines and GLP

The study was conducted according to SETAC (Lynch, 1995) and OECD 'Leaching in soil columns' draft 12/2000. Principles of GLP were complied with.

Materials and methods

The leaching of aged dodine, ^{14}C-guanidine (batch nº 011009, radiochemical purity of >97.9% and specific activity of 25 mCi/mL) and the non-labelled dodine (batch nº S01106 and purity of 96.2%. The radiolabelled dodine) was studied in one sandy loam soil form UK (see Table B.8 21).

Ageing was undertaken in one standard soil, Cranfield 243, a sandy loam from UK, with 1.7% organic carbon and soil moisture content equivalent to 48% water holding capacity, at 20°C ± 2°C in the dark for 3.25 days (78 h). The application rate was 1.2 mg dodine/kg dry soil (equivalent to 900 g a.s./ha).

Aged residues were transferred to the top of a Cranfield 243 soil column with 5.0 cm i.d. x 40 cm. The aged residues were leached with 0.01 M $CaCl_2$, under unsaturated conditions (536 mL of leachate, i.e., 273 mm rainfall during 65 hours). The experiments were performed in duplicate and leachates collected in fractions. At the end of the leaching process, soil columns were subsectioned

and partitioned into extractable/unextractable fractions. The extractable fraction served to determine metabolites and a.s. by means of TLC.

Table B.8 21 - Soil used to investigate lixing behaviour of aged soil residues of dodine

Soil designation **Origin** **Soil type**	**Cranfield 243** **Rase/ UK** **Sandy loam**
US Textural analysis sand silt clay	 70.4% 16.4% 13.2%
pH value	5.9
Organic carbon %	1.7
Cation exange Capacity (meq/100g)	12.1
Water holding capacity (%)	52.4

Findings

The radiochemical purity of the spiked solution was determined as 99.1 %. The ageing process was terminated after 78h days of incubation when the content of dodine was down to 32.4%. Results of the ageing process are given in the following Table.

Table B.8 22 - Distribution of activity in Cranfield 243 soil during ageing of dodine (% of applied)

Time (hours)	**CO_2**	**Non-extractable**	**Extractable**	**% recovery dodine (TLC/HPLC)**	**Mass balance**
0	-	9.0	89.1	88.5/89.3	98.3
72	24.8	20.0	47.3	47.0/39.9	91.9
78	23.1	32.5	39.0	38.8/32.4	94.7

At the end of the ageing period (78h), the extractable fraction of the soil contained 39% of the applied activity, 99.6% of which was dodine according to TLC (83.1% of the extractable fraction, i.e. 32.4% of applied according to HPLC). No extractable metabolites > 1 % of AR were present in the aged soils (<5% according to HPLC). The mass balance was 92-98% of applied and thus within acceptable range. Extraction efficiency from the soil at t=0 was acceptable (89%).

Table B.8 23 - Recovery of activity in leachate fractions of the Cranfield 243 soil (replicate A and B)

Fraction	Mass of fraction (g)		Recovery (% of applied)	
	A	B	A	B
1	52.20	45.09	0.00	0.00
2	60.81	62.90	0.00	0.00
3	59.92	57.22	0.02	0.02
4	53.59	57.63	0.03	0.04
5	64.21	66.22	0.04	0.04
6	41.57	62.44	0.02	0.02
7	36.35	57.00	0.01	0.01
8	38.88	59.75	0.01	0.01
drain	2.01	3.84	0.00	0.00
Total	409.54	472.09	0.13	0.14

No significant amounts (< 0.2% of applied) of dodine aged residues were detected in the leacheate upon leaching under unsaturated flow conditions (see Table B.8 23).

The major part of the activity in the two columns at the end of the leaching process consisted of extractables (≥ 78% of total soil associated activity) was recovered in the top section of the column (0-14.2 cm). The extractable radioactivity was mainly dodine (31-32% of applied in column sections 1-2). 88%-95% of total soil associated activity was recovered in top section of the soil column. Little movement of activity down the soil column was observed. In both columns, the soil associated activity was only 1.7-5.2% of applied in section 2 (7.1 – 14.2 cm) and decreased to 0.1 % in the two lower sections. No significant metabolites were observed after leaching.

After leaching, the mass balance was 78.7% for column A and 73.2% for column B. The incomplete mass balance is most likely the result of incomplete trapping of' $^{14}CO_2$ just before leaching (during ageing period 24.4% and 25.4% for column A and B, respectively), during or after leaching (11.4% and 1.7% for column A and B, respectively), since the mass balances up to the start of leaching were >90%. This is supported by the observation that during ageing, dodine degraded quickly and almost solely to CO_2.

B.8.2.4 Lysimeter and field leaching studies (Annex IIA 7.1.3.3; Annex IIIA 9.1.2.2)

No data submitted.

B.8.2.4.2 Field Leaching Studies

No data submitted.

B.8.2.5 Summary of adsorption/desorption and mobility in soil

The adsorption and desorption characteristics of dodine were studied by Williams (1991) in a sand, a sandy loam, a clay loam and a silt loam soil, that covered a range of organic carbon contents between 0.05% and 2.10%, a range of pH from 6.4 to 7.6 and a clay content from 4% to 38%. The mass balance in the four soils ranged between 98.5 and 103%.

Adsorption K_{oc} values ranged from $5.51x10^5$ to $1.29x10^7$, the K_{oc} values for desorption were also high, ranging between $3.62x10^5$ and $5.88x10^{7,}$ indicating that once adsorbed to soil dodine has low

potential to leach into the groundwater. Hence dodine can be classified as immobile in soil. No influence of pH on soil sorption was observed from data available.

The leaching characteristics of dodine in aged soil were studied by Van Noorloos (2002) in one sandy loam soil, with organic matter content of 2.9%, pH 5.9 of and clay content of 13%. Dodine was applied at a rate of 1.2 mg/kg dry soil and aged under aerobic conditions at soil moisture content equivalent to 48% of water holding capacity, for 78 hours.

No significant amounts (< 0.2% of applied) of dodine aged residues were detected in leachates under unsaturated flow conditions. Most of the activity (88-95%) was found to remain in the top of the soil columns consisting mainly of dodine. Little movement of activity down the soil occurred. In the bottom section of the column no significant amounts of activity were found (0.1%). No significant metabolites were observed after leaching. It can be concluded that dodine show low potential to leach into groundwater.

B.8.2.6 Assessment of adsorption/desorption and mobility in soil

The adsoption/desorption characteristics of dodine were studied using a batch equilibrium method. Adsorption coefficients higher than 500000 were determined for tested soils. Dodine was classified as immobile in soil. This result is supported by a dodine column leaching study with aged residues, where dodine showed low potential to leach (see Table B.8 24).

Table B.8 24 - Adsorption/desorption of dodine in soil

Soil Type	OC %	pH	Kd (mL/g)	Koc (mL/g)	1/n
Sand	0.1	7.6	6440	$1.29x10^7$	0.8770
Sandy loam	0.8	6.5	2202	$5.51x10^5$	0.9735
Clay loam	1.3	6.4	18019	$2.77x10^6$	0.9955
Silt loam	4.2	7.4	15228	$7.25x10^5$	0.9962
Arithmetic mean/median			10472	$423.6x10^4$	
pH dependence, Yes or No			No		

B.8.3 Predicted environmental concentration in soil (PECs) Annex IIIA 9.1.3)

Dodine formulated as a 400g/l SC formulation (A.S. 400SC) will be applied five times, spaced by 7 days, at maximum rate of 0.9 kg dodine/ha. Since there were no major metabolites formed in soil, predicted environmental concentrations (PEC) were calculated for dodine only. The initial predicted environmental concentration in soil (PECs) is a worst case estimation based on the maximum rate being applied with canopy interceptions of 20%. For calculation purposes, it is assumed that the residues of dodine are uniformly distributed in the soil to a depth of 5 cm (bulk density 1.5 g/cm3). The initial PECS was calculated using the following formula:

$$PEC_s = A \times \frac{(1 - f_{int})}{(100 \times depth \times bd)}$$

where PECs = predicted environmental concentration of the a.s. in soil (mg/kg) immediately following a single application

A = application rate (g a.s./ha)

fint = fraction intercepted by crop canopy

depth = mixing depth (cm)

bd = bulk density (g/cm3)

Short-term and long-term PECS were calculated to estimate the concentration of dodine in soil using a time-weighted average (TWA) according to the following formula.

$$PEC_l = PEC_i \cdot \frac{DT_{50}}{t_1 \cdot \ln(2)} \left(1 - e^{(-t_1 \cdot \ln(2)/DT_{50})}\right)$$

where PEC_l = time-weighted average (TWA) concentration, PECi = initial concentration, and t_1 = time period. This PEC_l reflects the average concentration an organism would be exposed to within a given time period t_l.

Using the realistic worst-case DT_{50} of 18.6 days from field studies, the actual and TWA PEC_S values have been calculated for dodine. The calculated actual and TWA PECs values are presented in Table B.8 25.

Table B.8 25 - Actual PECs values and time-weighted average concentrations for dodine calculated using the first-order DT50 value of 18.6 days

$PEC_{(s)}$ (mg/kg)	Single application Actual	Single application Time weighted average	Multiple application Actual	Multiple application Time weighted average
Initial	0.96		3.04	
Short term 24h	0.940	0.923	2.98	2.928
2d	0.921	0.889	2.928	2.821
4d	0.889	0.825	2.823	2.618
Long term 7d	0.8429	0.738	2.675	2.341
28d	0.5945	0.337	1.887	1.070
50d	0.4342	0.149	1.378	0.472
100d	0.2508	$1.19x10^{-06}$	0.796	$7.32x10^{-02}$

B.8.4 Fate and behaviour in water (Annex IIA 7.2.1; Annex IIIA 9.2)

B.8.4.1 Hydrolysis rate of relevant metabolites, degradation and reaction products (Annex IIA 7.2.1.1) (as far as not covered by point 2.9)

Annex II point 7.2.1.1 requires a hydrolysis study to be conducted for relevant metabolites, degradation and reaction products, which account at any time for > 10% of the a.s. added unless sufficient information on the degradation is available for the test performed on the parent material, which is not the case, since metabolites, degradates or reaction products do not make up for > 10% of the parent compound (dodine)

In the hydrolysis of dodecylguanidine HCl as a function of pH at 25°C study (see Annex II A 2.9.1/01), it was concluded that dodecylguanidine HCl is hydrolytically stable in water without significative pH dependence. The DT_{50} estimated were 576, 914 and 1198 days for pH 5, 7 and 9 respectively (Daly D. *et al*, 1991).

B.8.4.2 Direct phototransformation of relevant metabolites, degradation and reactions products in water (Annex IIA 7.2.1.2)

Annex II point 7.2.1.1 requires a photolysis study to be conducted for relevant metabolites, degradation and reaction products, which account at any time for > 10% of the a.s. added unless sufficient information on the degradation is available for the test performed on the parent material, which is not the case, since metabolites degradates or reaction products do not make up for > 10% of the parent compound (dodine).

The photolytic degradation of dodine in water was studied and in natural water and in a sterilised water at pH 7. Both water samples were continuously irradiated for a period of 30 days under a sunlight-simulating light source (Xenon lamp ≥290nm) at temperature of 25°C.

It was concluded that dodine degraded under the test conditions with a photolytic DT_{50} of 174 days for sterilised water at pH 7 and a DT_{50} of 12.6 days in natural water. In the dark controls of the sterilised sample no degradation of dodine was observed, however in the natural water was possible to observe some degradation. Up to three photodegradation products were formed (detected by HPLC), only one of which, exceeded 10% of AR in the irradiated solution, the maximum of 42% was detected in the natural sample and 15% AR in the sterilised sample at pH 7. In the dark control of natural water this metabolite was also detected (max. 13%). The metabolite was identified as guanidine.

Based on OECD draft guideline and the OPPTS guideline (40°N) 30 days in sunlight correspond to approximately 10-13 days in the test system. As according to the guidelines metabolites are considered relevant in case they exceed 10% of applied within 30 days in sunlight, for buffer pH 7. At 40°N only guanidine might be a relevant metabolite (D.J. Slangen and B. van Noorloos, 2004).

The photolytic DT_{50} under natural conditions at 40°N was 403-456 days in sterilised water at pH 7 and 34-38 days in natural water.

B.8.4.3 Ready biodegradability of the active substance (Annex IIA 7.2.1.3.1)

Determination of 'ready' biodegradability: carbon dioxide (CO2) evolution test (modified sturm test) with dodine technical

Desmares-Koopmans, M.J.E. (2002)

Guidelines and GLP

The study was conducted according to OECD # 301 B. Principles of GLP were complied with.

Materials and methods

The biodegradability of dodine (batch nº S01/ 06 and purity of 96.2%) was studied in activated sludge obtained from municipal sewage treatment plant, "Watersharp de Maaskant", 's-Hertogenbosch, Netherlands. The sludge was stored under continuous aeration (conc. suspended

solids: 3.8 g/I). Before use sludge was settled for 30-90 min and the decanted liquid was used as inoculum at 10 ml/L of mineral medium.

Test was conducted for 28 days and the last CO_2 measurement occurred on day 29.

The test vessels were 2 glass brown bottles, the mineral medium was composed with CO_2-free air and the CO_2-absorber $Ba(OH)_2$, according to the guidelines.

The test substance was tested in duplicate at 31 mg per 2 litres, corresponding to 10 mg TOC/L. Two bottles only with inoculum were the blank controls. One bottle with the inoculum and approx. 40 mg/I of Na- acetate (equivalent to a TOC of 12 mg/I) was the positive control. One bottle with test substance, Na-acetate and inoculum together was the toxicity control.

The test started by bubbling CO_2-free air through the solutions at a rate of approx. 30-100 ml/min.

Weighed amounts of dodine technical were added to test bottles (test substance bottle A: 31.1 mg; test substance bottle B: 31.0 mg and toxicity control bottle: 32.3 mg). Ten ml of milli-RO water were added to each weighing bottle and after vigorous shaking the resulting suspension was added quantitatively to the test medium, since dodine technical was poorly soluble in water. The test solutions were continuously stirred during the test.

Findings

The theoretical CO_2 production of dodine was calculated to be 2.30 mg CO_2/ mg. The test was conducted in duplicates with theoretical CO_2 productions of 71.5 mg and 71.3 mg per 2 litres. The criteria for acceptability of the experiments were met.

The results for the blank and positive controls were the expected. The validity of the test system was proved, since in the positive control substance was degraded at 60% within 9 days, the total of CO_2 release in the blank reached a total of 51 mg CO_2 per 2 litters of medium and the difference between duplicate values of degradation of dodine technical, in percentage, was always less than 20.

The absence of toxic effects on biodegradation was also demonstrated in toxicity control, since more than 25% of degradation occurred within 14 days (based on $ThCO_2$), therefore the test substance has no inhibitory effects on microbial activity. Under the test conditions dodine was shown to be not readily biodegradable.

The results obtained for the test item (in duplicate: bottles A and B) are summarised below:

Table B.8 26 - CO_2 production (in mg) and percentage of degradation of dodine

	Bottle A		**Bottle B**	
Day	**Cumulative**	**Degradation**	**Cumulative**	**Degradation**
	CO_2	%	CO_2	%

	Bottle A		Bottle B	
2	0.2	0	0.0	0
5	0.7	1	0.1	0
7	1.0	1	0.3	0
9	2.2	3	0.9	1
14	2.6	4	2.0	3
23	5.9	8	2.0	3
26	5.9	8	2.0	3
27	6.0	8	2.0	3
29	6.8	9	2.7	4
29	6.8	9	2.7	4
29	6.8	9	2.7	4

B.8.4.4 Water/sediment study (Annex IIA 7.2.1.3.2)

Aerobic aquatic metabolism of Metasol DGH.

Cady C., Cranor W. (1992)

Guidelines and GLP

The study was conducted according to US EPA FIFRA N-162-4. Principles of GLP were complied with.

Materials and methods

A 30-day aerobic aquatic metabolism study was conducted with radiollabelled dodine hydrochloride (^{14}C-DGH with batch nº. of 2729-104, radiochemical purity of 98.5% and specific activity of 27.69 mCi/mmol), using a Nebraska (USA) sandy loam soil flooded with well water.

The study was conducted under dark conditions at 25°C. Bacterial and fungal viability were assessed using plate count analysis and was found to be representative of microbially active soil/water system. After receipt at the laboratory the soil was sieved through a 2 mm mesh screen. The soil characteristics are summarized in Table B.8-27. The water used was taken from a well and was characterized in terms of alkalinity, hardness, pH, temperature, conductivity and dissolved oxygen.

Table B.8 27 - Soil used to investigate the behaviour of dodine HCl in an aerobic aquatic system

Soil designation **Origin** **Soil type**	**Nebraska/ US** **Sandy loam**
Textural analysis sand silt clay	 56% 26% 18%
pH value	6.8
Organic matter %	1.6
Cation exange capacity (meq/100cc)	11.6
Bulk density	1.51
Moisture at field capacity 1/3 bar	17.37

Thirty six metabolism tubes were prepared, each containing 10 g of soil (dry weight) and 30 ml of water. Dosing of the test soils was done by injecting 195 µg of ^{14}C-DGH dose solution into 25 culture tubes (equivalent to 12 mg DGH/kg dry soil (12 kg DGH/ha) or 4 mg DGH/l water). Each dosed sample was capped and shaken manually for a few seconds to homogenize before being placed uncapped into the metabolism vessel. The vessel was placed in the dark at 25°C. The remaining 11 samples served as controls.

Soil and water samples were collected and extracted at 0, 1, 2, 7, 14, 21 and 30 days after application. The radioactive volatiles were trapped in KOH, ethylene-glycol and H_2SO_4 solutions. The radioactivity in the aqueous phase was detected by LSC. The extractable residues in the soil where determined by multiple step extraction with methanol followed by LSC with identification by HPLC. The extraction in the humic-fulvic fraction was conducted with KOH and HCl followed by LSC.

Findings

The results obtained with the ^{14}C-labelled dodine HCl, in terms of distribution of radioactivity and metabolites at different sampling dates are summarized in Table B.8 28.

Table B.8 28 - Recovery of applied radioactivity in % after application of 14C-dodine HCI to flooded soil under aerobic aquatic conditions at a rate of 12 kg dodine HCl/ha (12 mg dodine HCl/kg dry soil)

Days application	**Water phase (%)**	**$^{14}CO_2$ (%)**	**Extractable (%)**	**Bound residues (%)**	**Total (%)**
0	1.58	n/a	95.87	2.55	100.00
1	0.59	0.03	96.92	3.27	100.81
2	0.68	0.02	96.68	3.60	101.01
7	0.96	0.76	95.78	4.06	101.56
14	0.84	1.40	89.24	4.19	95.66
21	0.46	1.97	90.09	5.20	97.73
30	0.75	2.31	87.26	5.33	95.64

Recoveries of applied radioactivity were generally good throughout the study with all of the individual results at each time point falling within the range 96-102% AR. The non-extractable ^{14}C-residues rose steadily from 2.55% of IMD (initial measured dose) at day 0 to 5.33% (of which 4.8% in the humic acid fraction, 1.4% in the fulvic acid fraction and 93.9% in the humin phase) at day 30.

Cumulative volatiles reached 2.31 % of the IMD after 30 days and were identified as $^{14}CO_2$. Concurrently, the sums of residues in the soil extract and water layer dropped from 97.45% at day 0 to 88.01 % after one month of aerobic incubation. HPLC analysis of the soil extracts showed that approx. 98% of the radioactivity found in these extracts after 30 days was the parent compound. This corresponds to 85.98% of the IMD after 30 days. The water phase contained less than 2% of the IMD already on day 0. In the water phase, TLC analysis revealed that 12% of the radioactivity was the parent DGH. A polar degradate was observed at a concentration of approx. 0.01 ppm but could not be identified with HPLC-MS.

A first-order degradation curve was constructed for ^{14}C-DGH, allowing for the calculation of the degradation half-life of DGH. The slope of the first-order degradation curve was determined statistically to be different from zero and the calculated DT50 was 227 days.

Comments from RMS:

The significance of this study is doubtful to assess the behaviour of dodine in a natural water/sediment system since a soil has been used instead of sediments, no equilibrium period was respected, the samples were shaken after dosing increasing adsorption of the a.s. onto the soil particles (especially for dodine which has a natural potential to adsorb being positively charged). Study not reliable for risk assessment.

The fate of dodine in two water/sediment systems

Slangen P.J. (2004)

Guidelines and GLP

The study was conducted according to 95/36/EC (July 1995) - SETAC (Lynch, 1995) - CTB guideline (01 /2001) - BBA guideline (IV, 5-1). Principles of GLP were complied with.

Materials and methods

The study was conducted with radiolabelled dodine (batch nº 011009, radiochemical purity >97.9%) and non-radiolabelled dodine (batch S01106, purity 96.2%). The test material was incubated aerobically in the laboratory in two non- contaminated water/sediment systems at 20°C in the dark for 84 days. Sediments and water were collected at two different locations in the Netherlands at the top of the sediment layer with a water depth of 1 m (Lake of Oostvaardersplassen - OVP) or 3.5 m (Pool of Schoonrewoerdsewiel - SW). After receipt at the laboratory, the sediments were sieved through a 2 mm mesh screen and the water was passed through a 150 Nm sieve. The sediments characteristics are summarized in Table B.8 29. The water used was fully characterized in terms of appearance, hardness, pH, temperature, conductivity and dissolved oxygen.

Table B.8 29 - Sediments used to investigate the behaviour of dodine in two aerobic water/sediment systems

Sediment designation	OVP	SW
Redox pot. (mV, at sampling)	- 400	- 400
% sand (63 µm-2 mm)	2.9 – 4.3	85.1 – 86.7
% silt (2 µm-63µm)	78.5 – 80.4	8.5 – 9.2
% clay (<2 µm)	15.3 – 18.5	4.8 – 5.6
% organic C (dry weight basis)	2.0 – 2.1	1.7 -1.8
pH	7.6 – 8.4	7.3 – 8.1
CEC (meq/ 100 g dry weight)	12.8 – 13.0	5.1 – 6.6
Sampling depth	Top layer	Top layer
Texture	Silt loam	Loamy sand / Sand
Total P (mg/kg air dry weight)	754 – 938	766 – 848
Total N (mg/kg air dry weight)	1750 - 2016	1245 – 1539
ATP (µ/kg dry weight)	287	131
Colour	Grey/ green	Black

Eighteen metabolism flasks were prepared for each water/sediment system, each flask containing a sediment layer of 2-3 cm (200g) and a water layer of 5-6 cm (480 m1) on top of the sediment layer. Equilibration for both test systems lasted approx. 10 weeks, in dark at 20ºC. For each system, 12 (OVP) to 14 (SW) metabolism vessels were spiked at a nominal concentration of 100 µg as/L. The vessels were incubated at 20°C, in the dark, for 84 days. Sediment and water samples were collected and extracted at 1 hour, 5 hours and 1, 2, 5, 8 (SW only), 9 (OVP only), 20, 48 and 84 days after application.

The radioactive volatiles were determinate by NaOH and ethylene- glycol traps. The determination of radioactivity in the aqueous phase was conducted by LSC before and after a multiple step extraction with ethyl acetate and/or dichloromethane and n-hexane. The determination of residues (extractable) in the soil was conducted by multiple step extraction with methanol, dichloromethane and n-hexane followed by LSC. The parent compound was determined in the extracts by TLC.

Findings

The results obtained with the ^{14}C-labelled dodine, in terms of distribution of radioactivity and metabolites at different sampling dates are summarized in Table B.8 30.

Table B.8 30 - Recovery of applied radioactivity in % after application of 14C-dodine to two water/sediment systems

Days After application	Water phase (after release of dissolved CO_2) (%)	$^{14}CO_2$ (after release of dissolved CO_2) (%)	Sediment (extractable) (%)	Sediment (Bound residues) (%)	Total
SW					
1 hr	94.2	-	-	-	94.2
5 hrs	38.4	0.4	33.6	23.5	95.9
1 day	27.4	4.5	30.2	34.6	96.8
2 d	25.7	11.5	24.2	31.2	92.6
5 d	24.0	28.3	8.2	14.3	74.9
8 d	7.2	56.7	8	14.9	86.8
8 d	0.8	63.9	6.7	13.9	85.2
20 d	0.5	84.4	2.0	9.4	96.2
48 d	0.4	83.5	1.8	14.0	99.6
84 d	0.3	89.3	0.8	13.8	104.3
OVP					
1 hr	114.9	-	-	-	114.9
5 hrs	56.4	0.1	4.6	32.8	94.0
1 day	27.0	1.7	7.2	57.7	93.6
2 d	22.9	4.9	6.8	56.8	91.4
5 d	11.8	26.1	6.2	45.2	89.3
8 d	10.3	31.7	5.5	43.7	91.1
8 d	0.7	52.9	3.7	23.1	80.4
20 d	0.7	68.5	1.7	21.7	92.8
48 d	0.3	69.1	1.5	23.0	93.9
84 d	0.3	72.1	0.8	33.4	106.6

Measured redox potentials indicated aerobic conditions in the water layer and anaerobic conditions in the sediment layer of both systems throughout the incubation period. The extraction efficiency from the water was >90% for both systems. The extraction efficiency for the sediments was validated for the SW system prior to the start of the study and was found to be acceptable (69% recovery). For OVP, although generally not required for the second system, the extraction efficiency was also verified and was found to be lower (27% recovery) indicating clearly the different natures of the two types of sediment used and the strong adsorption potential of the active substance. Mass balances of applied radioactivity were within the range 80- 115% (OVP) and SW (75-104%) and so did not always meet the criterion of 90-110%.

Dissipation of dodine from the water layer was the result of transfer to the sediment as well as of mineralization. Dissipation of dodine from the sediment was the result of mineralization and formation of bound or strongly adsorbed residues. Complete mineralization into CO_2 was the major degradation process (72% OVP, 89% SW after 84 days). The amount of bound residues increased to 58% (OVP) and 35% (SW) after 1 day and dropped to values of 33% (OVP) and 14% (SW) after 84 days. The sum of unknown, organo-extractable metabolites never exceeded 2% of applied in the total system. Non-organo-extractable polar metabolites were also formed, one of them in significant amounts (up to 14.5% of applied after 2 days, then degrading quickly). It was speculated that one of the metabolites in the non organo-extractable fraction is urea, though probably not the major metabolite.

Dissipation of dodine from the water layer as well as total system of both OVP and SW water/sediment systems could be described by simple 1st order kinetics with r^2 values being 0.94-0.98 as given in the following Table.

Table B.8 31 - DT_{50} and DT_{90} values for dodine dissipation from OVP and SW water/sediment systems

Water/sediment systems	OVP		SW	
	water layer	whole system	water layer	whole system
DT_{50} (days)	0.37	0.51	0.12	0.92
DT_{90} (days)	1.2	1.7	0.4	3.1
r^2 (n)	0.979 (5)	0.978 (9)	0.980 (5)	0.948 (9)

As loss of CO_2 is thought to be the reason for the low mass balances (<90%) at some sampling points, and as mass balances are only slightly below 90%, results were still deemed acceptable. The results give a valid basis for this conclusion and the target of the study was met.

B.8.4.6 Summary of the fate and behaviour in water

Hydrolytic stability

Dodine was considered hydrolytically stable to pH values of 4, 7 and 9, at 25ºC. The DT_{50} values estimated were 576, 914 and 1198 for pH 5, 7 and 9 respectively. No degradation products were detected in the study.

Photolytic stability

The photolytic degradation of dodine in water was studied in buffer pH 7 and in natural water, the sterilized water was continuously irradiated for a period of 30 days under a sunlight-simulating light source (Xenon lamp ≥290nm) at temperature of 24.2°C. It was concluded that dodine degraded under the test conditions with a photolytic DT_{50} of 174 days in buffer pH 7 a DT50 of 12.6 days in natural water. In the dark controls the photolytic DT_{50} under natural conditions at 40°N was 403-456 days in buffer pH 7 and 34-38 days in natural water. One photodegradation product was detected, in irradiated and in natural water (irradiated and dark) that exceeded 10% of AR, and was identified as guanidine.

Biological degradation – "Ready biodegradability"

A readily biodegradation study ("modified Sturm test") of dodine was conducted by Desmares-Koopmans (2002). Dodine was applied at a rate of 31 mg per 2 litres, corresponding to 10 mg TOC/L. It was concluded that under test dodine was shown to be not readily biodegradable.

Water/sediment degradation

The fate of dodine in a water sediment system was studied by Slangen (2004). Sediments and water were collected at two different locations in the Netherlands in Lake of Oostvaardersplassen - OVP, (clay content between 15-18%, sediment pH between 7.6-8.4, and organic carbon content in sediment of 2%) and in a Pool of Schoonrewoerdsewiel – SW (clay content between 5-6%, sediment pH between 7.3-8.1, and organic carbon content in sediment of 1.8%).

The recovery of AR from all samples was > 90% AR. The non-extractable radioactivity increased through the incubation representing a maximum of 57% AR at 2nd day, in SW. $^{14}CO_2$ is the major degradation product, increasing to 89% after 84 days, in OVP.

It was possible to conclude that dodine disappeared rapidly from the water phase due to adsorption into the sediment and rapid mineralization to CO_2 with a DT_{50} of 0.12 - 0.37 days for the water layer and a DT_{50} of 0.51 - 0.92 days for the whole system. A DT_{90} of 0.4 - 1.2 days and 1.7 - 3.1 days was calculated for the water layer and for the whole system respectively. One highly polar metabolite was detected at 13.4% but could not be identified with certainty.

Table B.8 32 - DT_{50} and DT_{90} values for Dodine dissipation from OVP and SW water/sediment systems

Water/ sediment system	pH water	pH sed	T°C	DT_{50} system (d)	DT_{90} system (d)	r^2 system	DT_{50} water (d)	DT_{90} water (d)	r^2 water	Method of calculation
Lake OVP	7.6	8.4	20	0.51	1.7	0.978	0.37	1.2	0.979	1st order
Pool SW	7.3	8.1	20	0.92	3.1	0.948	0.12	0.4	0.980	1st order
Geometric mean				0.71	2.4		0.25	0.8		

Degradation in the saturated zone

No data submitted.

B.8.4.7 Assessment of the fate and behaviour in water

Dodine was hydrolytically stable at pH7 and 25ºC with DT_{50} of 914 days, under alkaline conditions dodine showed more stability and at acidic conditions dodine was less stable. Hydrolysis is not likely to be a significant route of dissipation of dodine in water. Photolysis will not play an important role for dodine degradation in water, since a DT_{50} of 174 days in buffer pH 7 was estimated, however in natural water conditions the DT_{50} was significantly lower (12days). Under dark conditions no degradation occurred (DT_{50} of 450 days), but in natural water the degradation occurred with a DT_{50} of 30 days, indicating that the degradation conditions are more related with water conditions than with light radiation.

Dodine is not readily biodegradable.

The water sediment study conducted with dodine showed that the a.s. dissipates rapidly from the water phase, by mineralisation and dissipation into sediment, with a DT_{50} < 1 day in the water phase. In sediment dodine dissipates rapidly from the sediment by mineralisation or by formation of bound residues strongly adsorbed to the sediment. In the whole system was estimated a mean DT_{50} of 0.71 days.

B.8.5 Impact on water treatment procedures (Annex IIIA 9.2.2)

No data submitted. Dodine is not expected to reach Waste Water Treatment Plants.

B.8.6 Predicted environmental concentrations in surface water and in ground water (PECsw, PECgw) (Annex IIIA 9.2.1, 9.2.3)

B.8.6.1 Predicted Concentrations in Surface waters – PECsw (Annex IIIA 9.2.1)

Dodine is used for the control of fungal disease in pome fruits and stone fruits like apple/pear, cherry, peach and plums. A brif summary of intended uses is given in the following table.

Table B.8 33 - Summary table of dodine intended uses

Crop	Disease	Growth stage	Application rate (g a.i./ha)	Maximum nr. of applications	Application interval
Apple	Apple scrab (*Venturia inaequalis*)	BBCH 01 to BBCH 69	680-900	5	7-10
Pear	Pear scrab (*Venturia inaequalis*)	BBCH 01 to BBCH 69	680-900	5	7-10
Cherry	*Blumeriella jappai = Coccomyces hiemalis*	BBCH 01 to BBCH 69	800	5	7-10
Peach	*Taphrina deformans*	BBCH 01 to BBCH 69	900	5	7-10

The Predicted concentration of dodine in surface water and in sediment was estimated regarding the worst case application scenario, i.e. 5 early applications of 900 g a.s/ha in orchards, with 7 days interval and interception of 40% (average crop cover).The assessment of PECsw and PECsed followed the STEP 1, STEP 2, STEP 3 and STEP 4 (for buffer zones). For STEP 1 and STEP 2 was the FOCUS model version 1.1, was used for pome/stone fruit, for North and South of Europe, regarding early applications and crop interception of 40%. The STEP 3 simulation was conducted with SWASH version 2.1 and drift calculator 1.1, MACRO version 4.3b or PRZM version 3.21β models to simulate potential surface water exposure and TOXSWA version 2.2.1 to simulate the fate and behaviour of the compound in the water body. As in the guideline 7193/VI/99 rev0 (Guidance document of calculation PEC values of plant protection products for soil, ground water, surface water and sediment) the route of contamination via drains in only possible when Koc < 500. In case of dodine, which has a Koc ranging from 5.51 x 105 to 1.27 x 107, there is no need to consider this source of loading. So, the TOXSWA calculations were only performed on scenarios where the product enter the compartment surface water by spray drift and run off, i.e., R1 pond, R1 stream, R2 stream, R3 stream and R4 stream. Five applications by air blast with an interval of 7 days between each application were considered.

Table B.8 34 - Summary table of specific parameters for FOCUS sw modelisation

Parameter	Value
Version control no. of FOCUS calculator	Version 1:1
Molecular weight (g/mol)	287.4
Vapour pressure (Pa at 50ºC)	5.49×10^{-6}
Water solubility (mg/L)	930 mg/l at 20ºC and pH 6.9
K_{OC} (L/kg)	423.65×10^{4}
1/n	0.9
DT_{50} soil (d)	13.25 days (mean of field studies)
DT_{50} water/sediment system (d):	0.92 (representative worst case from water /sediment studies)
DT_{50} water (d)	0.37
DT_{50} sediment (d)	0.92
Crop interception (%)	40 (average cover) for Step 1 and 2 Default values for Step 3 and 4
Application rate	900 g/ha
Number of applications	5
Interval between applications (d)	7

Note that, for STEP 2, 3 and 4 only the initial PEC (global maximum), the 4 day and the 21 day values were presented since they are the representative input parameter for aquatic risk assessment.

Table B.8 35 - FOCUS Step 1 PECsw and PEDsed for dodine application to pome/stone fruit

FOCUS STEP1 Scenario	Day after overall maximum	PEC_{SW} (µg/L)		PEC_{SED} (µg/kg)	
		Actual	TWA	Actual	TWA
	0	**87.644**		**$2.25E^{+03}$**	
	1	0.032	43.838	$1.37E^{+03}$	$1.81E^{+03}$
	2	0.015	21.930	644.090	$1.39E^{+03}$
	4	0.003	**10.969**	142.736	858.861
	7	0.000	6.269	14.891	515.019
	14	0.000	3.134	0.076	258.914
	21	0.000	**2.090**	0.000	**172.614**
	28	0.000	1.567	0.000	129.461
	42	0.000	1.045	0.000	86.307
	50	0.000	0.878	0.000	72.498
	100	0.000	0.439	0.000	36.249

Table B.8 36 - FOCUS Step 2 PECsw and PEDsed for dodine application to pome/stone fruit

FOCUS STEP 2 Scenario	Day after overall maximum	PEC_{SW} (µg/L)		PEC_{SED} (µg/kg)	
		Actual	TWA	Actual	TWA
Northern EU	0	**69.348**		**618.900**	
	4	0.015	**9.606**	30.384	204.420
	21	0.000	**1.830**	0.000	**40.947**
Southern EU	0	**69.348**		1220.000	
	4	0.034	**10.969**	142.736	858.862
	21	0.000	**1.831**	0.000	**60.469**

Table B.8 37 - FOCUS Step 3 PECsw and PEDsed for dodine application to pome/stone fruit

FOCUS STEP 3 Scenario	Water body	Day after overall maximum	PEC_{SW} (µg/L)		PEC_{SED} (µg/kg)	
			Actual	TWA	Actual	TWA
R1	Pond	0	3.063		1.658	
		4	0.053	0.930	0.665	1.433
		21	0.004	0.454	0.798	0.812
R1	Stream	0	41.862		4.786	
		4	0.000	1.682	1.398	2.919
		21	0.000	0.813	1.722	1.456
R2	Stream	0	55.589		5.345	
		4	0.000	1.140	0.873	2.893
		21	0.000	0.434	0.446	1.347
R3	Stream	0	**59.298**		**12.710**	
		4	0.001	**4.146**	6.177	7.505
		21	0.000	**2.255**	1.930	**4.750**
R4	Stream	0	42.110		10.977	
		4	0.000	1.927	1.763	6.067
		21	0.000	0.968	0.000	2.226

Table B.8 38 - FOCUS Step 4 PECsw and PEDsed for dodine application to pome/stone fruit (10 m buffer)

FOCUS STEP 4 Scenario	Water body	Day after overall maximum	PEC_{SW} (µg/L)		PEC_{SED} (µg/kg)	
			Actual	TWA	Actual	TWA
R1	Pond	0	1.915		1.037	
		4	0.033	0.581	0.416	0.499
		21	0.003	0.284	0.499	0.508
R1	Stream	0	17.042		2.310	
		4	0.000	0.685	0.295	1.266
		21	0.000	0.331	0.779	0.594
R2	Stream	0	22.633		5.232	
		4	0.000	0.464	0.855	2.822
		21	0.000	0.179	0.182	0.910
R3	Stream	0	**24.149**		6.215	
		4	0.000	**1.754**	1.444	3.784
		21	0.000	**0.960**	0.805	2.126
R4	Stream	0	17.143		**10.977**	
		4	0.431	0.797	1.763	6.067
		21	0.000	0.394	0.000	**1.527**

Table B.8 39 - FOCUS Step 4 PECsw and PEDsed for dodine application to pome/stone fruit (20 m buffer)

FOCUS STEP 4 Scenario	Water body	Day after overall maximum	PEC_{SW} (µg/L)		PEC_{SED} (µg/kg)	
			Actual	TWA	Actual	TWA
R1	Pond	0	0.554		0.300	
		4	0.003	0.168	0.119	0.260
		2	0.000	0.0821	0.136	0.147
R1	Stream	0	4.149		1.235	
		4	0.000	0.167	0.270	0.803
		21	0.000	0.0806	0.000	0.266
R2	Stream	0	5.511		5.174	
		4	0.000	0.113	0.846	2.786
		21	0.000	0.0470	0.045	0.688
R3	Stream	0	**5.887**		2.859	
		4	0.000	**0.511**	0.666	2.021
		21	0.000	**0.250**	0.201	0.760
R4	Stream	0	4.174		**10.976**	
		4	0.431	0.210	1.763	6.067
		21	0.000	0.096	0.000	**1.527**

Table B.8 40 - FOCUS Step 4 PECsw and PEDsed for dodine application to pome/stone fruit (30 m buffer)

FOCUS STEP 4 Scenario	Water body	Day after overall maximum	PEC_{SW} (µg/L)		PEC_{SED} (µg/kg)	
			Actual	TWA	Actual	TWA
R1	Pond	0	0.240		0.130	
		4	0.001	0.072	0.052	0.112
		21	0.001	0.036	0.062	0.064
R1	Stream	0	1.392		1.234	
		4	0.000	0.078	0.215	0.705
		21	0.000	0.270	0.000	0.210
R2	Stream	0	1.849		5.161	
		4	0.000	0.038	0.844	2.778
		21	0.000	0.019	0.015	0.664
R3	Stream	0	**1.981**		2.477	
		4	0.000	**0.245**	0.705	1.656
		21	0.000	**0.097**	0.111	0.497
R4	Stream	0	1.400		**10.976**	
		4	0.431	0.202	1.763	6.067
		21	0.000	0.051	0.000	**1.527**

Table B.8 41 - FOCUS Step 4 PECsw and PEDsed for dodine application to pome/stone fruit (35 m buffer)

FOCUS STEP 4 Scenario	Water body	Day after overall maximum	PEC_{SW} (µg/L)		PEC_{SED} (µg/kg)	
			Actual	TWA	Actual	TWA
R1	Pond	0	0.173		0.094	
		4	0.001	0.052	0.037	0.080
		21	0.001	0.255	0.045	0.046
R1	Stream	0	0.917		1.234	
		4	0.000	0.064	0.206	0.688
		21	0.000	0.021	0.000	0.201
R2	Stream	0	1.218		5.159	
		4	0.000	0.025	0.843	2.777
		21	0.000	0.014	0.010	0.660
R3	Stream	0	**1.309**		2.472	
		4	0.000	**0.200**	0.662	1.592
		21	0.000	**0.072**	0.073	0.442
R4	Stream	0	0.922		**10.976**	
		4	0.431	0.202	4.404	6.067
		21	0.000	0.540	0.001	**1.527**

Table B.8 42 - FOCUS Step 4 PECsw and PEDsed for dodine application to pome/stone fruit (40 m buffer)

FOCUS STEP 4 Scenario	Water body	Day after overall maximum	PEC_{SW} (µg/L)		PEC_{SED} (µg/kg)	
			Actual	TWA	Actual	TWA
R1	Pond	0	0.129		0.070	
		4	0.001	0.039	0.277	0.060
		21	0.001	0.019	0.033	0.034
R1	Stream	0	0.638		1.234	
		4	0.000	0.055	0.200	0.678
		21	0.000	0.018	0.000	0.195
R2	Stream	0	0.848		5.157	
		4	0.000	0.024	0.843	2.776
		21	0.000	0.011	0.007	0.657
R3	Stream	0	**0.914**		2.470	
		4	0.000	**0.173**	0.637	1.556
		21	0.000	**0.056**	0.051	0.421
R4	Stream	0	0.642		**10.976**	
		4	0.431	0.202	1.763	6.067
		21	0.000	0.051	0.000	**1.527**

The scenario R3 was identified as the representative worst case scenario, for surface water, as result of application of dodine to pomme/stone fruits. The worst case scenario identified for sediment was the R4.

B.8.6.2 Predicted Concentration in Ground waters – PECgw – (Annex IIIA 9.2.3)

The leaching potential of dodine has been determined using FOCUS PELMO model version 3.3.2, all nine scenarios have been considered, Châteaudun, Hamburg, Jokioinen, Kremsmünster, Okehampton, Piacenza, Porto, Sevilla and Thiva. The model simulations were carried out using

realistic worst-case input parameters (see Table B.8 42) and one realistic application scenario was taken in account. Five applications at highest dose, 900 g a.s. /ha, with 7 days application interval in apple orchards.

Dodine is a fungicide for apple/pear, cherry and peach. The "apple scenario" is the only available in the different models and can be considered representative of the intended uses. Since the application on apples is made from the bud opening (BBCH 01) until 28 days before harvest (BBCH 69), early application will be considered for modelisation. Regarding the FOCUSgw guidance (2000) recommendations, the applications were simulated to be carried out on the soil surface and the application rates were manually corrected for interception accordingly. The interception varies according to the time of application, 50% without leaves, 65% flowering, 70% foliage development and 80% full foliage.

For PELMO and PEARL models early applications were considered, ranging from March to May, for PRZM model, the application is relative to the emergence. For all models interseptions of 50% for the first two applications, 65% for the third and fourth applications and 70% of interceptions for the last application were considered. This can be considered a conservative approach because the foliage development is reduced and the interception is therefore smaller.

For the model MACRO, only one scenario (Chateaudun) is taken into account. The application range between April and May, and the same scheme of interception was considered.

Table B.8 43 - Input data and parameters in FOCUS for calculation of PECgw for dodine

Parameter	Value
Molecular mass	287.4 (20ºC)
Vapour pressure (Pa)	5.49×10^{-6} (50ºC)
Water solubility (mg/L)	930 (20ºC)
Koc (mL/g)	423.65×10^{4} (arithmetic median)
Freundlich 1/n	0.9 (default value)
Henry's law constant ($Pa*m^3mol^{-1}$)	1.7×10^{-3}
DT_{50} soil	13.25 d (geometric median)
Application rate	900 g/ha
Crop scenario/Number of applications/ interception	Apple scenario 5 early applications, from March to May: 1st: 50% interception 2nd: 50% interception 3rd: 65% interception 4th: 65% interception 5th: 70% interception

Table B.8 44 - Predicted concentrations of dodine at 1 meter soil depth following the use of dodine 400 SC in apples using the FOCUS gw Models

Scenario	80th percentile annual average concentration at 1m			
	PELMO	PEARL	PRZM	MACRO
Chateaudun	0.000	0.000	0.000	0.000
Hamburg	0.000	0.000	0.000	-
Jokioinen	0.000	0.000	0.000	-
Kremsmunster	0.000	0.000	0.000	-
Okehampton	0.000	0.000	0.000	-
Piacenza	0.000	0.000	0.000	-
Porto	0.000	0.000	0.000	-
Sevilla	0.000	0.000	0.000	-
Thiva	0.000	0.000	0.000	-

The model predicted that dodine would not be found at annual average concentrations as defined by FOCUS at concentrations greater than 0.001 µg/L, at 1 m depth, in any Châteaudun, Hamburg, Jokioinen, Kremsmünster, Okehampton, Piacenza, Porto, Sevilla and Thiva scenarios.

B.8.6.3 Monitoring data (Annex IIA 7.4)

No data submitted

B.8.7 Fate and behaviour in air (Annex IIA, 7.2.2; Annex IIIA 9.3)

No data submitted.

Comments by RMS:

Studies able to describe the behaviour of dodine in air, in particular the potential to volatilise from soil surface and/or from leaveswould be necessary. However due the low vapour pressure it is expected that no volatilization of the substance will be very low. At least an estimation of the degradation of dodine from reaction with OH should be presented.

B.8.7.1 Summary and Assessment of the fate and behaviour in air

Regarding the physical-chemical properties of dodine, in particular the vapour pressure of 5.49 x 10^{-6} Pa at 50ºC and the Henry's law constant < 1.7 x 10^{-3} Pa m^3/mol at 20ºC, it is possible to expect a very little potential of dodine to volatilise.

B.8.8 Predicted environmental concentrations in air (PECa) (Annex IIIA 9.3)

The predicted environmental concentrations in air (PEC_A) are expected to be negligible for dodine, since was estimated a vapour pressure, at 50ºC, lower than 5.49 x 10^{-6} Pa and a Henry's law constant < 1.7 x 10^{-3} Pa m^3/mol at 20ºC were estimated.

B.8.9 Definition of the residue (Annex IIA 7.3)

Soil: Dodine only

Dodine was shown to be degraded in all aerobic laboratory and field studies. The major degradation product was CO_2. Bound residues remain at low levels .No major metabolites were identified, only minor metabolites accounting for a very low percentage of applied radioactivity. These compounds have no toxicological or environmental significance. Thus the relevant residue in soil is dodine.

Surface water and sediment: Dodine only

The water sediment study conducted with dodine show that the a.s. dissipates rapidly from the water phase, by mineralisation and dissipation into sediment. In sediment dodine dissipates rapidly from the sediment by mineralisation or by formation of bound residues strongly adsorbed to the sediment. No major metabolites were detected in water/sediment systems. These compounds have no toxicological or environmental significance. Thus the relevant residue in water is dodine.

Ground water: Dodine only

The $FOCUS_{gw}$ model predicted that dodine would not be found at annual average concentrations as defined by FOCUS at greater than 0.001 $\mu g\ l^{-1}$, at 1 m depth, in Châteaudun, Hamburg, Jokioinen, Kremsmünster, Okehampton, Piacenza, Porto, Sevilla and Thiva scenarios for the intended uses.

Air: Dodine only

As default dodine might be considered as residue in air, although the Henry constant and vapour pressure indicate low potential for dodine volatilisation.

B.8.10 Monitoring data concerning fate and behaviour (Annex IIA 7.4)

No data submitted.

B.8.10.1 Summary of monitoring data

No data submitted.

B.8.11 References relied on

Annex point / reference number	Author(s)	Year	Title Source (where different from company) Company, Report No GLP or GEP status (where relevant), Published or not	Data Protection claimed Y/N	Owner
Annex IIA					
IIA, 7.1.1.1.1/01	Cooper J.D.L. et al	1996	14C-Dodine: Aerobic soil metabolism in two soils at 25°C. Rhone-Poulenc Agriculture, report # P94/162, GLP, unpublished	Y	CAG
IIA, 7.1.1.1.1/02	Lowden P *et al*.	1997	C14-dodine: Rate and route of degradation in three soil types under aerobic conditions with C14- guanidine labelled dodine and in one soil under aerobic conditions with C14-chain labelled dodine. Rhone Poulenc Agriculture, report # 10521, GLP, unoublished	Y	CAG
IIA, 7.1.1.1.2/01	Cady C.	1993	Anaerobic aquatic metabolism of Metasol DGH. ABC Lab, report # 38684, GLP, unpublished	Y	CAG
IIA, 7.1.1.1.2/02	Mislankar S.G.	2001	Dodine : photodegradation on soil. Aventis Crop Science, report # 2000X18609, GLP, unpublished	Y	CAG
IIA, 7.1.1.2.2/01	Norris F.A.	1999	Terrestrial Soil Dissipation after application of A.S. Brand 65W fruit fungicide to bare ground plots simulating an orchard or grove. Rhone Poulenc Ag, report # 45751 96X10342, GLP, unpublished	Y	CAG
IIA, 7.1.2/01	Williams M, Hargadine S.	1991	Soil/sediment adsorption-desorption of DGH, ABC Laboratories, report # 38683GLP, unpublished	Y	CAG
IIA, 7.1.3.2/01	Van Noorloos B, Slangen P.J.	2002	Aged leaching of Dodine. Notox, NL, report # 327094, GLP, unpublished	Y	CAG
IIA, 7.2.1.3.1/01	Desmares-Koopmans M.J.E	2002	Determination of 'ready' biodegradability: carbon dioxide (CO_2) evolution test (modified sturm test) with dodine technical. Notox, NL, report # 327105, GLP, unpublished	Y	CAG
IIA, 7.2.1.3.2/01	Cady C., Cranor W.	1992	Aerobic aquatic metabolism of Metasol DGH ABC Laboratories, report # 38682, GLP, unpublished	Y	CAG
IIA, 7.2.1.3.2/02	Slangen P.J.	2004	The fate of Dodine in two water/sediment systems. Notox / NL, report # 344677, GLP, unpublished	Y	CAG
Annex IIIA					
IIIA 9.6.0/01	Corman C.	2006	Determination of DT50 of dodine in soils	Y	CAG
IIIA 9.6/01	Cornille I.	2006	Evaluation of predicted environmental concentration in surface water (PECsw) for dodine following the use of A.S. 400SC, unpublished	Y	CAG
IIIA 9.7/01	Cornille I.	2006	Evaluation of predicted environmental concentration in surface water (PECgw) for dodine following the use of A.S. 400SC, unpublished	Y	CAG