STUDY TITLE

[Guanidine-^{14}C]Clothianidin: Time dependent Sorption from Four European Field Dissipation Soils

GUIDELINES

- OECD: Guideline 106, Adsorption/Desorption, 2001 (in parts)
- OECD: Guideline 307, Aerobic and Anaerobic Transformation in Soil, 2002 (in parts)

STUDY DIRECTOR

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STUDY COMPLETION DATE

2011-04-11

STUDY SPONSOR

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TEST FACILITY

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STUDY NUMBER

M131 1911-7

Total number of pages: 133

Data Confidentiality Statement

This page is intentionally left blank for the purpose of submitting administrative information that is required by regulations promulgated by various countries.

Certification of Good Laboratory Practice

This study was conducted in compliance with the current OECD Principles of Good Laboratory Practice (Appendix 22) and the German Chemical Law (Chemikaliengesetz), current version of Annex 1.

The experimental part of the study was not inspected by QAU in the 4th quarter of 2009 and in the 2nd quarter of 2010. However, this did not affect the integrity and correctness of the data and results.

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Quality assurance statement

Report No **M1311911-7**
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Print Date: 14 APR 2011

Quality Assurance (GLP)

Quality Assurance Statement

Title: **[Guanidine-14C]Clothianidin: Time-dependent Sorption from Four European Field Dissipation Soils**

Study: M1311911-7

Phase(s) of this study was/were inspected and records of these inspections were submitted to the study director, principal investigator (if relevant) and management as shown below.
This report has been inspected by the GLP Quality Assurance. As far as can be ascertained, the reported results accurately reflect the original data of the study.

Phase of Study	Inspection	Report
Study plan	21 SEP 2009	
Application	28 SEP 2009	28 SEP 2009
Evaluation	16 OCT 2009	16 OCT 2009
Processing	27 JAN 2010	27 JAN 2010
Application	06 JUL 2010	07 JUL 2010
Processing	09 NOV 2010	09 NOV 2010
Draft report	02 FEB 2011 - 31 MAR 2011	31 MAR 2011
Final report	12 APR 2011 - 14 APR 2011	

5.1.2.e WOO 2011-04-14
5.1.2.e WOO
GLP Quality Assurance

Study Information and Study Personnel

Study Number:	M 131 1911-7
Test item:	Clothianidin
Study Initiation Date:	2009-09-18 (study protocol)
Study protocol:	Dated 2009-09-18 1st amendment dated 2009-12-09 2nd amendment dated 2010-05-04 3rd amendment dated 2010-06-09
Study Completion Date:	2011-04-11
Experimental Start Date:	2009-09-22 (preparation of stock solution)
Experimental Termination Date:	2010-12-16
Study Director:	5.1.2.e WOO
Head of Test Facility and Representative of the Sponsor:	5.1.2.e WOO
Additional Investigators:	5.1.2.e WOO and 5.1.2.e WOO (spectroscopy) 5.1.2.e WOO radioactivity measurement) 5.1.2.e WOO (technician)
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QAU:	Bayer CropScience AG BCS-D-SPQ-GLP/QA Alfred-Nobel-Str. 50 D-40789 Monheim, Germany

All dates in the report are written as yyyy-mm-dd (unless within citations).

The format of the report bases on the Data Evaluation Report (DER) by EPA/PMRA. The relevant tables for the DER are highlighted and can be selected by the titles (see Table of Contents, Tables and Appendices).

Table of Contents

Abbreviations and Symbols

Abbreviation	Definition
µCi	Microcurie
µg	Microgram
µL	Microliter
µm	Micrometer
ACN	Acetonitrile
acc.	According
a.e.	Active equivalent
a.s.	Active substance
AR	Applied radioactivity
AU	Absorbance units
BBA	Biologische Bundesanstalt für Land- und Forstwirtschaft, Germany
Bq	Becquerel; disintegrations per second
BKG	Background
°C	Degree Celsius or Centigrade
CAS	Chemical Abstract Services
CEC	Cation exchange capacity (meq/100 g)
cm	Centimeter
cm^3	Cubic centimeter
cont.	Containing
contd.	Continued
CPA	Crop protection agent
cpm	Counts per minute
d	Day
DAT	Days after treatment
DIN	Deutsche Industrie Norm
DM	Dry matter
dpm	Disintegrations per minute
DT50/75/90	Dissipation time of 50/75/90% of test substance
EC	European Commission
EU	European Union
g	Gravitational constant
g	Gram
GEOmean	Geometric mean
GLP	Good Laboratory Practices
ha	Hectare
h	Hour(s)
HPLC	High performance liquid chromatography
i.d.	Inside diameter
ID	Identification
IUPAC	International Union of Pure and Applied Chemistry
kBq	Kilobecquerel
kg	Kilogram
L	Liter
LC	Liquid chromatography
LC-MS/MS	Tandem liquid chromatograph-mass spectrometer-mass spectrometer
LOD	Limit of detection
LOQ	Limit of quantification
LSC	Liquid scintillation counting
m/z	Mass to charge ratio

Remark: Not all entries necessarily appear within this report.

Abbreviations and Symbols, continued

Abbreviation	Definition
M	Molar
m	Meter
MBq	Megabecquerel
mCi	Millicurie
mg	Milligram
min	Minute
mL	Milliliter
mm	Millimeter
mmole	Millimole
mole	Mole
MS	Mass spectrometry
mV	Millivolt
MW	Molecular weight
N/A	Not applicable
nd	Non-detect (below limit of detection)
n.d.	Not determined
NER	Non-extractable residues
nm	Nanometer
NMR	Nuclear magnetic resonance
no.	Number
OC	Organic carbon
OECD	Organization for Economic Cooperation and Development
OM	Organic matter
ppb	Parts per billion or micrograms per kilogram
ppm	Parts per million or microgram per gram or milligrams per kilogram
psi	Pounds per square inch
PU	Polyurethane
QAU	Quality assurance unit
RA	Radioactivity
R^2 (or r^2)	Square of correlation coefficient
Rep.	Replicate
Rf	Ratio of compound movement to solvent front movement on TLC
ROIs	Regions of interest on TLC
RP	Reversed phase
rpm	Revolutions per minute
RSD	Relative standard deviation
R_t	Retention time
R_{TDS}	Ratio of concentration of test item in soil [µg/g]/ concentration of test item in solution [µg/mL]
s	Second
SETAC	Society of Environmental Toxicology and Chemistry, France
SS	Solvent system
STD	Standard deviation
$T_{1/2}$ (or $t_{1/2}$)	Half-life
TLC	Thin layer chromatography
USDA	United States Department of Agriculture
UV	Ultraviolet
v	Volume
WHC	Water holding capacity
wt	Weight

1 EXECUTIVE SUMMARY

The time-dependent sorption of [guanidine-^{14}C]Clothianidin was studied in a silt loam (Hoefchen am Hohenseh plot 4011, Replicate A and B, organic carbon 1.1%, pH 5.7 ($CaCl_2$), Burscheid, Germany), a sandy loam (Wellesbourne, Replicate C and D, organic carbon 0.8%, pH 6.1 ($CaCl_2$), Wellesbourne, United Kingdom), a clay loam (Mas du Coq, Replicate E and F, organic carbon 0.8%, pH 7.7 ($CaCl_2$), St. Etienné du Gres, France) and a sandy loam (Vilobi d'Onyar, Replicate G and H, organic carbon 0.8%, pH 6.1 ($CaCl_2$), Vilobi d'Onyar, Spain) for 120 days under aerobic conditions in the dark at 20°C and about 55% WHCmax (max. water holding capacity). Clothianidin was applied at a rate of 20 µg a.s./ 100 g soil, equivalent to approx. 150 g a.s./ha assuming a soil density of 1.5 g/cm³ and a soil depth of 5 cm.

The study was based in principle on OECD Guideline 307 and for the respective sorption parts on OECD Guideline 106. The study was carried out in compliance with valid GLP regulations.

The test system consisted of biometer-type flasks with traps for the collection of CO_2 and volatile organic compounds. Samples were analyzed after 0, 1, 3, 9, 21, 28, 35, 49, 63, 77, 98 and 120 days of incubation. In order to determine the sorption behavior, the treated soils were shaken with aqueous 0.01 M $CaCl_2$ solution for 24 hours at ambient temperature. The suspensions were centrifuged and the supernatants were analyzed separately. Then, the soil samples were extracted four times at ambient temperature with 80 mL acetonitrile/water (50/50, v/v, ambient extracts). With respect to the fast formation of non-extractable residues, the extraction was repeated once at an elevated temperature in microwave (aggressive extracts). The $CaCl_2$-exctracts as well as the ambient and aggressive organic extracts were analyzed by LSC and radio-HPLC. Identification of the transformation products was achieved by LC-MS/MS experiments and/or Co-chromatography. TLC analysis of selected samples was used to confirm the results.

The test conditions outlined in the study protocol were maintained throughout the study. The material balances ranged from 96.7% up to 104.2% of the applied amount of radioactivity (means of two replicates).

Extractable ^{14}C-residues decreased from 99.0%, 102.0%, 101.0%, and 100.9% of the applied amount at DAT-0 to 37.7%, 61.8%, 71.2% and 63.6% of the applied RA at the end of the study in soils Hoefchen am Hohenseh plot 4011, Wellesbourne, Mas du Coq and Vilobi d'Onyar, respectively.

The formation of bound residues increased with the overall metabolism of test compound. The non-extractable residues at DAT-0 varied from 2.2% – 3.0% of AR and were generated during the 24h lasting desorption step (see section 4.2.1). At study termination they increased to 30.8%, 20.2%, 18.1%, and 19.8% of AR in soils Hoefchen am Hohenseh plot 4011, Wellesbourne, Mas du Coq and Vilobi d'Onyar, respectively. They were separated into humin, humic acid and fulvic acid fractions for the last sampling interval.

The test item Clothianidin degraded moderately fast. Its amount in the extracts decreased from 96.9-99.8% on DAT-0 to 21.5 – 60.8% of AR after incubation for 120 days.

In all soils the degradation behavior of Clothianidin was best described using biphasic models. In soils Hoefchen am Hohenseh plot 4011, Mas du Coq and Vilobi d'Onyar, the degradation followed double first order in parallel kinetics (DFOP) according to the lowest chi^2 values. For soil Wellesbourne, the degradation was slightly better described using the FOMC kinetic model. The calculated half-lives were in the range of 9.7 to 174.6 days (geometric mean: 65.1 days).

Besides carbon dioxide six degradation products were detected and identified in the course of the incubation:

TZNG (FHW0107C) was formed at maximum amounts 1.0% of the applied amount in soil Hoefchen am Hohenseh plot 4011 (DAT-98), 3.0% in soil Wellesbourne (DAT-120), 2.1% in soil Mas du Coq (DAT-120) and 1.9% in soil Vilobi d'Onyar (DAT-77 and DAT-120). The amounts of the metabolite increased during the course of the study.

The second transformation product in soil was MNG (FHW0107F) which was found at maximum concentrations of 6.4% of the AR in soil Hoefchen am Hohenseh plot 4011 (DAT-120), 6.8% in soil Wellesbourne (DAT-77), 5.3% in soil Mas du Coq (DAT-120) and 4.5% in soil Vilobi d'Onyar (DAT-98).

The third identified transformation product in soil was TZMU (FHW0107E) which was observed in maximum concentrations of 10.6% of the AR in soil Hoefchen am Hohenseh plot 4011 (DAT-9), 5.9% in soil Wellesbourne (DAT-9), 2.2% in soil Mas du Coq (DAT-9) and 4.5% in soil Vilobi d'Onyar (DAT-9). The amounts of the metabolite decreased towards the end of the study.

The fourth identified transformation product in soil was TMG (FHW0107G) which was found at maximum concentrations of 0.8% of the AR in soil Hoefchen am Hohenseh plot 4011 (DAT-28, 35 and 77), 0.3% in soil Wellesbourne (DAT-28 and 35), 0.3% in soil Mas du Coq (DAT-98) and 0.4% in soil Vilobi d'Onyar (DAT-63). The amounts of the metabolite tend to decrease towards the end of the study.

The fifth identified transformation product in soil was NTG (FHW0107H) which was found at maximum concentrations of 2.8% of the AR in soil Hoefchen am Hohenseh plot 4011 (DAT-98), 3.1% in soil Wellesbourne (DAT-120), 2.1% in soil Mas du Coq (DAT-63 and 98) and 3.5% in soil Vilobi d'Onyar (DAT-120).

The sixth identified transformation product in soil was TZFA (BCS-CQ81511) which was found at maximum concentrations of 6.7% of the AR in soil Hoefchen am Hohenseh plot 4011 (DAT-35), 2.8% in soil Wellesbourne (DAT-21), 0.7% in soil Mas du Coq (DAT-9) and 1.3% in soil Vilobi d'Onyar (DAT-49). The amounts of the degradation product tend to decrease towards the end of the study.

In addition several minor transformation products were observed. Their sum at the different sampling times accounted for ≤ 2.4% (Hoefchen am Hohenseh plot 4011), 2.3% (Wellesbourne), 1.8% (Mas du Coq) and 2.1% (Vilobi d'Onyar) of the applied radioactivity.

At the end of the study at DAT-120, evolved $^{14}CO_2$ accounted for 29.3%, 16.2%, 9.4%, and 15.9% of the applied RA in soils Hoefchen am Hohenseh plot 4011, Wellesbourne, Mas du Coq and Vilobi d'Onyar, respectively. Only once a very small amount of volatile organic compounds was measured in the polyurethane foam (Replicate A, DAT-120, 0.3% of AR).

The sorption of Clothianidin to soil increased in the course of the study. The calculated R_{TDS} values were 1.3, 0.9, 0.6 and 0.8 mL/g for soils Hoefchen am Hohenseh plot 4011, Wellesbourne, Mas du Coq and Vilobi d'Onyar, respectively, at the beginning of the study (DAT-0). With time of aging in soil, these values increased to 4.8, 2.5, 1.6, and 2.5 mL/g on DAT-120 for the four soils.

Based on the results received within this study, a degradation pathway can be proposed. Metabolite TZNG was formed by demethylation of the test item. Metabolite MNG was formed by cleavage of the thiazolyl-moiety. Metabolite NTG was either formed by demethylation of MNG or by the cleavage of the thiazolyle moiety of TZNG. Metabolites TMG, TZMU and TZFA are characterized by a loss of the NO_2 moiety. The amount of formed carbon dioxide indicates the complete mineralization of Clothianidin and/or its transformation products in soil.

Result Synopsis:

Soil	**Hoefchen am Hohenseh plot 4011**	**Wellesbourne**	**Mas du Coq**	**Vilobi d'Onyar**
Type	Silt loam	Sandy Loam	Clay loam	Sandy loam
DT_{50} of Clothianidin	9.7 days	86.9 days	174.6 days	122.0 days
TZNG (% of AR)	max. 1.0%	max. 3.0%	max. 2.1	max. 1.9
MNG (% of AR)	max. 6.4%	max. 6.8%	max. 5.3%	max. 4.5%
TZMU (% of AR)	max. 10.6%	max. 5.9%	max. 2.2%	max. 4.5%
TMG (% of AR)	max. 0.8%	max. 0.3%	max. 0.3%	max. 0.4%
NTG (% of AR)	max. 2.8%	max. 3.1%	max. 2.1%	max. 3.5%
TZFA (% of AR)	max. 6.7%	max. 2.8%	max. 0.7%	max. 1.3%
Increase of sorption: $\frac{R_{TDS}\ DAT\text{-}120}{R_{TDS}\ DAT\text{-}0}$	3.7	2.7	2.6	3.1

2 INTRODUCTION

2.1 Objective of Study and Guidelines Followed

2.1.1 Purpose of Study

The aim of the present study was to investigate the changes of the sorption parameters K_d and K_{oc} (calculated as R_{TDS} –values) of Clothianidin affected by a preceding aging period in soil by time-dependent desorption experiments.

The concentration of the test item (and possible metabolites) was determined throughout the study, including formation of volatile products. Material balances were established at each sampling interval.

2.1.2 Relevant History and Background Information

Clothianidin (TI-435) is a systemic insecticide against sucking and biting insects for use in corn, cereals, canola and sugar beets. It is chemically related to the class of chloronicotinyles.

Information about its desorption from field accumulation soils in dependence of an aging period on soil is necessary for the overall assessment of the environmental fate and was investigated in this study.

2.1.3 Guidelines

The study was conceived based on the

- OECD Guideline 106 [1] (in parts)
- OECD Guideline 307 [2] (in parts)

2.1.4 Guideline Deviations

A guideline for a TDS study is not available. The study was based on OECD Guideline 106 and OECD Guideline 307.

2.2 Justification of Study Application Rate

The applied amount of Clothianidin was based on the maximum field use rate, which is 150 g Clothianidin/ha. The amount corresponding to 150 g a.s./ha was applied for the experiments.

Based on the conversion according to the OECD guideline [2] (soil depth of 5 cm and a bulk density of 1.5 g cm^{-3}), the projected concentration in soil corresponds to 20 µg Clothianidin per 100 g of soil calculated as dry matter (DM).

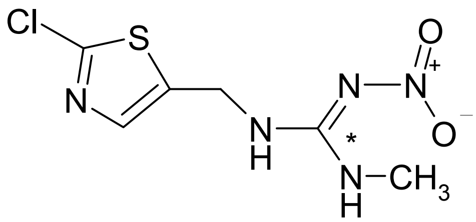
3 MATERIALS AND METHODS

3.1 Test and Reference Substances

A tabular overview of test and reference items, including chemical structure, CAS Nomenclature, CAS number, and molecular weight is given in Figure 1. Purity and content of the test item were determined prior to the applications.

3.1.1 Test Substance

- Common name: Clothianidin (report name)
- Compound Name: TI-435
- Chemical formula: $C_6H_8ClN_5O_2S$
- Chemical structure:



(* denotes positions of [^{14}C] label used)

- Molar mass: 249.7 g/mol
- IUPAC Nomenclature: (E)-1-(2-chloro-1,3-thiazol-5-ylmethyl)-3-methyl-2-nitroguanidine
- CAS Number: 210889-92-5
- Radiolabel used: [Guanidine-^{14}C]Clothianidin
- Site of radiolabel: (see for * in figure above)
- Specific activities: 4.34 MBq/mg (117.92 µCi/mg)
- Formulation: Pure ^{14}C-labeled product was used
- Physical state: solid, dried in vacuum
- Sample ID: KATH 6360
- Reference Synthesis: KML 2998-1
- Source: The certified radiolabeled test substance was provided by Dr. 5.1.2.e WOO, Bayer CropScience AG, Product Technology, D-42096 Wuppertal. In the course of the study the identity was confirmed by NMR, LC-MS and LC-MS/MS in the stock solution by the laboratory of DI A. Lagojda, BCS-D-ENSA. The spectra are presented in Figure 6 and Figure 7.
- Radiochem. purity: >98% (TLC, scan); >98% (HPLC, radioactivity detector)

- Chemical purity: >98% (HPLC, UV-detector, 210 nm)
- Storage stability: Not specified. The test item was dissolved in 6 x 1 mL methanol after arrival at the testing facility, resulting in approx. 1.5 mg Clothianidin /mL and stored in a freezer in the dark. The purity of test substance was verified after application. The storage stability of the test item in soil extracts was determined in the course of the study.

Physico-chemical properties of Clothianidin:

Parameter	**Values**	**Comments**
Water solubility	0.33 g/L at 20°C	6155-122 M-026209-04-1
Vapor pressure/volatility (p at 20 °C)	3.8 x 10^{-11} Pa	6155-115A M-026219-03-2
pK_a	11.09 at 20°C	6155-122 M-026209-04-01
log P_{OW}	0.7 at 25°C	6155-122 M-026209-04-1
Hydrolysis	Stable at pH 4, pH 7, pH 9	DEFT012 M-048047-01-1

3.1.2 Reference Substances

3.1.2.1 Non-Radiolabeled Clothianidin

- Sample ID: A1283742 (Original Sample ID: AE1283742 00 1B99 0001)
- Report name: Clothianidin
- Source and release: Bayer CropScience AG, BCS-D-EnSa-MeA
  D-40789, Germany
- Storage stability: expiry date 2012-05

3.1.2.2 FHW0107C

- Report name: TZNG
- Sample ID: FHW0107C
- Source and release: Bayer CropScience AG, BCS-D-EnSa-MeA
  D-40789, Germany
- Storage stability: expiry date 2014-09

3.1.2.3 FHW0107D

- Report name: TZU
- Sample ID: FHW0107D

- Source and release: Bayer CropScience AG, BCS-D-EnSa-MeA
  D-40789, Germany
- Storage stability: expiry date 2014-09

3.1.2.4 FHW0107E

- Report name:: TZMU
- Sample ID: FHW0107E
- Source and release: Bayer CropScience AG, BCS-D-EnSa-MeA
  D-40789, Germany
- Storage stability: expiry date 2014-09

3.1.2.5 FHW0107F

- Report name: MNG
- Sample ID: FHW0107F
- Source and release: Bayer CropScience AG, BCS-D-EnSa-MeA
  D-40789, Germany
- Storage stability: expiry date 2014-09

3.1.2.6 FHW0107G

- Report name: TMG
- Sample ID: FHW0107G
- Source and release: Bayer CropScience AG, BCS-D-EnSa-MeA
  D-40789, Germany
- Storage stability: expiry date 2014-09

3.1.2.7 FHW0107H

- Report name: NTG
- Sample ID: FHW0107H
- Source and release: Bayer CropScience AG, BCS-D-EnSa-MeA
  D-40789, Germany
- Storage stability: expiry date 2014-09

3.1.2.8 BCS-CQ81511

- Report name: BCS-CQ81511
- Sample ID: KATH 15179-4-5
- Source and release: 5.1.2.e WOO (Bayer Schering Pharma Aktiengesellschaft, Isotope Chemistry, Wuppertal, Germany)

The reference substances were provided by the laboratory of Dr. 5.1.2.e WOO and 5.1.2.e WOO (Bayer CropScience AG, Structure Elucidation) or by 5.1.2.e WOO

(Bayer Schering Pharma Aktiengesellschaft, Isotope Chemistry, Wuppertal, Germany). Copies of the Certificate of Analysis are archived within the raw data. For formulas, CAS nomenclature, chemical purity, molar mass, SMILES code, etc. of reference items see Figure 1. The reference items were dissolved in 1.8 mL of methanol for use as standards and stored in a freezer.

3.1.3 Control Substances

Control substances were not used.

3.2 Reagents and Equipment

A list of the used reagents and equipment will be found in Appendix 1.

3.3 Test System

3.3.1 Soils

Designation	Origin	Soil Type
Northern Europe		
Hoefchen am Hohenseh plot 4011 (ID: Replicate A and B)	Germany	Silt Loam
Wellesbourne (ID: Replicate C and D)	UK	Sandy Loam
Southern Europe		
St. Etienne du Gres, Mas du Coq (ID: Replicate E and F)	France	Clay Loam
Vilobi d'Onyar (ID: Replicate G and H)	Spain	Sandy Loam

3.3.1.1 Justification of Soils

Soils were taken from agricultural used areas in northern and southern Europe and cover representative soil types with a representative range of physicochemical properties. The selected soils are well-known from field dissipation as well as from soil metabolism studies.

3.3.1.2 Collection of Soils

The soils were sampled freshly from the field. A description of soil collection and storage is given in Appendix 2.

3.3.1.3 Properties of Soils

The physicochemical characteristics of the four soils are presented in Table 1. The pH values of the soils Hoefchen am Hohenseh plot 4011, Wellesbourne, Mas du Coq and Vilobi d'Onyar in aqueous 0.01 M $CaCl_2$ solution were 5.7, 6.1, 7.7, and 6.1, respectively. The organic carbon contents were 1.1%, 0.8%, 0.8% and 0.8% and the clay contents were 24%, 16%, 30% and 14% (USDA), respectively.
The microbial biomass was performed at commencement, in the middle and at the end of the study (Dr. T. Leicher, BCS -D-ETX).

3.3.1.4 Preparation of Soils

Prior to the start of the test, the soils were successively sieved to a particle size of ≤ 2 mm and stored at ambient temperature in the laboratory for a maximum of 25 days until pre-incubation. The soil moisture was determined by drying aliquots of the soils at 105°C using a balance for humidity determination (Mettler Toledo HB43). Water (VE) was added to each soil aliquot in a way that the final moisture was 55% of the maximum water holding capacity (WHC_{max}). Aliquots of the soils, each corresponding to 100 g dry weight, were filled into 300-mL Erlenmeyer flasks on 2009-09-25. The weights of all test vessels were recorded (see Section 3.5.2).

3.3.2 Experimental Apparatus

3.3.2.1 Equipment Used

A test system is presented in Figure 2.

3.3.2.2 Description of Experimental Set-up/Sampling Scheme

The incubation systems were static systems and consisted of Erlenmeyer flasks (300 mL) with 100 g soil (DM) for each sampling interval. After application, the flasks were closed with trap attachments, which were easily permeable for oxygen. The traps contained soda lime for absorption of CO_2 and a polyurethane foam plug for adsorption of volatile organic compounds.
Four spare samples per soil were incubated as well as samples for the determination of biomass.

Samples were taken 0, 1, 3, 9, 21, 28, 35, 49, 63, 77, 98 and 120 days after application. At each sampling interval, two samples per soil were removed from the climatic chamber and processed.

For more information of experimental design see also Table 2.

3.3.2.3 Trapping of $^{14}CO_2$ and Volatile Organic Compounds

The incubation vessels were closed with trap attachments containing soda lime for absorption of CO_2 and polyurethane foam plug for adsorption of volatile organic compounds (see also Figure 2).

3.4 Preparation and Application of Test Substance

Stock solution
The entire amount of the supplied [Guanidine-^{14}C]Clothianidin was dissolved in 6 mL methanol (ID: I 12614). The concentration of the test item in the stock solution (6439.06 kBq/mL) was determined by analyzing two dilutions of the stock solution (50 µL of stock solution + 4950 µL of methanol) with LSC. The identity of the test item in the stock solution was confirmed by NMR, LC-MS and LC-MS/MS (ID: Ja57SA01). LC-MS, LC-MS/MS and NMR- Spectra (LC-MS/MS, serial No. 13902) are presented in Figure 6 and Figure 7. The stock solution was stored in a freezer (< -20°C) when it was not used for processing.

Application solution
For the preparation of the application solution, 2.16 mL of the stock solution (ID: 12614) were dissolved in 29.84 mL of methanol and 32 mL of water (Ja57 App1). The concentration of the test item was determined by LSC (26397 Bq/100 µL or 60.8 µg/mL).

3.4.1 Application Procedures

On 2009-09-28, 329 µL of Ja57 App1 were added to each soil sample. After application, the samples were carefully shaken. Agglomeration of soil was avoided.

Soil moisture was documented by recording the starting weight (see Section 3.5.2) of the test vessels. Then, the test vessels (except DAT 0 vessels) were closed with trap attachments (see Figure 2).

3.4.1.1 Application Rate

Prior, during and after application, 329 µL aliquots of the application solution were dosed into 20 mL volumetric flasks in order to determine the exact amount of applied test item and the accuracy of application. The volumetric flasks were made up to volume with acetonitrile/water (1/1; v/v) and measured by LSC (2 x 100 µL). In total 5 aliquots of the application solution were analyzed.
A dilution of the application solution (ID: Ja57 AÜ 1) was analyzed by HPLC for identity and purity. The purity of Clothianidin was 97.96% (HPLC see Figure 5).

Applied amount of test item:

ID	**LSC** [Bq/0.1 mL]	**Mean** [Bq]	**AR** [Bq]	**Applied a.s.** [µg/test vessel]
JA57 AÜ 1	409.37 412.70	411.04	82208	
Ja57 AÜ2	430.45 437.35	433.90	86780	
Ja57 AÜ 3	431.01 433.17	432.09	86418	
Ja57 AÜ4	435.53 439.90	437.71	87542	
Ja57 AÜ 5	438.64 440.21	439.43	87886	
Mean (application)			**86167**	19.9
RSD %			2.66	

The applied amount of Clothianidin in this study was 19.9 µg/vessel which is equivalent to 86167 Bq. This was regarded as 100% of the applied radioactivity for all calculations.

3.4.1.2 Evaporation Procedures

No evaporation procedure was performed.

3.4.1.3 Control Substances Application

A control substance was not used.

3.5 Test System Maintenance and Sampling

3.5.1 Experimental Conditions and Monitoring

Incubation system: Erlenmeyer- flasks with trap attachments
Oxygen conditions: Aerobic
Light conditions: Dark
Temperature: 20 °C ± 2 °C
Soil moisture: 55% of the maximum water holding capacity.

For more details see Table 2.

3.5.2 Moisture Maintenance Procedures

The water which evaporated from soil was determined once on 2009-12-11 by weighing all remaining flasks without the traps. The weights were compared with the original weights and the evaporated water (approx. 3 g) was replaced by adding distilled water).

3.5.3 Sampling Intervals and Collection for Soils, $^{14}CO_2$ and Volatile Organics

The sampling intervals and the processing dates are given in Section 3.3.2.2. At the processing dates, the vessels of each soil system treated with active substance were completely sacrificed. Each test vessel was covered with an individual trap to determine the amount of $^{14}CO_2$ and organic volatiles (except DAT-0).

3.5.4 Sample Storage Conditions

Directly after sampling, the soils were extracted within one day. All extracts were further analyzed within a maximum of six days (e.g. LSC, first chromatographic analysis). During the time of processing, samples were stored at room temperature (< 1 day) or in a refrigerator (< 1 week). After processing, the extracts were deep frozen. Samples taken from DAT-1 to DAT-63 were first analyzed with a preliminary HPLC-method. They were re-analyzed with the evaluation method within a maximum of 91 days (Appendix 10).

The trap attachments were stored at room temperature until work-up. The analysis dates for CO_2 and organic volatiles are summarized in the following table:

Sampling Interval (DAT)	Date of Analysis of $^{14}CO_2$	Date of Analysis of organic volatiles
1, 3	2009-10-06	2009-10-07
9	2009-10-30	2009-10-30
21, 28	2009-10-29	2009-10-30
35	2009-11-05	2009-11-05
49	2009-11-16	2009-11-16
63	2009-12-03	2009-12-03
77	2010-01-11	2010-01-05
98	2010-01-12	2010-01-05
120	2010-02-01	2010-01-27

3.5.5 Maintenance and Collection of Volatile Traps

Prior to opening the incubation vessel (for moistening or sampling of soil), the entire vessels and traps were placed in a desiccator. Volatile (radioactive) compounds, possibly still present in the vessel, were transferred into the trap attachment by evaporating the entire system carefully (test vessel with trap) with low pressure.

3.6 Analytical Methodology

3.6.1 Sample Preparation and Processing

- Extraction procedures for all intervals:

In order to determine the desorption behavior of the test item in soil, the entire soil sample was first extracted with aqueous $CaCl_2$ solution. After shaking for 24 hours to gain equilibrium, the suspension was centrifuged and the supernatant was removed. The remaining soil was extracted with organic solvents at room temperature (ambient organic extracts) and under elevated temperature conditions (70°C, aggressive organic extracts). A scheme of processing can be found in Figure 3.

Extraction procedure:

Extraction step	Solvent	Volume	Duration	Temper-ature	Cycles	Combine extracts
1	0.01 M $CaCl_2$ solution	100 mL (with soil moisture)	24 h (shaking)	Ambient temperature, about 20°C	1x	-
2-5	Acetonitrile / water 50/50 (v/v)	80 mL	30 min (shaking)	Ambient temperature, about 20°C*	4x	Yes
6	Acetonitrile / water 50/50 (v/v)	80 mL	10 min (microwave)	70°C	1x	-

After each extraction step the suspension was centrifuged for about 15 minutes (about 5000 x g) and the supernatant was either decanted directly ($CaCl_2$ extracts) or using a folded filter (ambient and aggressive organic extracts).

The supernatants resulting from step 2-5 (ambient extracts) were combined. The 0.01 M $CaCl_2$ desorption solution as well as the ambient and aggressive organic extracts were analyzed separately for volume and radioactivity. All extracts were analyzed by HPLC either directly ($CaCl_2$ extracts) or after concentration (ambient and aggressive organic extracts). In addition, selected extracts (DAT-120, Replicates A, C, E and G) were analyzed with a confirmation method (TLC).

For the first replicates of samples taken on DAT-0, the ambient organic extracts (steps 2-5) were analyzed individually by LSC in order to determine the extracted amount of radioactivity for each individual step. The results are summarized in Appendix 11, showing that the extracted activity remarkably decreased with each extraction step.

The folded filters used for decanting were dried, quartered, compressed and combusted in order to determine the collected amount of radioactivity.

The amount of bound residues in the extracted soil samples was determined by combustion of soil aliquots after freeze drying and homogenization.

- Clean-up procedures:

No clean up procedures were used.

- Sample concentration procedures:

Aliquots of the ambient and aggressive organic extracts were concentrated prior to HPLC-analysis. For this purpose, 20 µL of dobanol were added to 5 mL aliquots of the extracts. The samples were concentrated for 90 min in a vacuum concentrator and analyzed by HPLC. The volumes of the concentrates were determined for representative samples (about 1.5 mL).

The recovery of radioactivity after the concentration step was tested using the ambient organic extracts of samples taken on DAT-0, DAT-63 and DAT-120 and the aggressive organic extracts of samples taken on DAT-63 and DAT-120. As a result, the recoveries in the single concentrates ranged from 87.1% - 99.2% and 93.6% - 101.6% for the ambient and aggressive organic extracts, respectively, which shows that no significant amount of radioactivity was lost during the concentration procedure. Furthermore the recovery of radioactivity after concentration was determined by means of samples that were also used for storage stability tests (see section 5).

- Special processing for metabolite isolation:

3.6.1.1 First metabolite isolation

260 mL of sample Ja57 49 C SO and 260 mL of sample Ja57 49 D SO were combined and concentrated to about 260 mL using a rotary evaporator. The concentrate was sucked through a Strata-x- cartridge (8B-S100-JDG, polymeric reversed phase) and the eluent was collected. The cartridge was rinsed with 10 ml of water and eluted with 10 mL of methanol. Prior to the solid phase extraction, the cartridge had been pre-conditioned with 5 mL of methanol and 10 mL of water.

Aqueous Phase after SPE:

The aqueous phase eluted from the SPE-cartridge and the 10 mL of water used to rinse the cartridge were combined and 75 g of NaCl were added. The solution was shaken with 100 mL of acetonitrile. The organic phase was concentrated to dryness using a rotary evaporator. The remainder was dissolved in 10 mL of methanol. The solution was centrifuged and analyzed by LSC. The aqueous phase was also analyzed by LSC. As a result 6.4% of the radioactivity was found in the aqueous phase and 0.6% of the radioactivity was found in the organic phase.

The aqueous phase was additionally shaken with 2 x 50 mL of ethyl-acetate. The organic phases were concentrated to dryness using a rotary evaporator and the remainder was dissolved in methanol/water. The solution was analyzed by LSC. As a result the radioactivity remaining in the water phase could not be extracted. The respective fractions were removed.

Eluat:

The methanolic eluat of the SPE cartridge was concentrated to about 1.8 mL using a speedvac concentrator. The concentrate was fractionated by HPLC. Six different fractions were collected and analyzed by LSC. All fractions were concentrated to dryness using a speedvac concentrator and transferred into HPLC-vials using 2 x 500 µL of methanol. The samples were subjected to HPLC and TLC analysis.

In order to verify the distribution of radioactivity in the sample, a combined HPLC/LSC analysis and several TLC measurements with different eluents were performed.

3.6.1.2 Second metabolite isolation

Respectively 250 mL of the extracts Ja57 35 A SO and Ja57 35 B-SO were combined and concentrated to a volume of about 15 mL using a rotary evaporator. The concentrate was sucked through a Strata-X-cartridge which was rinsed with 10 mL of water and eluted with 10 mL of methanol. Prior to the solid phase extraction, the cartridge had been pre-conditioned with 5 mL of methanol and 10 mL of water.

Prior and after SPE-clean-up the aqueous phase was analyzed by LSC. In addition, the methanolic eluat was analyzed for radioactivity. The eluate was concentrated to dryness using a speedvac concentrator and transferred into a HPLC-vial using methanol/water. 150 µL aliquots of the sample were fractionated via HPLC. Three fractions (TMG, 5-9 min, U19) were collected and concentrated via speedvac.

The TMG-fraction was further cleaned up via HPLC and analyzed by LSC. The polar fraction collected in the range of 5-9 min was concentrated and not further processed. The U19 fraction was cleaned up in another HPLC-run. The fraction was concentrated to dryness using a speedvac concentrator, transferred into a HPLC vial using methanol and sent to the laboratory of DI A. Lagojda for structure elucidation.

- Characterization of bound residues:

The bound residues in the four soils were characterized and fractionated into humin, humic acid and fulvic acid fractions. A flow chart showing the procedure is presented in Figure 4.

3.6.2 <u>Sample Analysis</u>

3.6.2.1 Chromatographic and Spectroscopic Procedures

<u>High Performance Liquid Chromatography (Preliminary and Evaluation Method)</u>

All extracts were subjected to HPLC analysis using a Purospher STAR C18e HPLC column. The analysis of the extracts was started using a preliminary HPLC-method (Ja57.m, 2009-09-04 or 2009-10-02).

During the course of the study, several HPLC-methods were tested in order to improve the peak separation. Finally, the preliminary method was modified by adding ammoniumformiate to eluent A (evaluation method). The evaluation method (Ja57.m 2009-12-17 or 2010-01-21) was used for the quantification of the test item and the transformation products in the extracts sampled from DAT-1 onwards. Extracts of samples which were first analyzed with the preliminary HPLC-method were re-analyzed with the evaluation method. The dates of sampling and first and second analysis are summarized in Appendix 10.

For selected samples taken on DAT-120, the eluent of the HPLC-runs was collected and analyzed by LSC. The mean recovery of radioactivity was determined to be

102.9% (range: 97.3 – 114.7%) which shows that no radioactivity was lost during the HPLC-run.

Instrument **Software**	HP 1100 (Agilent) Chemstation /Gina Star Version 2.18
Column	Purospher Star C18e, 250 mm x 4.6 mm (5 µm) + Guard Column 4 * 4 mm CAT 1.50359, No. Hx755206
Oven temperature	40 °C
Flow rate	1 mL/min
Detection Radioactivity UV	 Flow through detector (Ramona Star, Raytest) with solid scintillator cell (glass 370 µL, Raytest) Agilent 1100 variable wavelength detector (254 nm)
Preliminary Method: Ja57.m (2009-09-04, 2009-10-02)	
Eluent A:	Milli-Q-Water, 1 % formic acid
Eluent B:	Acetonitrile, 1 % formic acid
Evaluation Method: Ja57 (2009-12-17, 2010-01-21)	
Eluent A:	Aqueous ammoniumformiate [5g/L], 1% formic acid
Eluent B:	Acetonitrile, 1% formic acid
Separation	Gradient: 0.0 min 0% B 2.0 min 0% B 45.0 min 25% B 50.0 min 95% B 54.0 min 95% B 55.0 min 0% B 5.0 min Posttime
Injection volume	up to 600 µL

Soil extracts were qualitatively analyzed by HPLC either directly ($CaCl_2$ extracts) or after a concentration step (ambient and aggressive organic extracts, see section 3.6.1).

The assignment of HPLC peaks to test item and degradates was done using their retention times.

Retention times of the test item and relevant metabolites (DAT-120, replicate C, $CaCl_2$ extract, see Figure 11, page 77):

6.57 min.: NTG
8.77 min.: MNG

20.90 min.:	TZFA
24.33 min.:	TMG
32.45 min.:	TZMU
37.40 min.:	TZNG
42.12 min.:	Clothianidin

The quantification of the test item and the degradation products in extracts was calculated based on the distribution of the HPLC - zones and the amount of RA in the extracts.

Representative HPLC-chromatograms measured in extracts of samples taken on DAT-0, DAT-63 and DAT-120 from soil Wellesbourne (Replicate C) are shown in Figure 11.

Thin Layer Chromatography (TLC), Confirmation Method

Aliquots of the solutions were spotted onto the plates as bands using an automatic spotter (TLC sampler 4 automatic, Camag). The distribution of radioactive zones on the plates was measured using a Bio-Imaging Analyzer (BAS 2000, Fuji Co.) and quantified with the software package AIDA (Version 4.14.024, Raytest).

TLC analyses were performed using silica gel plates (Merck Si60, F_{254}, size 200 x 200 mm) with the solvent systems given below.

TLC solvent systems: Ethylacetate/i-propanol/VE-water/formic acid (65/ 23/ 11/ 1)

Ethylacetate/i-propanol/VE-water/triethylamine (50/ 20/ 5/ 10)

First, the upper solvent system was run over the first 5 cm. After the silica gel plates were dried, the second solvent system was used for separation over the whole plate. The confirmation method was used to analyze the first replicates of each type of extract sampled on day 120.

Several further TLC methods were used to confirm the peak separation at different sampling times.

Identification Methods (Parent and Relevant Metabolites)

MS-Spectrometry

The electro-spray ionization MS spectra (ESI) were obtained with a LTQ Orbitrap XL mass spectrometer (Thermo, San Jose, CA, U.S.A.).

The chromatographic conditions for the MS experiments are given below. The HPLC instrument used for chromatography was an Agilent HP1100 (Agilent, Waldbronn, Germany). The flow from the HPLC column was split between UV-detector followed by a radioactivity detector (Ramona Star, Raytest, Straubenhardt, Germany) and MS spectrometer.

Samples	HPLC Instrument and Setup	Solvents	Gradient
MS Instrument: LTQ Orbitrap XL (FT-MS)			
JA57SA01 JA57SA02 JA57SA03 JA57SA04 JA57SA05 JA57SA06	HPLC: Agilent HP 1100 Column: Nucleodur C18 Gravity, 3 µm, 250 x 2 mm (MN) Flow: 0.2 mL/min	A: 0.1% formic acid in water B: 0.1% formic acid in acetonitrile	0-1 min 5% B, at 25 min 95% B, at 35 min 95% B

NMR-Spectroscopy

The 600 MHz NMR-spectrum of sample JA57SA01 was recorded in a tube with a BRUKER AV 600 instrument (Bruker, Karlsruhe, Germany). Sample ID and solvent (supplier: Merck, Wilmad or Aldrich) are given in the spectrum header.

3.6.2.2 Metabolite Identification and Characterization Procedures

<u>LC-MS and LC-MS/MS</u>
The identification and confirmation of relevant ^{14}C-peaks in the HPLC-chromatograms was performed using fractions of combined ambient organic extracts of soil Wellesbourne (DAT-49) or soil Hoefchen am Hohenseh (DAT-35). Samples were worked up as described in see section 3.6.1. The collected fractions were analyzed by LC-MS/MS (section 3.6.2). In the extracts of soil Wellesbourne (section 3.6.1.1), the substances appearing at retention times of 41.08, 36.38, 31.18 and 21.28 min were characterized as the test item Clothianidin, TZNG, TZMU and TMG. The identity of these substances was also confirmed by comparing their retention times with the retention times of the non-labeled reference items prior to LC-MS and LC-MS/MS analysis.

For the identification of the peak appearing at a retention time of about 17.5 min, two ambient organic extracts of soil Hoefchen am Hohenseh (Ja5735-A-SO and Ja5735-B-SO) were used. The peak was isolated as described in section 3.6.1.2. According to LC-MS and LC-MS/MS the substance was identified as TZFA (Figure 9).

<u>Co-Chromatography</u>

Several extracts sampled on DAT-28 were spiked with 20 µL of the following non-labeled reference items and analyzed with HPLC (Ja57.m, 2009-10-02):

Ja57 28 D SOe	FHW0107C (TZNG)
Ja57 28 A SHe	FHW0107D (TZU)
Ja57 28 A SOe	FHW0107E (TZMU)
Ja57 28 C DE	FHW0107F (MNG)
Ja57 28 B SHe	FHW0107G (TMG)
Ja57 28 G DE	FHW0107H (NTG)

As a result, the peaks of the non-labeled reference items TZNG, TZMU, MNG, TMG and NTG corresponded well with the ^{14}C-peaks detected in the respective samples. For TMG and NTG the ^{14}C-peaks were, however, very small. A representative chromatogram is presented in Figure 10. TZU was not detected in the extracts.

A concentrated organic extract of soil Hoefchen am Hohenseh (Ja57 63 A SOe) was fortified with 2 µL of a non-labeled TZFA solution and analyzed with HPLC. As a result, the peak in the ^{14}C- chromatograms appeared at the same time as the TZFA-peak in the UV-chromatogram, corrected for delay between both detectors.

3.6.2.3 Radiocarbon Determination Procedures

- Volatile Compounds

- Organic volatiles adsorbed to PU foam plug

The PU foam was extracted with 50 mL ethyl acetate. An aliquot (5 mL) of each extract was submitted for radioactivity measurement.

- $^{14}CO_2$

For determination of $^{14}CO_2$, the soda lime contained in the trap attachments was dissolved in 60 mL 18% hydrochloric acid using the apparatus described in Appendix 9. The liberated CO_2 was absorbed by a special absorption / scintillation cocktail (Carbosorb E / Permafluor E$^+$, Perkin-Elmer Life Science) and the radioactivity was subsequently measured by liquid scintillation counting (*e.g.* LKB-Wallac 1219 Spectral (Perkin-Elmer Life Science), average counting efficiency = about 69.7-91.3% and background = 26-29 cpm). The LOD is considered to be two times of the radioactivity in the background (approx. 1 Bq).

The amount of radioactivity assigned to $^{14}CO_2$ in selected samples (Ja57 12 A/C/E/G) was determined qualitatively and quantitatively by precipitation of formed [^{14}C]barium carbonate. The radioactivity liberated from the CO_2 cocktail using acetic acid was transferred into a sodium hydroxide solution. This solution was treated with 1mL 1 M aqueous sodium carbonate solution and 8 mL of 1 M aqueous barium chloride solution to precipitate the radioactive carbon dioxide. The complete precipitation and the absence of radioactivity in the supernatant after centrifugation were taken as evidence for the identity of carbon dioxide as final degradation product (Barium carbonate precipitation, Appendix 12).

- Liquid Samples

The soil and PU-foam extracts were measured in 2 mL and 7 mL Quicksafe A + 5% water (Zinsser Analytic) using a liquid scintillation counter (*e.g.* LS 6500, Beckman, counting efficiency = about 82.74 - 92.01% and background = 13 to 23 cpm). The alkaline extracts obtained from the characterization of the bound residues were measured in 10 mL of Rotiszint eco plus (Carl Roth GmbH + Co. KG, background = 22 - 23 cpm). The LOD is considered to be two times of the radioactivity in the background (approx. 0.8 Bq).

- Soil Samples

The extracted soil samples were freeze-dried and milled. Radioactivity in soil was determined by combusting three portions (about 1 g) of each sample in an oxidizer

(OX 500, Harvey Instruments). The folded filters used for the decanting of extracts were dried, quartered, compressed and combusted. The resulting carbon dioxide of soil samples and filters was absorbed in Oxysolve C 400 (Zinsser Analytic) and measured in a liquid scintillation counter (*e.g.* LKB-Wallac 1219 Spectral (Perkin Elmer Life Science), average counting efficiency 69.7 - 91.3% and background = 17 to 19 cpm). The LOD is considered to be two times of the radioactivity in the background (approx. 0.6 Bq).

3.6.2.4 Characterization of Bound Residues

Bound residues were separated into humin, humic acid and fulvic acid by addition of sodium hydroxide and subsequently precipitation of the supernatant with hydrochloric acid. A flow chart showing the procedure can be found in Figure 4.

3.7 Calculations

3.7.1 Sample Calculations

Calculations were performed using the computer software ®Microsoft Excel. In general, the program uses nine decimal places for calculations. The results given are rounded to one or two digits. Thus, rounding errors may occur if recalculations are made using the listed (rounded) figures. Example calculations are presented in Appendix 8.
Calculations of percent of applied radioactivity were based on the applied radioactivity (see 3.4.1.1), which was set as 100%.

- *Concentration in the soil extracts and volatile traps*

$$\text{Concentration}\left[\%\ \text{applied}\right] = 100 * \frac{Bq_{aliquot} * \dfrac{ml_{total}}{ml_{aliquot}}}{Bq_{applied}}$$

- *Amount of non-extractable radioactivity*

$$\text{Non-extractables}\left[\%\ \text{applied}\right] = 100 * \frac{Bq_{aliquot} * \dfrac{g_{dry\ soil\ total}}{g_{dry\ soil\ aliquot}}}{Bq_{applied}}$$

- *Concentration of metabolites in a certain phase*

$$\text{Metabolite}_{phase}\left[\%\text{applied}\right] = \frac{\left(\%_{chromatogramm}\right) * \left(\%_{applied,phase}\right)}{100}$$

3.7.2 Half-lives for the parent compound:

Degradation rates and DT_{50} values of Clothianidin for soils Hoefchen am Hohenseh plot 4011, Wellesbourne, Mas du Coq and Vilobi d'Onyar were calculated using three different kinetic models. First order kinetics (SFO), first order multi compartment (FOMC) and double first order parallel kinetics (DFOP) were calculated. The most suitable approach was chosen according to FOCUS based on the chi² value and visual inspection [4]. The Kinetic Modeling Software KinGUI 1.1 was used to determine the kinetics [5]. For optimal goodness of fit, the initial value was also allowed to be estimated by the model. The data points for day 0 were set to the total material balances.

For the kinetic evaluation of the data the following models were tested:

Simple first order model (SFO):

$$M_P(t) = M_0 \exp^{(-kt)}$$

$M_P(t)$ = Total amount of chemical present at time t
M_0 = Total amount of chemical present at time t = 0
k = Rate constant

First order multi compartment model (FOMC):

$$M_P(t) = M_0 \left(\frac{t}{b} + 1 \right)^{(-a)}$$

$M_P(t)$ = Total amount of chemical present at time t
M_0 = Total amount of chemical present at time t = 0
a = Shape parameter determined by CV of k values
b = Location parameter

Bi-exponential model (double first order in parallel, DFOP):

$$M_P(t) = M_1 \exp^{(-k_1 t)} + M_2 \exp^{(-k_2 t)}$$

$M_P(t)$ = Total amount of chemical present at time t
M_1 = Amount of chemical applied to compartment 1 at time t = 0
M_2 = Amount of chemical applied to compartment 2 at time t = 0
k_1 = Rate constant in compartment 1
k_2 = Rate constant in compartment 2

Metabolites:

The formation and degradation of metabolites was not described with models in the course of this study.

3.7.3 Time-Dependent Sorption

3.7.3.1 R_{TDS} Values

For determination of the test item desorption dependent on the incubation (aging) time, the first extraction at each sampling date was performed using aqueous $CaCl_2$-solution. The soil to solution ratio was 1:1.

The R_{TDS} values were calculated by

$$R_{TDS} = \frac{\text{concentration of test item in soil extract } [\frac{\mu g}{g}]}{\text{concentration of test item in } CaCl_2 \text{ solution } [\frac{\mu g}{mL}]} = \frac{c_{extr}}{c_{des}}$$

A detailed explanation of R_{TDS} calculations for DAT-0 (sample JA57 00 A) is given in Appendix 16 (example of soil Hoefchen am Hohenseh plot 4011).

3.7.4 Statistical Methods

- Arithmetic means were used in case of all LSC measurements.
- The mathematical models used are referenced in 3.7.1 and 3.7.2.
- Outlier rejection criteria were not used.

3.7.5 Definitions of Detection Limits (LOD and LOQ)

- Amount radioactivity in soil extracts:

  The limit of detection (LOD) of the LSC counter was set to twofold the background radioactivity. The latter was determined to be about 23 cpm (0.4 Bq) at maximum which results in a LOD of about 0.8 Bq sample. The background was subtracted automatically from each sample after measurement. The limit of quantification (LOQ) was set to three times the background radioactivity, i.e. about 1.2 Bq (0.27 ng). The lowest amount measured in the soil extracts was about 45 Bq, i.e. about 37.5 times higher than the LOQ.

  The amounts of radioactivity extracted from the PU-foam used to collect the volatile organic compounds were lower than the LOQ in most of the samples. Therefore, it was concluded, that no significant amounts of organic volatiles were formed (except for Replicate A, DAT-120).

- Carbon dioxide and combustion of soils:

  The LOD values for the determination of carbon dioxide and the combustion of soils are 1 Bq and 0.6 Bq respectively (section 3.6.2.3). The lowest amounts of radioactivity measured were 47 Bq and 6.7 Bq for carbon dioxide and the bound residues, respectively, which is 47 and 11 times higher than the LOD.

- HPLC (evaluation method):

  The LOQ in HPLC depends on the peak width and the chromatographic behavior of each metabolite. The radioactive zones or peaks in HPLC (regions of interest, ROIs) were selected manually according to the positions in the chromatogram. The limit of detection for a single peak in the sum of all soil extracts can be estimated from the data sheets for distribution of HPLC analysis (Appendix 4, sum of $CaCl_2$, ambient organic and hot organic extracts) as percentage of applied radioactivity. The LOD was in the range of 0.1% AR.

Based on a LOD of 0.1% of the applied radioactivity (for the sum of all extracts) the limit of quantification (LOQ) is estimated to be about 0.3% of AR (3 times x LOD = 0.3%). However, values between LOD and LOQ were also taken into account in the tables and for calculations.

3.8 Additional Test on Extraction

A simplified extraction method was tested with two representative soils Hoefchen 4011 and Vilobi d´Onyar. Vessels (300 mL Erlenmeyer flasks) with 100 g soil (DM) were be prepared according Section 3.5, but with different soil batches. The test item was applied with the same amount of test item and the vessels were incubated (moisture, temperature, dark) in the same way as before. One set of samples was analyzed using the standard TDS-method (aqueous $CaCl_2$ extraction, ambient and hot organic extract, see Section 3.6.1). The second set was extracted using a simplified one-step method with microwave and organic solvent.

Six soil samples were prepared for each soil and incubated for 0, 58 and 124 days. Samples of DAT-0 and DAT-58 were stored in a deep freezer first and analyzed together with samples DAT-124, which were analyzed immediately after sampling. At each sampling point the extractability was investigated in duplicates:

Extraction methods:

a) according the TDS-method (aqueous $CaCl_2$ extraction, ambient and hot organic extract),

b) using a simplified extraction method:
The soil in a test vessel was carefully mixed and subdivided into five parts of similar amounts and each of the five portions were transferred into 500 mL-centrifuge vessels. To each of these vessels with soil, 40 ml extraction solvent (acetonitrile/water= 80/20, v/v) were added and the vessels were loosely closed with screw tops.

A total of 10 vessels were placed in a microwave. The temperature gradient program was increasing to 70°C within 3 minutes and then the temperature was constant for 15 minutes while mixing with a magnetic stirring bar.

The samples were cooled down to ambient temperature by placing them at room temperature. The samples were centrifuged at ca. 3000 x g for 10 minutes. The supernatants of the five corresponding portions of the same soil sample were collected in a separate flask, shortly mixed and the volume was determined.

After LSC, aliquots of the extracts were concentrated using a speedvac concentrator and analyzed for parent and metabolites by radio – HPLC.

4 RESULTS AND DISCUSSION

In the present study, the degradation and sorption behavior of Clothianidin was investigated in four soils.

4.1 Properties of Test System

4.1.1 Soil Properties

The degradation and sorption of Clothianidin was investigated in four soils originating from northern and southern regions of the EU (Germany, UK, France and Spain). The soils varied in relevant soil parameters such as texture (20% - 72% sand, 12% - 54% silt, 14% - 30% clay), organic carbon (0.8% - 1.1%) and pH (in aqueous $CaCl_2$: 5.7 – 7.7). The physicochemical characteristics of the used soils are shown in detail in Table 1.

4.1.2 Physical and Biological Conditions

From 2009-09-28 onwards, the test vessels were incubated in a thermostatically controlled incubation room (bldg. 6670, room no. 163) in the dark at a temperature of 19.3°C ± 0.2°C for a maximum period of 120 days. The temperature was recorded by a data logger (Squirrel, Grant Co.). Data are presented in Appendix 15. The samples were closed with a trap attachment which allows sampling of volatile compounds, but is permeable for oxygen exchange (Figure 2).

The moisture of the soil was determined once during the study (2009-12-11). The lost water was replaced by distilled water (about 3 g).

Determinations of microbial biomass were carried out on DAT- 0, DAT-74 and DAT-120 using the method described by J. P .E. Anderson [3]. The microbial activity in samples with and without treatment with solvent decreased to about 55% - 83% of the initial value at the end of the study in the four soils. Results are shown in Table 4. Under the conditions of a laboratory experiment the decrease of biological activity is inevitable without any soil treatment. The reason is the gradual nutrient depletion of the soil which is caused by the lack of continuous supply of organic matter from natural decay processes.

Further information about the experimental design is summarized in Table 2.

4.2 Analytical Methodology

4.2.1 Verification of Extraction Procedures

Soils were at first extracted with aqueous $CaCl_2$ in order to determine the sorption of the test item to soil. The soils were intensively extracted four times at ambient temperature and once under reflux conditions with acetonitrile / water, then. The mean recoveries of the test item at sampling interval day 0 range from 99.0 - 102.0%. These results indicated that the extraction method was well suitable to extract the applied [Guanidine-^{14}C]Clothianidin from the soil matrix (Appendix 4 to Appendix 7)

In addition, a simplified one-step extraction method with microwave extraction (70°C, 15 min.) to be used in e.g. other studies was tested. The recovery in freshly applied samples as well as in aged samples was the same as for the first six-step extraction method. The extraction using the microwave extraction method was as efficient as the TDS method (Appendix 20).

4.2.2 Verification of Chromatographic Procedures

Dilutions of the application solution (Ja57 App1) were used to determine the detection limit of the preliminary HPLC method and to test if the results of the analysis are reproducible. For this purpose, aliquots containing nominally 500, 100, 50, 10, 5, 4, 3 and 2 Bq were injected into the HPLC-system. The peak containing 5 Bq could be clearly identified. As a result the limit of detection was defined as 5 Bq. The correlation between injected radioactivities and the area determined during the HPLC-analysis was very high (R^2=1).

The dilution of Ja57 App1 containing the highest amount of the test item was injected five times in order to test the reproducibility of the preliminary HPLC-method. The mean area attributed to the test item was determined to be 5323.88 counts per second with a relative standard deviation of 0.66%. This shows that the results were well reproducible.

Two extracts of samples taken on DAT-0 (preliminary method) and all extracts from samples taken on DAT-120 (evaluation method) were analyzed for radioactivity prior and after HPLC analysis. The mean recoveries were 96.8% (range: 96.6 – 97%) and was 102.9% (range: 97.3 - 114.7%) for the preliminary and the evaluation method, respectively. This shows that with both methods no radioactivity was lost on the HPLC-column.

For samples taken from DAT-1 onwards, the quantification of the test item and the metabolites in the soil extracts was performed using the evaluation method. Based on the detection level of minor metabolites in the sum of all extracts, the LOD was considered to be about 0.1% of the applied radioactivity for liquid samples. The limit of quantification (LOQ) can be calculated as 3-times the LOD, equivalent to LOQ = about 0.3%. However, data between LOD and LOQ were also used for evaluation as given.

Representative HPLC chromatograms showing the separation of parent and transformation products can be found in Figure 11. These results indicated that the chromatographic method was well suitable to analyze the applied [Guanidine-^{14}C]Clothianidin as well as the degradation products.

In order to confirm the results of the HPLC analysis, extracts sampled on DAT-120 were investigated with a second separation method (TLC). A comparison of both methods is given Figure 12. The results obtained with both methods are comparable and no additional metabolite was detected in significant amounts.

4.3 Material Balance

In the present study, 86167 Bq of [Guanidine-^{14}C]Clothianidin have been applied to each flask containing 100 g (as dry matter) of soil. The total applied radioactivity was taken as 100 % for calculation of material balances.

The mean recoveries of radioactivity for all soils and sampling times were in the range of 96.7% to 104.2% for the test systems, indicating that no radioactivity was lost in the course of the study. The exact figures are presented in Table 5 to Table 8. Ranges of total recovery, extracted amounts of radioactivity, radioactivity of volatile components - detected by processing absorbents in the trap attachments - and the portion of bound residues are presented in the table below.

Radioactivity Balances Given as Minimum and Maximum for the Soils Hoefchen am Hohenseh plot 4011, Wellesbourne, Mas du Coq and Vilobi d'Onyar:

(values as % of applied radioactivity, mean values in case of duplicates)

Soil	Hoefchen am Hohenseh plot 4011	Wellesbourne	Mas du Coq	Vilobi d'Onyar
Duration (d)	120	120	120	120
Total Recovery (%)	96.7 – 101.9	98.2 – 104.2	98.1 – 103.4	98.0 – 103.1
Extracted RA (%)	37.7 – 99.0	61.8 – 102.0	71.2 – 101.0	63.6 – 100.9
Max. Volatile RA (%)	29.5	16.2	9.4	15.9
Bound Residues (%)	3.0 – 31.5	2.2 – 20.2	2.4 – 18.1	2.3 – 19.8

Significant amounts of other volatile products apart from $^{14}CO_2$ could not be detected (≤ 0.3% of AR). Raw data of the distribution of radioactivity for all soils are shown in Appendix 13.

4.4 Distribution and Composition of Residues

- Mineralization (Trapped $^{14}CO_2$) and Other Volatiles

The amount of formed $^{14}CO_2$ increased steadily during the entire study period. At the end of the study, 120 days after application, evolved $^{14}CO_2$ was 29.3%, 16.2%, 9.4%, and 15.9% of the applied RA in soils Hoefchen am Hohenseh plot 4011, Wellesbourne, Mas du Coq and Vilobi d'Onyar, respectively (see Table 5 to Table 8). Only once a very small amount of volatile organic compounds was measured in the polyurethane foam (Replicate A, DAT-120, 0.3% of AR, see Table 5).

- Extracted Radioactivity

Extractable ^{14}C-residues decreased from 99.0%, 102.0%, 101.0%, and 100.9% of the applied amount at DAT-0 to 37.7%, 61.8%, 71.2% and 63.6% of the applied RA at the end of the study in soils Hoefchen am Hohenseh plot 4011, Wellesbourne, Mas du Coq and Vilobi d'Onyar, respectively (see Section 4.3). The percentage of extracted radioactivity in desorption solution ($CaCl_2$-extract), ambient and aggressive organic extracts is given in Table 5, Table 6, Table 7, and Table 8. The raw data in the individual soil extracts of soil Wellesbourne are presented in Appendix 14.

In the course of the experiment, a number of zones of RA were detected in the extracts and quantified by HPLC. The biotransformation (sum of desorption solution, ambient and aggressive organic extracts, ID: DE, SO, and SA) expressed as % of AR was calculated and is presented in Table 9, Table 10, Table 11 and Table 12 (mean of duplicates) as well as in Appendix 4, Appendix 5, Appendix 6, and Appendix 7 (individual samples).

The decline of the test item and the formation and/or decline of the major degradation products (measured in the soil extracts) and $^{14}CO_2$ are plotted in Figure 14. The raw data of HPLC analysis, i.e. the composition of extractable residues of the individual soil extracts for soil Wellesbourne are presented in Appendix 14.

The test item Clothianidin degraded moderately fast. After incubation for 120 days its amount in the extracts decreased to 21.5 – 60.8% of AR.

Besides carbon dioxide, six degradation products were detected and identified in the course of the incubation:

TZNG (FHW010JC) was formed at maximum amounts 1.0% of the applied amount in soil Hoefchen am Hohenseh plot 4011 (DAT-98), 3.0% in soil Wellesbourne (DAT-120), 2.1% in soil Mas du Coq (DAT-120) and 1.9% in soil Vilobi d'Onyar (DAT-77 and DAT-120). The amounts of the metabolite increased during the course of the study.

The second transformation product in soil was MNG (FHW010JF) which was found at maximum concentrations of 6.4% of the AR in soil Hoefchen am Hohenseh plot 4011 (DAT-120), 6.8% in soil Wellesbourne (DAT-77), 5.3% in soil Mas du Coq (DAT-120) and 4.5% in soil Vilobi d'Onyar (DAT-98).

The third identified transformation product in soil was TZMU (FHW010JE) which was observed in maximum concentrations of 10.6% of the AR in soil Hoefchen am Hohenseh plot 4011 (DAT-9), 5.9% in soil Wellesbourne (DAT-9), 2.2% in soil Mas du Coq (DAT-9) and 4.5% in soil Vilobi d'Onyar (DAT-9). The amounts of the metabolite decreased towards the end of the study.

The fourth identified transformation product in soil was TMG (FHW010JG) which was found at maximum concentrations of 0.8% of the AR in soil Hoefchen am Hohenseh plot 4011 (DAT-28, 35 and 77), 0.3% in soil Wellesbourne (DAT-28 and 35), 0.3% in soil Mas du Coq (DAT-98) and 0.4% in soil Vilobi d'Onyar (DAT-63). The amounts of the metabolite tend to decrease towards the end of the study.

The fifth identified transformation product in soil was NTG (FHW010JH) which was found at maximum concentrations of 2.8% of the AR in soil Hoefchen am Hohenseh plot 4011 (DAT-98), 3.1% in soil Wellesbourne (DAT-120), 2.1% in soil Mas du Coq (DAT-63 and 98) and 3.5% in soil Vilobi d'Onyar (DAT-120).

The sixth identified transformation product in soil was TZFA (BCS-CQ81511) which was found at maximum concentrations of 6.7% of the AR in soil Hoefchen am Hohenseh plot 4011 (DAT-35), 2.8% in soil Wellesbourne (DAT-21), 0.7% in soil Mas du Coq (DAT-9) and 1.3% in soil Vilobi d'Onyar (DAT-49). The amounts of the degradation product tend to decrease towards the end of the study.

In addition several minor transformation products were observed. Their sum at the different sampling times accounted for ≤ 2.4% (Hoefchen am Hohenseh), ≤ 2.3% (Wellesbourne), ≤ 1.8% (Mas du Coq) and ≤ 2.1% (Vilobi d'Onyar) of the applied radioactivity.

- Non-extractable Radioactivity (Bound Residues)

The remaining RA measured in the soil after all extraction steps was determined for each individual test vessel. The formation of bound residues increased with the overall metabolism of test compound. The non-extractable residues at DAT-0 varied from 2.2% – 3.0% of AR and were generated during the 24h lasting desorption step (see section 4.2.1). At study termination they increased to 30.8%, 20.2%, 18.1%, and 19.8% of AR in soils Hoefchen am Hohenseh plot 4011, Wellesbourne, Mas du Coq and Vilobi d'Onyar, respectively. The data are presented in Section 4.3; the corresponding graphs are presented in Appendix 3.

- Characterization of bound residues

Bound residues of samples from the last sampling day were separated into fractions of humin, humic acid, and fulvic acid.

Soil	Hoefchen am Hohenseh plot 4011	Welles-bourne	Mas du Coq	Vilobi d'Onyar
Total bound residues (%*)	29.9	19.3	16.1	18.1
Humin (%)	13.1	8.7	7.8	7.0
Humic acid (%)	1.7	0.9	0.1	0.7
Fulvic acid (%)	15.1	9.7	8.1	10.3

*) % of applied radioactivity of replicate 1 at study termination (set as 100%)

4.5 Identification and Characterization of Parent and Transformation Products

According to LC-MS and LC-MS/MS the identity of the test item in the stock solution (Figure 6) as well as in one extract sample (Figure 8) was confirmed by the molar mass pattern of fragmentation. In the stock solution, the identity of the test item was also established using NMR analysis (Figure 7).

The identification and confirmation of relevant ^{14}C-peaks in the HPLC-chromatograms was performed in fractions of combined ambient organic extracts of soil Wellesbourne (DAT-49) or soil Hoefchen am Hohenseh (DAT-35).

In the extracts of soil Wellesbourne (see section 3.6.1.1 for workup), the substances appearing at retention times of 41.08, 36.38, 31.18 and 21.28 min were characterized as the test item Clothianidin and the transformation products TZNG, TZMU and TMG using LC-MS and LC-MS/MS (Figure 9). The identity of these substances was also confirmed by comparing the retention times of the individual peaks with the retention times of the non-labeled reference items prior to LC-MS and LC-MS/MS analysis.

For the identification of the peak appearing at a retention time of about 20.32, two ambient organic extracts of soil Hoefchen am Hohenseh (Ja57 63 A SO and Ja57 63 B SO) were used. Samples were prepared as described in section 3.6.1.2. According to LC-MS and LC-MS/MS the peak was identified as TZFA (Figure 9).

Co-chromatography was used to identify the transformation products MNG and NTG and to confirm the identity of TZNG, TZMU, TMG and TZFA. Several extracts sampled at DAT-28 were spiked with 20 µL of solutions of the non-labeled reference items TZNG, TZU, TZMU, MNG, TMG and NTG and analyzed with HPLC. As a result, the peaks of the non-labeled reference items TZNG, TZMU, MNG, TMG and NTG corresponded well with the ^{14}C-peaks detected in the respective samples. For TMG and NTG the ^{14}C-peaks were however very small. A representative chromatogram (TZNG) is presented in Figure 10.

A concentrated organic extract of soil Hoefchen am Hohenseh (Ja57 63 A SOe) was fortified with 2 µL of a solution of the freshly synthesized non-labeled reference item TZFA and analyzed with HPLC. As a result, the peaks in the ^{14}C- and UV-chromatograms appeared at the same time.

[^{14}C]-carbon dioxide was identified by its binding to soda lime. It was liberated by strong acid followed by reabsorption with a specific CO_2 binding cocktail (Carbosorb). In addition, it was identified by precipitation of $BaCO_3$ (shown for the first replicate sampled at DAT-120 of each soil, Appendix 12).

4.6 Time-Dependent Sorption

In order to determine the sorption behavior under equilibrium conditions, the soil samples were shaken with 0.01 M aqueous $CaCl_2$ solution as first extraction procedure. The results of these desorption measurements affected by the aging period are presented in Table 13. Details for calculations can be found in Appendix 16 to Appendix 19. For graphical presentation see Figure 15.

The calculated R_{TDS} values were 1.3, 0.9, 0.6 and 0.8 mL/g for soils Hoefchen am Hohenseh plot 4011, Wellesbourne, Mas du Coq and Vilobi d'Onyar, respectively, at the beginning of the study (DAT-0). With time of aging in soil, these values increased to 4.8, 2.5, 1.6, and 2.5 mL/g on DAT-120 for the four soils. Regarding the aging time of 120 days, the mean R_{TDS} values increased by a factor of 2.6 to 3.7 (Mean: 3.0). R_{TDS} calculations can be found in Appendix 16 (for soil Hoefchen am Hohenseh plot 4011 as an example).

4.7 Kinetic Analysis of Data

4.7.1 Kinetics of Parent Compound Degradation

The data for the parent compound Clothianidin were evaluated according to FOCUS [4] as described in Section 3.7.2. The initial concentration at day 0 was included in the parameter optimization procedure. For calculation of DT_{50} values that trigger additional studies, the best available model should be used. The best fit kinetic model was chosen based on the chi^2 confidence criterion and visual assessment. The results are summarized in the table below (the best fits are highlighted in bold letters).

Calculation of DT_{50} for Clothianidin (parent)

Soil	Kinetic Model	DT_{50} [d]	DT_{90} [d]	Visual Assessment*	Chi^2
Hoefchen am Hohenseh plot 4011	SFO FOMC **DFOP**	31.4 10.5 **9.7**	104.4 682.1 **256.4**	-- + **+**	20.5 4.1 **3.6**
Wellesbourne	SFO **FOMC** DFOP	96.8 **86.9** 88.6	321.7 **> 1000** 530.1	-- + **+**	9.1 **2.2** 2.3
Mas du Coq	SFO FOMC **DFOP**	168.1 577.8 **174.6**	558.4 > 1000 **710.2**	-- + **+**	4.4 2.2 **1.8**
Vilobi d'Onyar	SFO FOMC **DFOP**	118.9 178.8 **122.0**	394.9 > 1000 **585.3**	-- + **+**	6.6 2.1 **1.8**

*Visual assessment: + good O medium -- bad

Except for the SFO model in soil Hoefchen am Hohenseh plot 4011, the chi^2 values of the fits were below 15. In all soils the degradation behavior was best described using biphasic models. In soils Hoefchen am Hohenseh plot 4011, Mas du Coq and Vilobi d'Onyar, the degradation of Clothianidin followed double first order in parallel kinetics (DFOP) according to the lowest chi^2 values. For soil Wellesbourne, the degradation was slightly better described using the FOMC kinetic model. The half-lives were in the range of 9.7 to 174.6 days (geometric mean: 65.1 days).
The corresponding curves for the best fits and the relevant statistical evaluations are plotted in Figure 16 (Hoefchen am Hohenseh plot 4011), Figure 17 (Wellesbourne), Figure 18 (Mas du Coq), and Figure 19 (Vilobi d'Onyar).

The biphasic degradation behavior may be related to the observed sorption behavior of the test item to soil. Increasing amounts of the applied test item may be adsorbed to protected compartments where binding is stronger but microbial degradation did not occur.

4.7.2 Decline of Metabolites

The degradation of the metabolites was not described within the scope of this study.

4.7.3 Degradation Pathway

Based on the results received within this study a degradation pathway can be proposed. Metabolite TZNG was formed by demethylation of the test item. Metabolite MNG was formed by cleavage of the thiazolyl moiety. Metabolite NTG was either formed by demethylation of MNG or by the cleavage of the thiazolyl moiety of TZNG. Metabolites TMG, TZMU and TZFA are characterized by a loss of the NO_2 moiety. They may be direct transformation products of the test item or formed from each other. The test item and/or the metabolites are further transformed to CO_2 or bound residues. The proposed pathway is given in Figure 20.

5 STORAGE STABILITY

All Extracts were deep frozen after processing and the first chromatographic analysis. However, since most of the HPLC-analyses were repeated using the evaluation method, the storage stability of the extract samples was tested.

The main storage stability test was done using the first replicates (A, C, E and G) of the $CaCl_2$, ambient and aggressive organic extracts taken on DAT-98 (first analysis was performed with the HPLC evaluation method). After a storage period for about 108 days which is somewhat larger than the maximum storage time between the first and second HPLC-analysis of soil extracts (Appendix 10), the extracts were thawed, shaken and analyzed by LSC. For the desorption solutions, HPLC-analysis was possible without any sample preparation. For HPLC-analysis of the ambient and aggressive organic extracts, 5 mL of each sample were transferred into centrifuge tubes, fortified with 20 µL of dobanol and concentrated using a vacuum concentrator for 90 min. The volume of the concentrated samples was determined. The concentrated solutions were transferred into Eppendorf vials and centrifuged (10 min, 16 000 x g); the supernatants were subjected to LSC and HPLC-analysis.

The recovery of radioactivity after the second analysis was on average 100.3% compared to the first analysis which shows that no radioactivity was lost during storage. The recovery of radioactivity after the concentration step was on average 95.8%. The second HPLC-analysis revealed that the test item and the transformation products remained stable during the time of storage. Representative chromatograms of an extract before and after storage are presented in Figure 13.

6 CONCLUSIONS

6.1 Major Outcomes of Study

The data gathered in the current laboratory investigation demonstrated that Clothianidin was moderately fast degraded in all four soils with a DT_{50} of 9.7 to 174.6 days (20°C) according to double first order in parallel or first order multi compartment kinetics.

In the course of the experiment six degradation products were detected and quantified together with the test item. The maximum amounts were 3.0% of AR for TZNG (DAT-120, soil Wellesbourne), 6.8% for MNG (DAT-77, soil Wellesbourne), 10.6% for TZMU (DAT-9, soil Hoefchen am Hohenseh plot 4011), 0.8% for TMG (DAT 28, 49 and 77, soil Hoefchen am Hohenseh plot 4011), 3.5% for NTG (DAT-120, soil Vilobi d'Onyar) and 6.7% of AR for TZFA (DAT-35, soil Hoefchen am Hohenseh plot 4011).

In addition several minor transformation products were observed. Their sum at the different sampling times accounted for ≤ 2.4% (Hoefchen am Hohenseh), ≤ 2.3% (Wellesbourne), ≤ 1.8% (Mas du Coq) and ≤ 2.1% (Vilobi d'Onyar) of the applied radioactivity.

The amount of $^{14}CO_2$ increased over the entire study period. At the end of the study 120 days after application, evolved $^{14}CO_2$ accounted for 29.3%, 16.2%, 9.4%, and 15.9% of the applied RA in soils Hoefchen am Hohenseh plot 4011, Wellesbourne, Mas du Coq, and Vilobi d'Onyar, respectively.

The formation of bound residues increased with the overall metabolism of test compound. The non-extractable residues at DAT-0 varied from 2.2% – 3.0% of AR and were generated during the 24h lasting desorption step (see section 4.2.1). At study termination they increased to 30.8%, 20.2%, 18.1%, and 19.8% of AR in soils Hoefchen am Hohenseh plot 4011, Wellesbourne, Mas du Coq and Vilobi d'Onyar, respectively.
The sorption of the test item to soil increased significantly in the course of the incubation by a factor of 2.6 – 3.7. Thus with time of aging in soil less test item will be available for the environment.

6.2 Significance of Results to Environmental Behavior of Test Compound

The test item Clothianidin will be degraded in soil under aerobic conditions. The DT_{50} in soil is expected to be 65.1 days (geometric mean). The high amount of $^{14}CO_2$ indicates complete mineralization.

7 RETENTION OF RECORDS

All documents/raw data concerning this study are collected and will be filed securely in a central place. After completion of the study these documents and the original report will be stored in the central GLP-Archive of Bayer CropScience AG in D-40789 Monheim, for at least as long as required by GLP-principles. Subsamples of the reference substances are stored at the provider of the substances. No subsample of the radiolabeled test substance will be stored, because of the instability of the substance (autoradiolysis). All samples will be discarded after completion of the final report.

8 REFERENCES

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9 ADDITIONAL INFORMATION

None

List of Tables

Table 1: Physicochemical Characteristics of Test Soils (DER Table 2)

Parameter	**Results/Units**			
Soil / Batch ID	**Hoefchen am Hohenseh plot 4011 (20090903)**	**Wellesbourne (20090918)**	**Mas du Coq (20090831)**	**Vilobi d'Onyar (20090918)**
Soil Taxonomic Classification (USDA)	(N/A)	(N/A)	(N/A)	(N/A)
Soil Series	(N/A)	(N/A)	(N/A)	(N/A)
Texture Class[A]	Silt Loam	Sandy Loam	Clay Loam	Sandy Loam
Sand[A] Silt[A] Clay[A]	22% 54% 24%	72% 12% 16%	20% 50% 30%	54% 32% 14%
pH in water: in $CaCl_2$: in KCl:	5.9 5.7 5.2	6.3 6.1 5.7	7.7 7.7 7.5	6.2 6.1 5.6
Organic Matter[B]	1.9%	1.4%	1.4%	1.4%
Organic Carbon[C]	1.1%	0.8%	0.8%	0.8%
Initial & Final Soil Biomass or Microbial Activity [mg microbial C /kg dry wt]	341	176	129	290
Cation Exchange Capacity (CEC)	12.2 meq/ 100 g	9.3 meq/ 100 g	11.2 meq/ 100 g	8.0 meq/ 100 g
Water Holding Capacity at 0.33 bar	16.3%	7.9%	17.7%	11.1%
WHCmax (g H_2O/ 100 g soil dry wt)	55.1 g	38.9 g	39.9 g	33.5 g
Bulk Density (disturbed)	1.1 g/mL	1.31 g/mL	1.19 g/mL	1.33 g/mL

[A]) According to USDA classification
[B]) Calculated: Organic matter = organic carbon * 1.724
[C]) Determination method: Combustion

Table 2: Experimental Design (DER Table 3)

Parameter		Description
Duration of the test		120 days
Soil condition		Fresh sampled from field, partially air dried
Soil sample weight		100 g (DM)/replicate
Test Conditions	g a.i./ha	150
	µg a.i./kg soil	200 (actual: 199)
Control conditions (if used)		N/A
Number of replications	Treatments	2
	Control	N/A
Test apparatus		Erlenmeyer flask, see Figure 2
Traps for $^{14}CO_2$ & organic volatiles		Solid soda lime traps & polyurethane foam plug, see Figure 2
Test material application	Identity of solvent	Methanol/water 1/1 (v/v)
	Volume of test solution used/treatment	329 µL
	Application method	The soil samples were treated directly as it would happen during a spray application (see 3.4.1)
	Evaporation of application solvent	No
Indication of test material adsorbing to walls of test apparatus		None
Experimental conditions	Temperature (°C)	19.3°C ± 0.2 °C (mean + RSD)
	Moisture content	55% WHCmax
	Moisture maintenance method	Weighing and adding lost water (purified water) on day 74
	Continuous darkness (Yes/No)	Yes
Other details		N/A

Table 3: Sampling Details (DER Table 4)

Parameter		**Description**
Sampling intervals		0, 1, 3, 9, 21, 28, 35, 49, 63, 77, 98 and 120 days post-application
Soil sampling procedures		See Figure 3
Collection of $^{14}CO_2$ and volatile organics		Soda lime & PU (see Figure 2)
Sampling intervals	Moisture content	On day 74
	Redox potential	N/A
	Sterility checks	N/A
	Other	none
Sample storage before analysis		Analysis of extracts by LSC and HPLC (first analysis) within a maximum of six days. During workup periods, samples were stored at room temperature (> 1 day) or in refrigerator (> 1 week). After analysis, samples were deep frozen.
Other observations		N/A

Table 4: Results of Microbial Biomass Determinations (as mg microbial carbon per kg of soil dry wt)

Incubation time	ID	Hoefchen am Hohenseh plot 4011	Wellesbourne	Mas du Coq	Vilobi d'Onyar
Day 0	BIO 1-	341	176	129	290
Day 74	BIO 2- BIO 2+	187 154	86 99	77 66	224 242
Day 120	BIO 3- BIO 3+	283 261	96 114	77 85	202 198

+: with application solution (solvent only)
-: without application solution

Table 5: Material Balance of Radioactivity in Soil Hoefchen am Hohenseh plot 4011 (Replicate A and B), expressed as percent of applied radioactivity

		Replicate No.	DAT 0	1	3	9	21	28	35	49	63	77	98	120
Volatiles														
	$^{14}CO_2$	A	n.a.	0.2	0.8	4.3	12.8	15.1	18.5	21.6	23.6	25.4	28.2	29.4
		B	n.a.	0.3	0.9	4.5	13.5	15.9	18.2	21.3	23.8	25.7	28.1	29.2
		Mean		**0.2**	**0.8**	**4.4**	**13.1**	**15.5**	**18.3**	**21.5**	**23.7**	**25.5**	**28.2**	**29.3**
	Volatile organics	A	n.a.	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	0.3
		B	n.a.	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1
		Mean												**0.17**
	Total	A	n.a.	0.2	0.8	4.3	12.8	15.1	18.5	21.6	23.6	25.4	28.2	29.7
		B	n.a.	0.3	0.9	4.5	13.5	15.9	18.2	21.3	23.8	25.7	28.1	29.2
		Mean		**0.2**	**0.8**	**4.4**	**13.1**	**15.5**	**18.3**	**21.5**	**23.7**	**25.5**	**28.2**	**29.5**
Extractable Radioactivity														
	Calcium chloride solution	A	22.7	21.6	21.0	13.4	8.6	8.0	7.2	6.3	5.8	5.9	5.3	4.9
		B	23.5	21.3	19.3	13.7	9.2	7.8	7.4	6.3	6.1	5.8	5.1	5.1
		Mean	**23.1**	**21.5**	**20.2**	**13.6**	**8.9**	**7.9**	**7.3**	**6.3**	**6.0**	**5.8**	**5.2**	**5.0**
	Ambient organic extract	A	73.7	68.1	65.5	52.8	40.6	40.6	36.3	32.5	30.3	29.3	26.7	25.3
		B	74.7	68.7	65.3	54.3	40.9	39.8	38.0	33.4	31.5	30.7	26.9	26.0
		Mean	**74.2**	**68.4**	**65.4**	**53.6**	**40.7**	**40.2**	**37.1**	**32.9**	**30.9**	**30.0**	**26.8**	**25.6**
	Aggressive organic extract	A	1.7	2.1	3.7	4.3	5.6	5.6	5.4	5.9	6.5	6.8	6.4	6.8
		B	1.7	2.1	3.6	4.2	5.6	5.7	5.4	6.0	6.6	7.0	6.5	7.3
		Mean	**1.7**	**2.1**	**3.7**	**4.3**	**5.6**	**5.7**	**5.4**	**5.9**	**6.6**	**6.9**	**6.4**	**7.0**
	Total	A	98.0	91.8	90.2	70.5	54.8	54.1	48.9	44.7	42.6	42.0	38.4	37.0
		B	99.9	92.2	88.2	72.2	55.7	53.4	50.8	45.7	44.2	43.4	38.5	38.4
		Mean	**99.0**	**92.0**	**89.2**	**71.4**	**55.2**	**53.8**	**49.8**	**45.2**	**43.4**	**42.7**	**38.4**	**37.7**
Bound Residue		A	2.9	6.3	9.1	22.6	30.5	29.5	30.2	30.0	30.9	31.2	30.5	30.8
		B	3.0	5.8	10.1	22.3	28.8	29.7	29.4	30.0	30.7	31.9	31.0	30.7
		Mean	**3.0**	**6.1**	**9.6**	**22.5**	**29.6**	**29.6**	**29.8**	**30.0**	**30.8**	**31.5**	**30.7**	**30.8**
Material Balance		A	100.9	98.4	100.1	97.4	98.0	98.8	97.5	96.3	97.2	98.6	97.1	97.5
		B	103.0	98.3	99.2	99.0	97.9	99.0	98.5	97.0	98.7	101.0	97.6	98.3
		Mean	**101.9**	**98.3**	**99.7**	**98.2**	**98.0**	**98.9**	**98.0**	**96.7**	**97.9**	**99.8**	**97.3**	**97.9**

n.a.: not analyzed, DAT: days after treatment

Material Balance	**Min**	**96.7**
	Max	**101.9**
	Mean	**98.6**
	rel. standard deviation	**1.4%**

Table 6: Material Balance of Radioactivity in Soil Wellesbourne (Replicate C and D), expressed as percent of applied radioactivity

		Replicate No.	DAT 0	1	3	9	21	28	35	49	63	77	98	120
Volatiles														
	$^{14}CO_2$	C	n.a.	0.2	0.3	2.0	5.8	7.3	8.2	10.404	12.2	13.2	15.0	16.1
		D	n.a.	0.2	0.4	2.0	6.0	7.2	8.2	10.9	12.3	12.9	14.5	16.4
		Mean		**0.2**	**0.3**	**2.0**	**5.9**	**7.3**	**8.2**	**10.6**	**12.3**	**13.1**	**14.8**	**16.2**
	Volatile organics	C	n.a.	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1
		D	n.a.	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1
		Mean												
	Total	C	n.a.	0.2	0.3	2.0	5.8	7.3	8.2	10.4	12.2	13.2	15.0	16.1
		D	n.a.	0.2	0.4	2.0	6.0	7.2	8.2	10.9	12.3	12.9	14.5	16.4
		Mean		**0.2**	**0.3**	**2.0**	**5.9**	**7.3**	**8.2**	**10.6**	**12.3**	**13.1**	**14.8**	**16.2**
Extractable Radioactivity														
	Calcium chloride solution	C	31.4	30.5	30.4	23.8	20.2	18.5	17.5	16.2	15.2	15.2	13.2	12.7
		D	32.9	30.7	29.9	24.3	20.5	18.4	17.8	16.3	16.1	15.6	13.4	12.5
		Mean	**32.1**	**30.6**	**30.2**	**24.0**	**20.3**	**18.5**	**17.6**	**16.3**	**15.7**	**15.4**	**13.3**	**12.6**
	Ambient org. extract	C	69.9	63.1	60.3	59.9	54.3	53.5	52.6	49.0	46.0	46.1	43.0	40.6
		D	67.8	62.1	62.9	59.9	53.8	52.6	52.5	50.4	47.2	45.5	42.8	40.6
		Mean	**68.8**	**62.6**	**61.6**	**59.9**	**54.0**	**53.1**	**52.6**	**49.7**	**46.6**	**45.8**	**42.9**	**40.6**
	Aggressive org. extract	C	1.1	1.5	2.7	3.7	5.0	5.5	5.3	6.1	6.7	7.0	7.3	8.4
		B	1.0	1.7	2.8	3.1	5.1	4.6	5.2	6.3	6.7	7.4	7.9	8.8
		Mean	**1.1**	**1.6**	**2.8**	**3.4**	**5.1**	**5.0**	**5.2**	**6.2**	**6.7**	**7.2**	**7.6**	**8.6**
	Total	C	102.4	95.1	93.4	87.5	79.4	77.5	75.4	71.3	67.9	68.3	63.5	61.8
		D	101.6	94.5	95.6	87.2	79.4	75.6	75.5	73.0	70.0	68.5	64.1	61.9
		Mean	**102.0**	**94.8**	**94.5**	**87.4**	**79.4**	**76.5**	**75.5**	**72.2**	**68.9**	**68.4**	**63.8**	**61.8**
Bound Residue		C	2.1	3.6	4.6	10.8	14.4	15.1	15.9	17.2	18.8	19.5	20.0	20.3
		D	2.3	3.3	4.9	10.9	14.8	16.0	16.2	16.9	18.8	19.3	19.8	20.1
		Mean	**2.2**	**3.5**	**4.7**	**10.8**	**14.6**	**15.5**	**16.0**	**17.0**	**18.8**	**19.4**	**19.9**	**20.2**
Material Balance		C	104.5	98.9	98.3	100.3	99.6	99.9	99.6	99.0	98.9	101.0	98.5	98.1
		D	104.0	98.0	100.8	100.1	100.3	98.8	99.9	100.7	101.0	100.6	98.4	98.4
		Mean	**104.2**	**98.4**	**99.6**	**100.2**	**99.9**	**99.3**	**99.7**	**99.8**	**100.0**	**100.8**	**98.5**	**98.2**

n.a.: not analyzed, DAT: days after treatment

Material Balance	**Min**	**98.2**
	Max	**104.2**
	Mean	**99.9**
	rel. standard deviation	**1.6%**

Table 7: Material Balance of Radioactivity in Soil Mas du Coq (Replicate E and F), expressed as percent of applied radioactivity

		Replicate No.	DAT 0	1	3	9	21	28	35	49	63	77	98	120
Volatiles														
	$^{14}CO_2$	E	n.a.	0.1	0.2	0.7	2.0	3.2	3.5	4.8	5.8	6.9	8.2	9.1
		F	n.a.	0.1	0.2	0.7	2.1	2.9	3.5	4.9	5.8	6.8	8.4	9.6
		Mean		**0.1**	**0.2**	**0.7**	**2.0**	**3.0**	**3.5**	**4.8**	**5.8**	**6.8**	**8.3**	**9.4**
	Volatile organics	E	n.a.	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1
		F	n.a.	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1
		Mean												
	Total	E	n.a.	0.1	0.2	0.7	2.0	3.2	3.5	4.8	5.8	6.9	8.2	9.1
		F	n.a.	0.1	0.2	0.7	2.1	2.9	3.5	4.9	5.8	6.8	8.4	9.6
		Mean		**0.1**	**0.2**	**0.7**	**2.0**	**3.0**	**3.5**	**4.8**	**5.8**	**6.8**	**8.3**	**9.4**
Extractable Radioactivity														
	Calcium chloride solution	E	37.0	34.4	31.7	28.8	25.1	23.2	22.4	21.6	19.6	20.5	17.2	16.5
		F	37.2	35.6	31.3	29.5	25.0	23.2	23.1	21.7	19.8	19.8	18.0	16.8
		Mean	**37.1**	**35.0**	**31.5**	**29.2**	**25.0**	**23.2**	**22.8**	**21.6**	**19.7**	**20.1**	**17.6**	**16.7**
	Ambient organic extract	E	62.4	59.9	60.4	60.3	56.4	56.6	55.8	54.4	52.9	52.7	48.9	47.4
		F	63.3	61.6	61.2	60.5	57.3	55.9	56.4	54.1	52.5	51.7	49.0	46.7
		Mean	**62.8**	**60.7**	**60.8**	**60.4**	**56.8**	**56.3**	**56.1**	**54.3**	**52.7**	**52.2**	**48.9**	**47.0**
	Aggressive Organic extract	E	1.1	1.5	2.8	3.6	4.8	5.5	5.5	5.6	7.1	8.1	7.9	7.6
		F	1.1	1.5	2.7	3.7	5.0	5.3	5.2	5.6	7.1	7.5	7.5	7.5
		Mean	**1.1**	**1.5**	**2.7**	**3.7**	**4.9**	**5.4**	**5.4**	**5.6**	**7.1**	**7.8**	**7.7**	**7.6**
	Total	E	100.5	95.9	94.9	92.7	86.3	85.3	83.7	81.7	79.6	81.3	74.0	71.5
		F	101.5	98.7	95.2	93.7	87.3	84.4	84.8	81.4	79.4	79.0	74.5	71.0
		Mean	**101.0**	**97.3**	**95.0**	**93.2**	**86.8**	**84.8**	**84.2**	**81.6**	**79.5**	**80.2**	**74.2**	**71.2**
Bound Residue		E	2.2	2.9	4.3	6.4	9.6	10.2	11.2	12.2	13.8	15.4	16.5	18.0
		F	2.6	3.0	4.1	6.4	9.7	10.3	11.2	12.2	14.0	15.5	16.5	18.1
		Mean	**2.4**	**2.9**	**4.2**	**6.4**	**9.7**	**10.2**	**11.2**	**12.2**	**13.9**	**15.5**	**16.5**	**18.1**
Material Balance		E	102.6	98.8	99.4	99.8	97.9	98.6	98.4	98.6	99.1	103.6	98.8	98.6
		F	104.1	101.8	99.5	100.8	99.1	97.6	99.5	98.6	99.3	101.3	99.4	98.7
		Mean	**103.4**	**100.3**	**99.4**	**100.3**	**98.5**	**98.1**	**99.0**	**98.6**	**99.2**	**102.5**	**99.1**	**98.7**

n.a.: not analyzed, DAT: days after treatment

Material Balance	**Min**	**98.1**
	Max	**103.4**
	Mean	**99.7**
	rel. standard deviation	**1.6%**

Table 8: Material Balance of Radioactivity in Soil Vilobi d'Onyar (Replicate G and H), expressed as percent of applied radioactivity

		Replicate	DAT											
		No.	0	1	3	9	21	28	35	49	63	77	98	120
Volatiles														
	$^{14}CO_2$	G	n.a.	0.2	0.3	1.7	5.0	6.6	7.9	9.7	11.3	12.8	14.7	15.6
		H	n.a.	0.2	0.3	1.7	5.2	6.7	7.9	10.1	11.6	13.0	14.6	16.2
		Mean		**0.2**	**0.3**	**1.7**	**5.1**	**6.6**	**7.9**	**9.9**	**11.4**	**12.9**	**14.7**	**15.9**
	Volatile organics	G	n.a.	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1
		H	n.a.	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1
		Mean												
	Total	G	n.a.	0.2	0.3	1.7	5.0	6.6	7.9	9.7	11.3	12.8	14.7	15.6
		H	n.a.	0.2	0.3	1.7	5.2	6.7	7.9	10.1	11.6	13.0	14.6	16.2
		Mean		**0.2**	**0.3**	**1.7**	**5.1**	**6.6**	**7.9**	**9.9**	**11.4**	**12.9**	**14.7**	**15.9**
Extractable Radioactivity														
	Calcium chloride solution	G	34.2	32.1	30.6	25.4	20.5	19.6	18.3	17.1	15.2	16.2	14.3	12.7
		H	34.4	32.0	29.3	25.6	20.3	19.0	18.5	17.7	16.2	15.9	14.3	13.6
		Mean	**34.3**	**32.0**	**29.9**	**25.5**	**20.4**	**19.3**	**18.4**	**17.4**	**15.7**	**16.0**	**14.3**	**13.1**
	Ambient organic extract	G	64.5	63.3	61.5	60.9	53.9	54.4	52.2	51.2	48.2	46.4	42.0	41.1
		H	66.3	62.2	62.0	61.0	53.5	54.1	52.4	51.4	48.6	46.1	42.5	41.4
		Mean	**65.4**	**62.8**	**61.8**	**60.9**	**53.7**	**54.2**	**52.3**	**51.3**	**48.4**	**46.3**	**42.2**	**41.3**
	Aggressive organic extract	G	1.0	1.7	2.7	4.2	4.7	6.3	6.7	6.6	9.1	9.0	9.3	9.2
		H	1.4	1.7	2.5	4.3	5.3	6.3	6.9	6.1	8.4	9.1	8.4	9.2
		Mean	**1.2**	**1.7**	**2.6**	**4.3**	**5.0**	**6.3**	**6.8**	**6.4**	**8.8**	**9.1**	**8.9**	**9.2**
	Total	G	99.6	97.1	94.8	90.4	79.1	80.3	77.2	74.9	72.6	71.7	65.6	63.0
		H	102.1	96.0	93.9	90.9	79.1	79.4	77.8	75.3	73.2	71.1	65.2	64.2
		Mean	**100.9**	**96.5**	**94.3**	**90.7**	**79.1**	**79.8**	**77.5**	**75.1**	**72.9**	**71.4**	**65.4**	**63.6**
Bound Residue		G	2.7	3.3	5.0	9.1	14.0	13.7	14.1	15.5	16.3	17.6	18.8	20.0
		H	1.9	3.1	5.6	8.6	13.5	13.3	14.6	14.8	16.2	17.7	19.4	19.7
		Mean	**2.3**	**3.2**	**5.3**	**8.8**	**13.8**	**13.5**	**14.3**	**15.1**	**16.3**	**17.7**	**19.1**	**19.8**
Material Balance		G	102.3	100.5	100.2	101.3	98.2	100.7	99.1	100.0	100.2	102.0	99.2	98.6
		H	103.9	99.2	99.8	101.2	97.9	99.3	100.3	100.2	101.0	101.8	99.2	100.1
		Mean	**103.1**	**99.9**	**100.0**	**101.2**	**98.0**	**100.0**	**99.7**	**100.1**	**100.6**	**101.9**	**99.2**	**99.3**

n.a.: not analyzed, DAT: days after treatment

Material Balance	**Min**	**98.0**
	Max	**103.1**
	Mean	**100.3**
	rel. standard deviation	**1.3%**

Table 9: Biotransformation of Clothianidin, Expressed as Percentage of Applied Radioactivity (mean ± s.d.), in Soil Hoefchen am Hohenseh plot 4011, Replicate A and B (DER Table 5)

		DAT											
Compound		**0**	**1**	**3**	**9**	**21**	**28**	**35**	**49**	**63**	**77**	**98**	**120**
Clothianidin	Mean	96.9	86.0	77.8	51.1	36.9	36.6	30.9	29.4	26.9	26.9	22.2	21.5
	SD	±0.6	±0.6	±0.6	±0.0	±1.3	±1.1	±1.1	±0.1	±0.6	±0.6	±0.2	±1.1
TZNG	Mean	n.d.	n.d.	0.0	0.1	0.5	0.5	0.6	0.8	0.8	1.0	1.0	1.1
	SD			±0.0	±0.1	±0.0	±0.0	±0.1	±0.1	±0.1	±0.0	±0.1	±0.0
MNG	Mean	n.d.	0.5	1.3	2.5	3.1	3.6	4.7	4.8	4.8	5.6	4.5	6.4
	SD		±0.1	±0.2	±0.1	±0.0	±0.0	±0.1	±0.4	±0.3	±0.3	±0.7	±0.0
TZMU	Mean	0.5	2.3	6.2	10.6	6.9	5.4	4.2	2.9	2.2	2.1	1.4	1.2
	SD	±0.0	±0.0	±0.3	±0.0	±0.4	±0.0	±0.1	±0.1	±0.3	±0.0	±0.0	±0.0
TMG	Mean	n.d.	0.1	0.3	0.6	0.6	0.8	0.8	0.7	0.6	0.8	0.6	0.5
	SD		±0.1	±0.0	±0.0	±0.0	±0.0	±0.1	±0.2	±0.0	±0.1	±0.1	±0.1
NTG	Mean	n.d.	0.1	<LOQ	0.0	0.2	0.2	0.8	0.6	1.5	1.5	2.8	2.2
	SD		±0.0		±0.0	±0.0	±0.0	±0.0	±0.1	±0.1	±0.2	±0.6	±0.1
TZFA	Mean	n.d.	0.8	1.8	4.2	5.3	4.9	6.7	4.3	5.0	4.0	4.6	3.8
	SD		±0.0	±0.0	±0.5	±0.0	±0.8	±0.1	±0.6	±0.5	±0.1	±0.3	±0.1
Unidentified	Mean	1.6	2.2	1.8	2.4	1.9	1.7	1.3	1.7	1.5	0.8	1.4	1.1
radioactivity	SD	±0.4	±0.1	±0.1	±0.2	±0.6	±0.1	±0.0	±0.3	±0.3	±0.0	±0.3	±0.1
Total	Mean	99.0	92.0	89.2	71.4	55.2	53.8	49.8	45.2	43.4	42.7	38.4	37.7
extractable res.	SD	±0.9	±0.2	±1.0	±0.9	±0.4	±0.4	±1.0	±0.5	±0.8	±0.7	±0.0	±0.7
$^{14}CO_2$	Mean	n.a.	0.2	0.8	4.4	13.1	15.5	18.3	21.5	23.7	25.5	28.2	29.3
	SD		±0.0	±0.0	±0.1	±0.3	±0.4	±0.1	±0.1	±0.1	±0.1	±0.1	±0.1
Volatile	Mean	n.a.	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	0.2
organics	SD												±0.2
Non-extractable	Mean	3.0	6.1	9.6	22.5	29.6	29.6	29.8	30.0	30.8	31.5	30.7	30.8
residues	SD	±0.1	±0.3	±0.5	±0.1	±0.8	±0.1	±0.4	±0.0	±0.1	±0.4	±0.3	±0.1
Total %	Mean	101.9	98.3	99.6	98.2	98.0	98.9	98.0	96.7	97.9	99.8	97.3	97.9
recovery	SD	±1.0	±0.1	±0.5	±0.8	±0.1	±0.1	±0.5	±0.3	±0.8	±1.2	±0.2	±0.2

n.d. : not detected, n.a. : not analyzed, DAT : day after treatment, SD : standard deviation

Table 10: Biotransformation of Clothianidin, Expressed as Percentage of Applied Radioactivity (mean ± s.d.), in Soil Wellesbourne, Replicate C and D (DER Table 5)

Compound	Replicate No.	DAT 0	1	3	9	21	28	35	49	63	77	98	120
Clothianidin	Mean	99.8	91.7	88.6	75.8	64.5	64.7	61.6	58.5	53.8	53.2	48.4	46.2
	SD	±0.7	±0.3	±0.8	±0.3	±1.5	±0.4	±0.0	±0.2	±1.3	±0.1	±0.4	±0.2
TZNG	Mean	n.d.	0.0	0.0	0.2	1.1	1.7	2.0	2.1	2.3	2.7	2.9	3.0
	SD		±0.0	±0.0	±0.1	±0.1	±0.0	±0.0	±0.1	±0.0	±0.1	±0.1	±0.3
MNG	Mean	n.d.	0.6	0.8	2.2	4.0	3.9	4.7	5.7	5.4	6.8	5.6	6.4
	SD		±0.2	±0.1	±0.0	±0.3	±0.3	±0.0	±0.2	±0.1	±0.4	±0.5	±0.2
TZMU	Mean	n.d.	0.9	3.0	5.9	5.1	3.0	2.5	2.1	1.7	1.5	1.0	1.0
	SD		±0.0	±0.3	±0.2	±0.9	±0.3	±0.0	±0.0	±0.1	±0.0	±0.0	±0.1
TMG	Mean	n.d.	0.0	0.1	0.1	0.2	0.3	0.3	0.1	0.1	n.d.	0.2	0.1
	SD		±0.0	±0.1	±0.1	±0.0	±0.0	±0.0	±0.1	±0.1		±0.1	±0.0
NTG	Mean	n.d.	n.d.	0.1	0.1	0.1	0.3	0.6	1.2	1.7	1.4	2.5	3.1
	SD			±0.1	±0.1	±0.0	±0.2	±0.0	±0.2	±0.2	±0.1	±0.1	±0.4
TZFA	Mean	n.d.	n.d.	0.4	1.9	2.8	2.1	2.7	2.0	2.6	1.9	1.9	1.5
	SD			±0.3	±0.0	±0.4	±0.3	±0.2	±0.2	±0.2	±0.1	±0.2	±0.2
Unidentified	Mean	2.3	1.6	1.5	1.1	1.5	0.5	1.0	0.4	1.3	0.7	1.3	0.5
radioactivity	SD	±0.3	±0.2	±0.6	±0.0	±0.0	±0.2	±0.2	±0.1	±0.2	±0.3	±0.2	±0.1
Total	Mean	102.0	94.8	94.5	87.4	79.4	76.5	75.5	72.1	68.9	68.4	63.8	61.8
extractable res.	SD	±0.4	±0.3	±1.1	±0.1	±0.0	±0.9	±0.0	±0.8	±1.0	±0.1	±0.3	±0.1
$^{14}CO_2$	Mean	n.a.	0.2	0.3	2.0	5.9	7.3	8.2	10.6	12.3	13.1	14.8	16.2
	SD		±0.0	±0.0	±0.0	±0.1	±0.1	±0.0	±0.2	±0.0	±0.2	±0.2	±0.2
Volatile	Mean	n.a.	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1
organics	SD												
Non-extractable	Mean	2.2	3.5	4.7	10.8	14.6	15.5	16.0	17.0	18.8	19.4	19.9	20.2
residues	SD	±0.1	±0.2	±0.2	±0.0	±0.2	±0.4	±0.2	±0.2	±0.0	±0.1	±0.1	±0.1
Total %	Mean	104.2	98.4	99.6	100.2	99.9	99.3	99.7	99.8	100.0	100.8	98.5	98.2
recovery	SD	±0.3	±0.4	±1.2	±0.1	±0.3	±0.6	±0.2	±0.9	±1.0	±0.2	±0.3	±0.1

n.d. : not detected, n.a. : not analyzed, DAT : day after treatment, SD : standard deviation

Table 11: Biotransformation of Clothianidin, Expressed as Percentage of Applied Radioactivity (mean ± s.d.), in Soil Mas Du Coq, Replicate E and F (DER Table 5)

		DAT											
Compound		**0**	**1**	**3**	**9**	**21**	**28**	**35**	**49**	**63**	**77**	**98**	**120**
Clothianidin	Mean	99.2	94.4	91.0	87.8	80.6	77.6	75.8	72.6	70.2	70.1	63.4	60.8
	SD	±0.3	±0.4	±0.8	±0.6	±1.2	±0.5	±0.2	±0.8	±0.6	±0.7	±0.7	±0.5
TZNG	Mean	n.d.	n.d.	n.d.	0.1	0.7	1.0	0.9	1.3	1.6	1.8	2.0	2.1
	SD				±0.0	±0.1	±0.0	±0.0	±0.2	±0.1	±0.2	±0.2	±0.1
MNG	Mean	n.d.	0.4	0.3	0.9	2.2	2.5	3.5	4.0	2.9	5.2	3.9	5.3
	SD		±0.1	±0.3	±0.0	±0.1	±0.4	±0.1	±0.0	±0.0	±0.7	±0.5	±0.2
TZMU	Mean	n.d.	0.8	1.8	2.2	1.8	1.8	1.8	1.7	1.6	1.5	1.5	1.2
	SD		±0.2	±0.0	±0.2	±0.2	±0.0	±0.0	±0.0	±0.1	±0.1	±0.2	±0.2
TMG	Mean	n.d.	n.d.	0.0	0.0	0.1	0.0	0.2	0.1	0.2	0.1	0.3	0.2
	SD			±0.0	±0.0	±0.1	±0.0	±0.1	±0.1	±0.2	±0.1	±0.0	±0.1
NTG	Mean	n.d.	0.2	0.1	0.1	n.d.	0.2	0.2	0.2	2.1	0.2	2.1	0.9
	SD		±0.2	±0.1	±0.1		±0.2	±0.2	±0.2	±0.0	±0.1	±0.2	±0.0
TZFA	Mean	n.d.	n.d.	0.1	0.7	0.4	0.4	0.6	0.2	0.1	n.d.	0.3	0.0
	SD			±0.1	±0.1	±0.1	±0.0	±0.1	±0.0	±0.1		±0.1	±0.0
Unidentified	Mean	1.8	1.6	1.8	1.2	1.0	1.4	1.3	1.3	0.9	1.2	0.8	0.8
radioactivity	SD	±0.2	±0.5	±0.2	±0.2	±0.2	±0.4	±0.5	±0.4	±0.4	±0.3	±0.0	±0.0
Total	Mean	101.0	97.3	95.0	93.2	86.8	84.8	84.2	81.6	79.5	80.1	74.2	71.2
extractable res.	SD	±0.5	±1.4	±0.2	±0.5	±0.5	±0.5	±0.5	±0.1	±0.1	±1.2	±0.2	±0.2
$^{14}CO_2$	Mean	n.a.	0.1	0.2	0.7	2.0	3.0	3.5	4.8	5.8	6.8	8.3	9.4
	SD		±0.0	±0.0	±0.0	±0.1	±0.1	±0.0	±0.0	±0.0	±0.0	±0.1	±0.2
Volatile	Mean	n.a.	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1
organics	SD												
Non-extractable	Mean	2.4	2.9	4.2	6.4	9.7	10.2	11.2	12.2	13.9	15.5	16.5	18.1
residues	SD	±0.2	±0.1	±0.1	±0.0	±0.0	±0.1	±0.0	±0.0	±0.1	±0.0	±0.0	±0.1
Total %	Mean	103.4	100.3	99.4	100.3	98.5	98.1	99.0	98.6	99.2	102.4	99.1	98.7
recovery	SD	±0.7	±1.5	±0.1	±0.5	±0.6	±0.5	±0.5	±0.0	±0.1	±1.2	±0.5	±0.1

n.d. : not detected, n.a. : not analyzed, DAT : day after treatment, SD : standard deviation

Table 12: Biotransformation of Clothianidin, Expressed as Percentage of Applied Radioactivity (mean ± s.d.), in Soil Vilobi d'Onyar, Replicate G and H (DER Table 5)

		DAT											
Compound		**0**	**1**	**3**	**9**	**21**	**28**	**35**	**49**	**63**	**77**	**98**	**120**
Clothianidin	Mean	99.1	93.5	89.3	81.7	71.1	70.3	67.9	64.0	61.5	59.5	53.5	51.6
	SD	±0.9	±0.6	±1.5	±0.7	±0.1	±0.1	±0.7	±0.4	±0.1	±0.7	±0.1	±0.5
TZNG	Mean	n.d.	0.0	n.d.	0.2	0.7	0.9	1.1	1.7	1.5	1.9	1.7	1.9
	SD		±0.0		±0.0	±0.1	±0.1	±0.1	±0.2	±0.1	±0.0	±0.0	±0.2
MNG	Mean	n.d.	0.2	0.6	1.5	2.4	3.0	3.6	4.0	2.9	4.3	4.5	3.8
	SD		±0.1	±0.0	±0.1	±0.1	±0.1	±0.4	±0.1	±0.3	±0.0	±0.1	±0.1
TZMU	Mean	0.1	0.7	2.5	4.5	2.9	2.4	2.1	2.0	1.7	1.5	1.2	1.1
	SD	±0.1	±0.0	±0.5	±0.2	±0.2	±0.1	±0.2	±0.1	±0.3	±0.1	±0.0	±0.1
TMG	Mean	n.d.	n.d.	0.1	0.1	0.2	0.2	0.3	0.3	0.4	<LOQ	0.1	0.2
	SD			±0.1	±0.1	±0.0	±0.1	±0.0	±0.1	±0.1		±0.0	±0.1
NTG	Mean	n.d.	n.d.	0.1	0.0	0.1	0.4	0.9	1.3	3.2	2.1	2.8	3.5
	SD			±0.1	±0.0	±0.1	±0.1	±0.2	±0.1	±0.2	±0.1	±0.1	±0.1
TZFA	Mean	n.d.	n.d.	0.3	1.1	1.0	1.2	1.0	1.3	1.2	0.7	1.2	0.9
	SD			±0.1	±0.2	±0.1	±0.1	±0.0	±0.2	±0.1	±0.0	±0.1	±0.1
Unidentified	Mean	1.6	2.1	1.5	1.5	0.8	1.5	0.6	0.5	0.6	1.5	0.4	0.7
radioactivity	SD	±0.2	±0.1	±0.4	±0.1	±0.1	±0.1	±0.0	±0.1	±0.2	±0.3	±0.1	±0.2
Total	Mean	100.9	96.5	94.3	90.7	79.1	79.8	77.5	75.1	72.9	71.4	65.4	63.6
extractable res.	SD	±1.2	±0.6	±0.5	±0.3	±0.0	±0.5	±0.3	±0.2	±0.3	±0.3	±0.2	±0.6
$^{14}CO_2$	Mean	n.a.	0.2	0.3	1.7	5.1	6.6	7.9	9.9	11.4	12.9	14.7	15.9
	SD		±0.0	±0.0	±0.0	±0.1	±0.0	±0.0	±0.2	±0.1	±0.1	±0.1	±0.3
Volatile	Mean	n.a.	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1
organics	SD												
Non-extractable	Mean	2.3	3.2	5.3	8.8	13.8	13.5	14.3	15.1	16.3	17.7	19.1	19.8
residues	SD	±0.4	±0.1	±0.3	±0.3	±0.3	±0.2	±0.2	±0.3	±0.0	±0.1	±0.3	±0.2
Total %	Mean	103.1	99.9	100.0	101.2	98.0	100.0	99.7	100.1	100.6	101.9	99.2	99.3
recovery	SD	±0.8	±0.7	±0.2	±0.0	±0.2	±0.7	±0.6	±0.1	±0.4	±0.1	±0.5	±0.7

n.d. : not detected, n.a. : not analyzed, DAT : day after treatment, SD : standard deviation

Table 13: R_{TDS} -Values of Time-Dependent Sorption

R_{TDS} values [mL/g], mean of duplicates

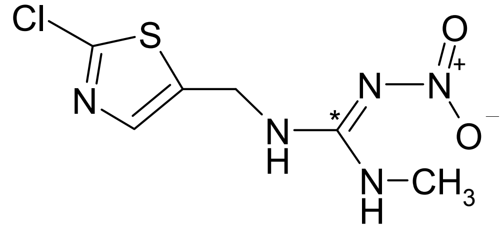
DAT	Hoefchen am Hohenseh plot 4011 (20090903)	Wellesbourne (20090918)	Mas du Coq (20090831)	Vilobi d'Onyar (20090918)
0	1.3	0.9	0.6	0.8
1	1.4	0.9	0.6	0.8
3	1.6	1.1	0.8	0.9
9	2.3	1.4	0.9	1.3
21	2.4	1.9	1.1	1.4
28	3.3	1.7	1.2	1.7
35	3.1	1.7	1.2	1.7
49	3.1	2.0	1.3	1.8
63	3.5	2.1	1.3	2.0
77	4.2	2.2	1.5	2.1
98	5.1	2.3	1.7	2.1
120	4.8	2.5	1.6	2.5
Factor: $\frac{R_{TDS}\ \text{DAT-120}}{R_{TDS}\ \text{DAT-0}}$	3.7	2.7	2.6	3.1
Mean	3.0			

For individual single values see Appendix 16 to Appendix 19.

List of Figures

Figure 1: Chemical Nomenclature and Structure of the Test and Reference Items (DER Figure 2)

Test Item

Common Name:	**Clothianidin**	
Chemical Code:	TI 435	
CAS Nomenclature:	(E)-N-[(2-chloro-5-thiazolyl)methyl]-N'-methyl-N''-nitroguanidine	
IUPAC Name	(E)-1-(2-chloro-1,3-thiazol-5-ylmethyl)-3-methyl-2-nitroguanidine	
CAS Number:	210889-92-5	
Molecular Weight:	249.7 g/mol	
		*: ^{14}C-labelling position of the test item
SMILES Codes:	s1c(CL) ncc1CNC (=NN (=O) (=O)) NC	

Reference Items

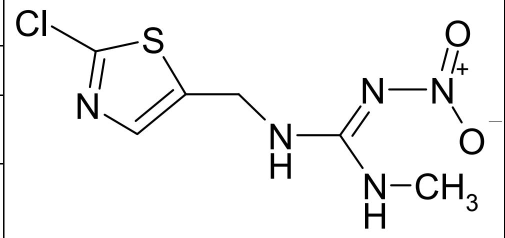
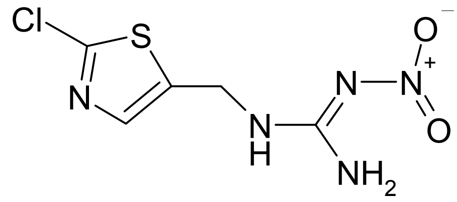
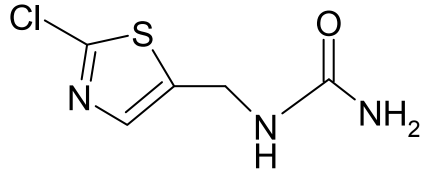
Report name:	**Clothianidin**	
Sample ID:	A1283742	
Batch:	AE1283742 00 1B99 0001	
CAS nomenclature:	(E)-N-[(2-chloro-5-thiazolyl)methyl]-N'-methyl-N''-nitroguanidine	
IUPAC Name	(E)-1-(2-chloro-1,3-thiazol-5-ylmethyl)-3-methyl-2-nitroguanidine	
CAS number:	210889-92-5	
Molecular weight:	249.7 g/mol	
Formula:	$C_6H_8ClN_5O_2$ S	
SMILES code:	s1c(CL) ncc1CNC (=NN (=O) (=O)) NC	

Figure 1: Chemical Nomenclature and Structure of the Test and Reference Items (DER Figure 2), continued

Reference Items

Report name:	**TZNG**	
Sample ID:	FHW0107C	
Batch:	TZNG	
IUPAC nomenclature:	Not available	
CAS number:	Not available	
Molecular weight:	235.65 g x mol^{-1}	
Formula:	$C_5H_6O_2N_5ClS$	
SMILES code:	s1c(CL) ncc1CNC (N) = NN(=O) (=O)	

Report name:	**TZU**	
Sample ID:	FHW0107D	
Batch:	TZU	
IUPAC nomenclature:	Not available	
CAS number:	Not available	
Molecular weight:	191.64 g x mol^{-1}	
Formula:	$C_5H_6N_3OClS$	
SMILES code:	s1c(CL) ncc1CNC (=O) (N)	

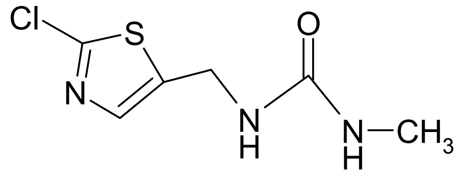
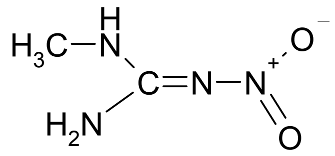
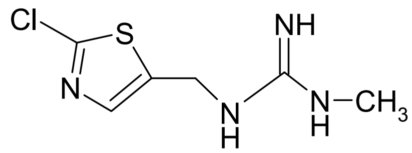
Report name:	**TZMU**	
Sample ID:	FHW0107E	
Batch:	TZMU	
IUPAC nomenclature:	Not available	
CAS number:	Not available	
Molecular weight:	205.66 g x mol^{-1}	
Formula:	$C_6H_8N_3OClS$	
SMILES code:	s1c(CL) ncc1CNC (=O)NC	

Figure 1: Chemical Nomenclature and Structure of the Test and Reference Items (DER Figure 2), continued

Report name:	**MNG**	
Sample ID:	FHW0107F	
Batch:	MNG	
IUPAC nomenclature:	Not available	
CAS number:	Not available	
Molecular weight:	118.10 g x mol^{-1}	
Formula:	$C_2H_6N_4O_2$	
SMILES code:	C(N)(=NN(=O) (=O)) NC	

Report name:	**TMG**	
Sample ID:	FHW0107G	
Batch:	TMG	
IUPAC nomenclature:	Not available	
CAS number:	Not available	
Molecular weight:	204.68 g x mol^{-1}	
Formula:	$C_6H_9N_4ClS$	
SMILES code:	s1c(CL) ncc1CNC (=N) NC	

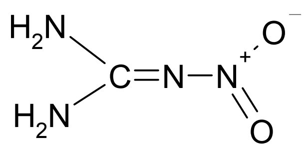
Report name:	**NTG**	
Sample ID:	FHW0107H	
Batch:	NTG	
IUPAC nomenclature:	Not available	
CAS number:	Not available	
Molecular weight:	104.07 g x mol^{-1}	
Formula:	$CH_4N_4O_2$	
SMILES code:	C(N)(N)=NN(=O)(=O)	

Figure 1: Chemical Nomenclature and Structure of the Test and Reference Items (DER Figure 2), continued

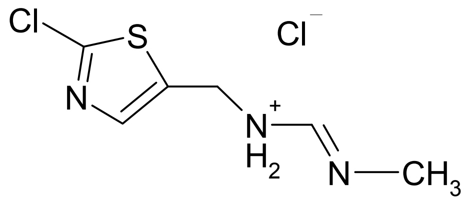
Report name:	**TZFA**	
Sample ID:	BCS-CQ81511	
Batch:	KATH 15179-4-5	
IUPAC nomenclature:	Not available	
CAS number:	Not available	
Molecular weight:	226.13 g x mol^{-1}	
Formula:	$C_6H_8ClN_3S_{1.}$ Cl	Mixture of E/Z isomers
SMILES code:	S1C(Cl)=NC=C1CNC=NC_Cl	

Figure 2: Incubation Vessel for Aerobic Soil Degradation Study

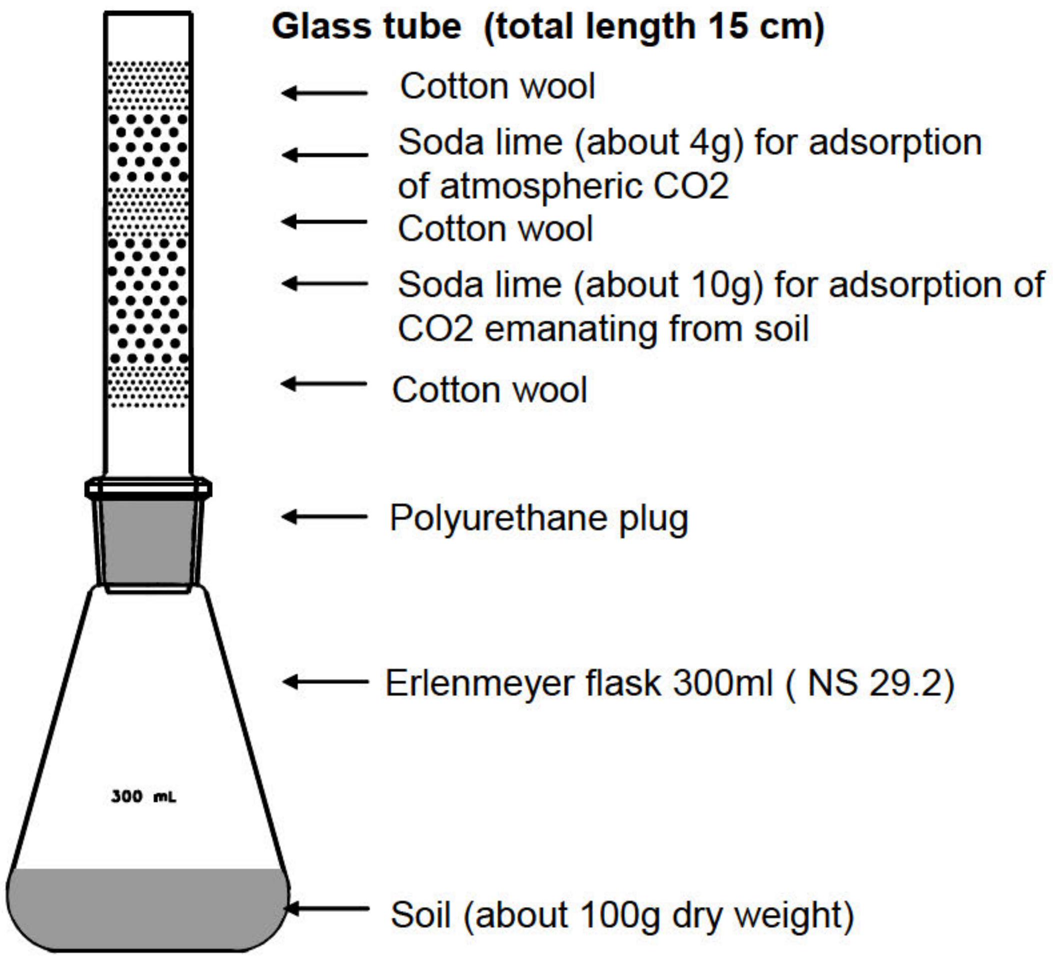


Figure 3: Flow Chart Showing the Procedure for Sample Analysis

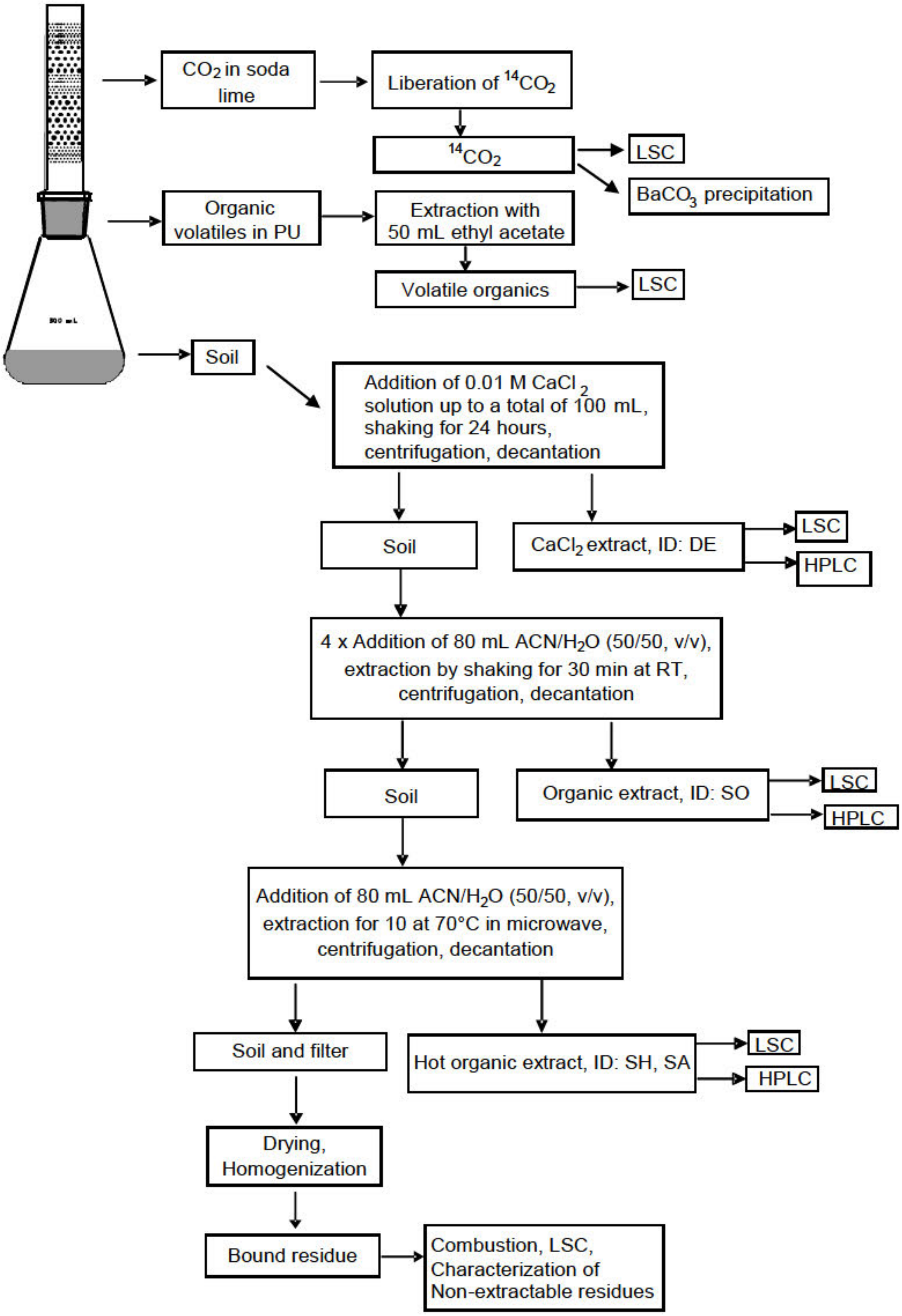


Figure 4: Flow Chart Showing the Procedure for Characterization of Bound Residues

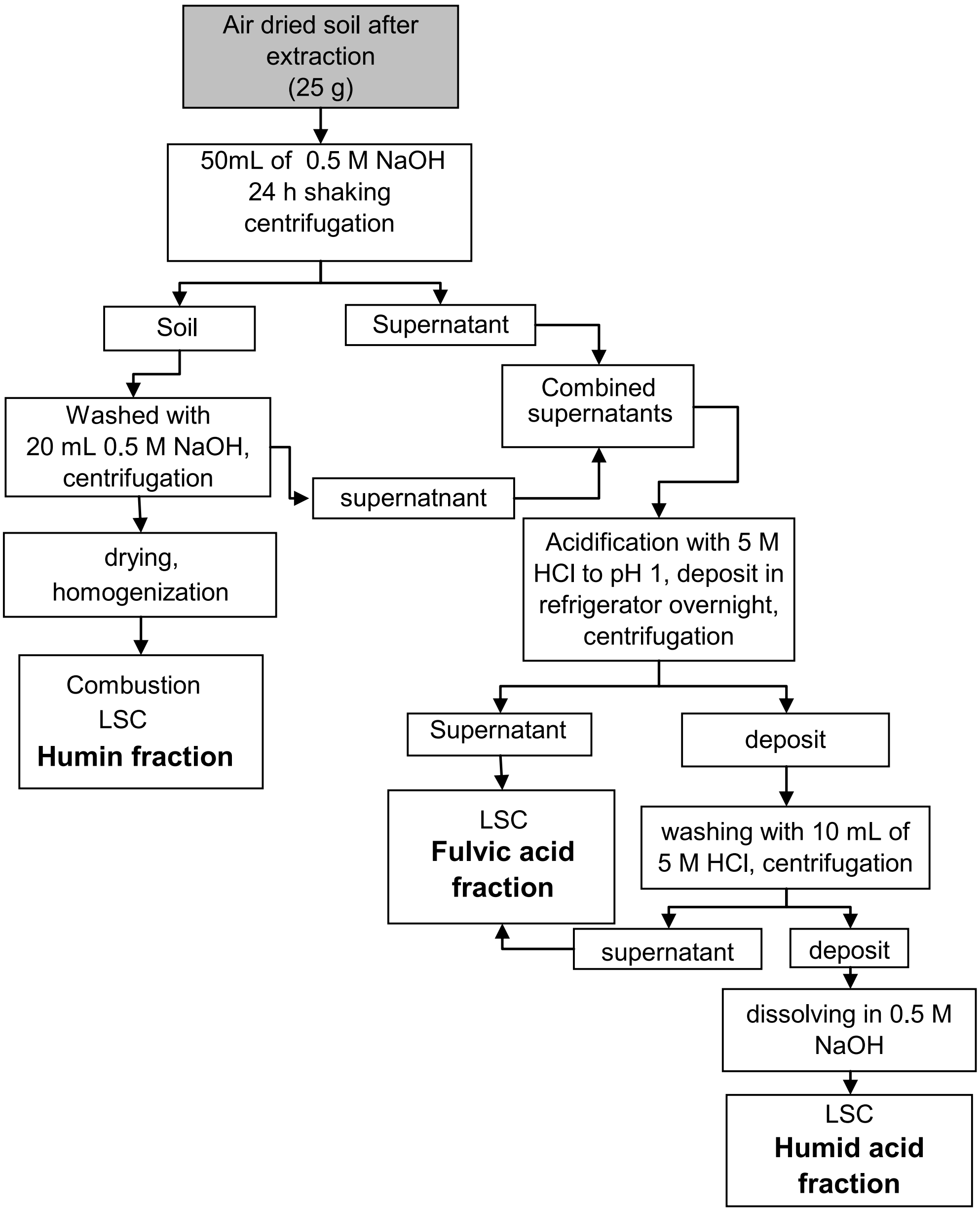
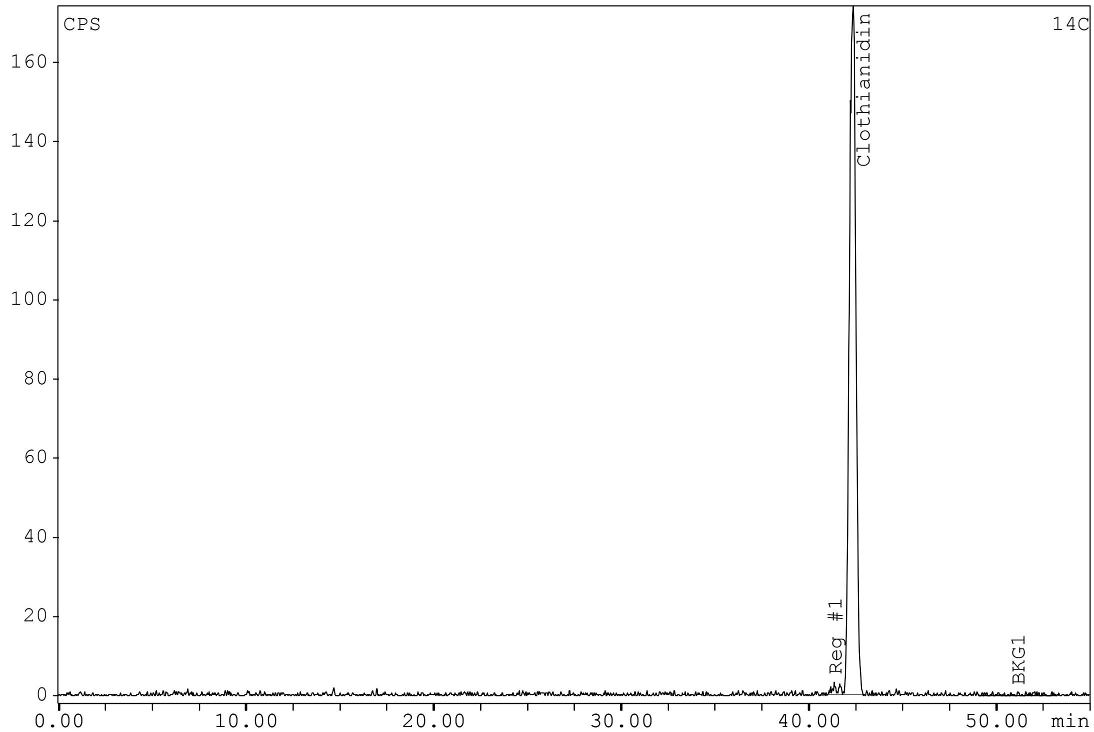


Figure 5: Chromatogram of [Guanidine-^{14}C]Clothianidin Application Solution

Sample ID: Ja57 AÜ1 (dilution of Application solution)

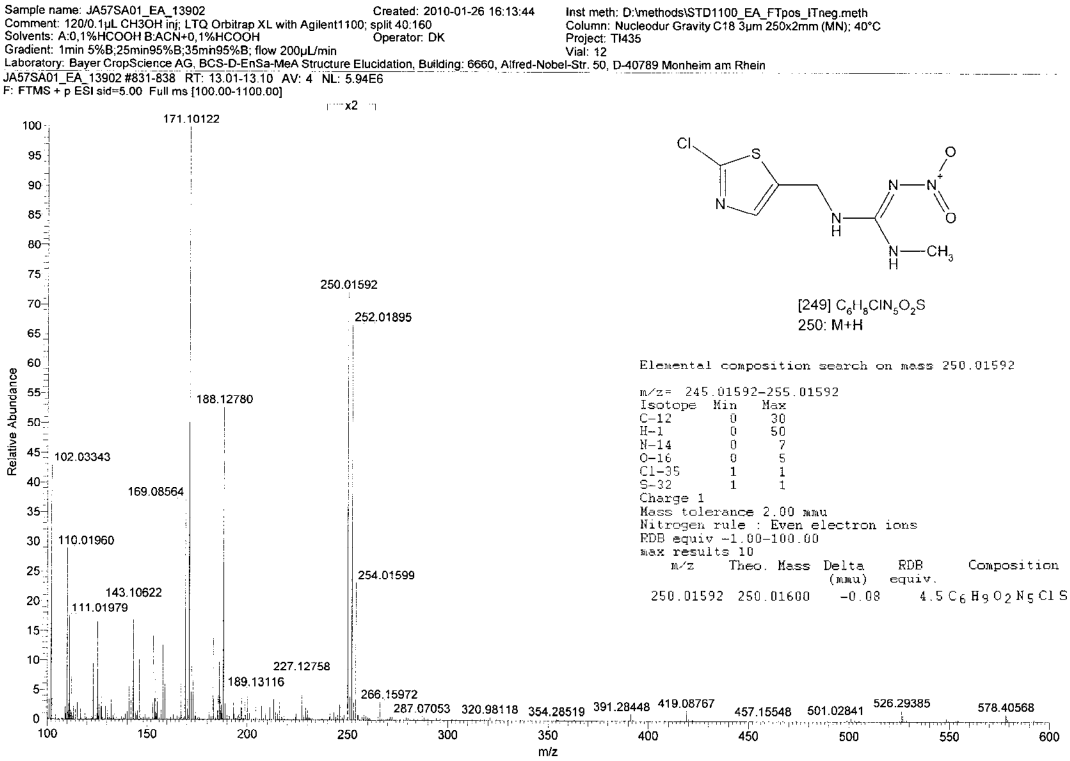
Integration Q:\D\MEF\Transfer\Unold\M1311911-7\MR218_02\JA57\JA57.011



Peak (^{14}C)				
	Ret. time	Area	Area	Total
		Counts	%	%
Reg #1	41.38	57.908	1.34	1.33
Clothianidin	42.35	4250.364	98.66	97.96
Sum Peaks		4308.272		
Area		4338.908		
BKG1		0.3080		

Figure 6: LC-MS and LC-MS/MS Spectra (ESI positive) of Clothianidin in Stock Solution

Spectrum ID: Ja57SA01



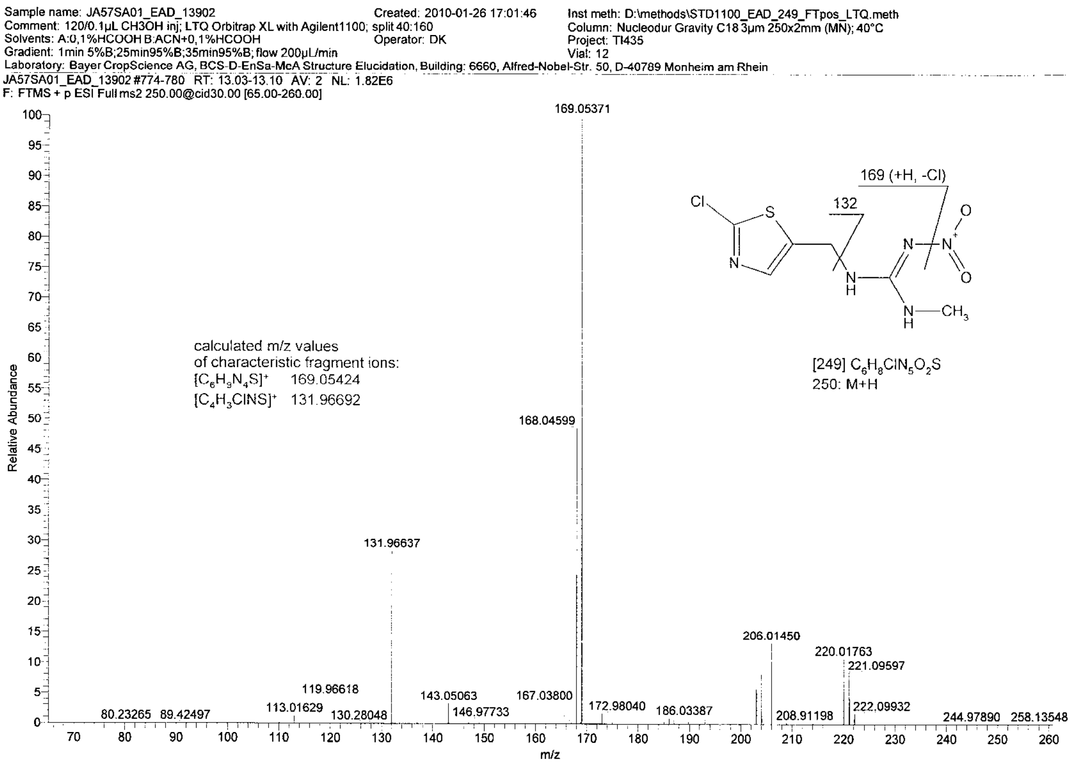


Figure 7: NMR-Spectrum of Clothianidin in Stock Solution

Spectrum ID: Ja57SA01

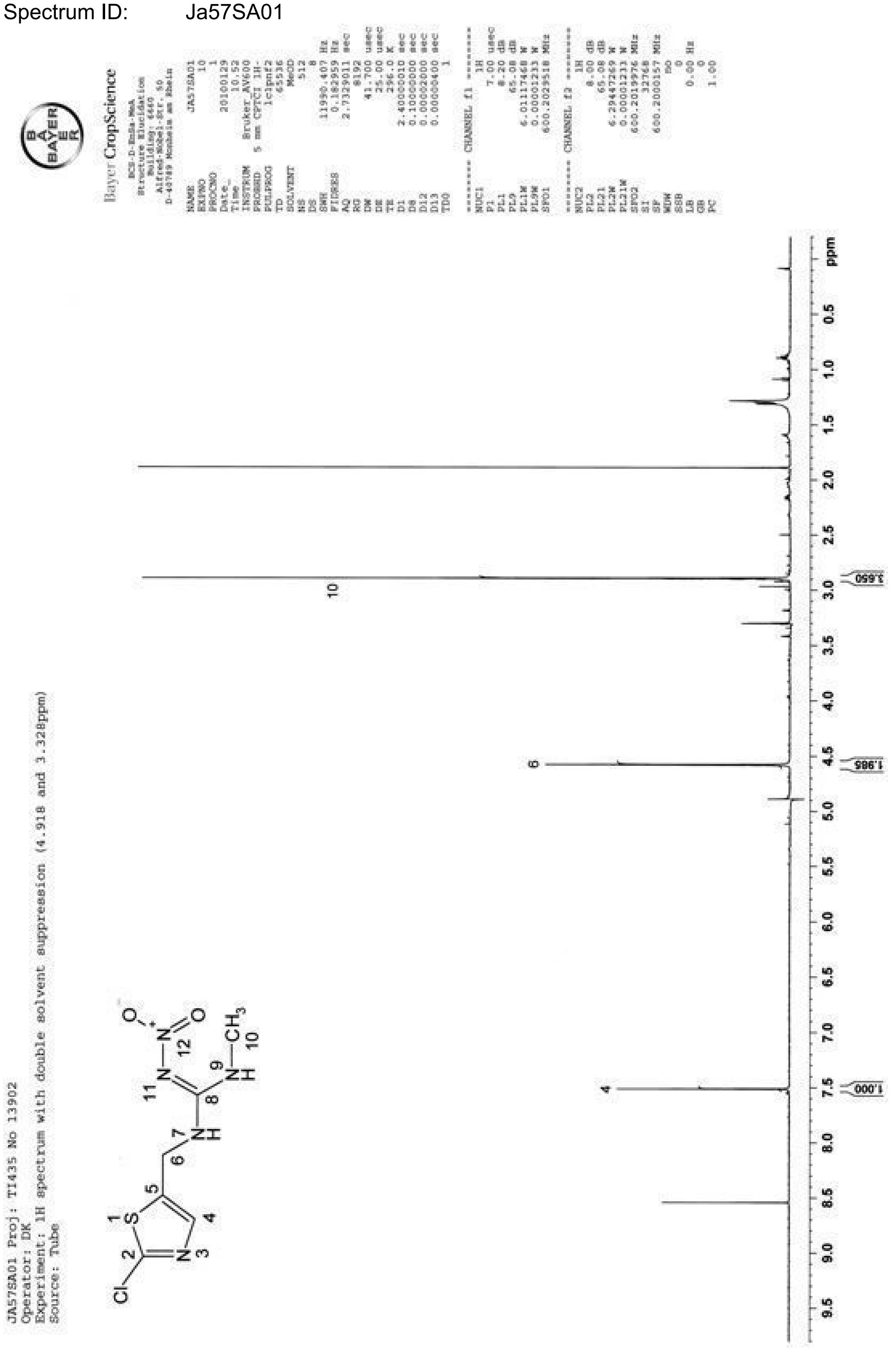
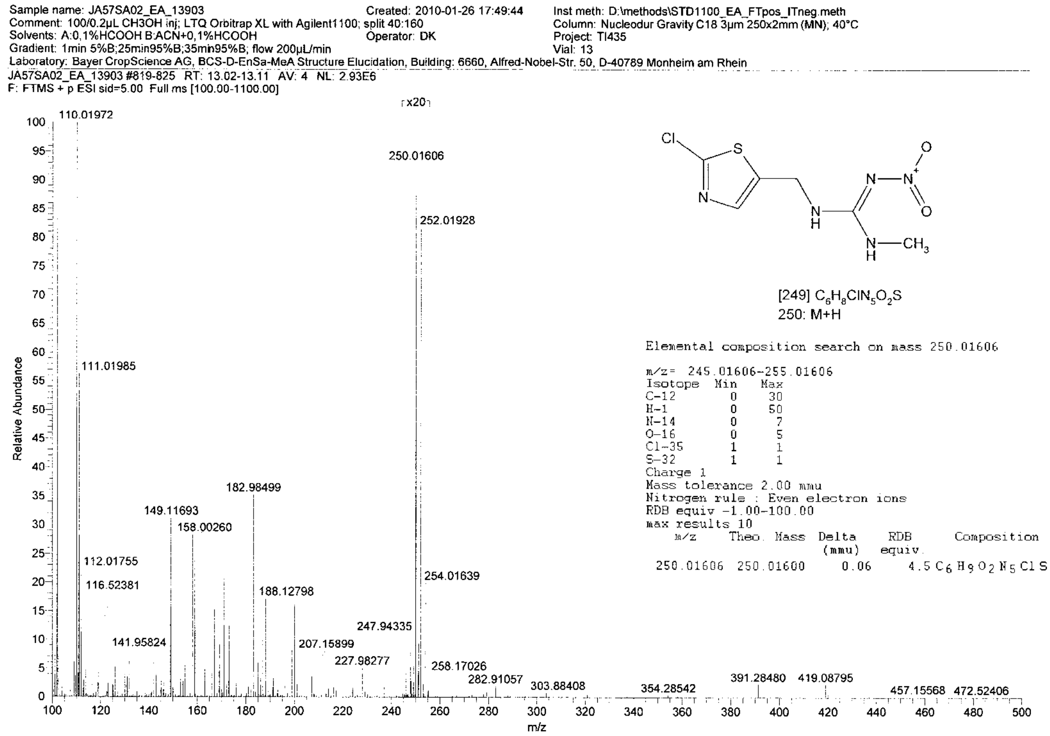


Figure 8: LC-MS and LC-MS/MS Spectra of test item in an ambient organic extract

The test item was characterized in a fraction of ambient organic extracts sampled at DAT-49 from Soil Wellesbourne (Ja57 49 C/D SO)

Spectrum ID: Ja57SA02



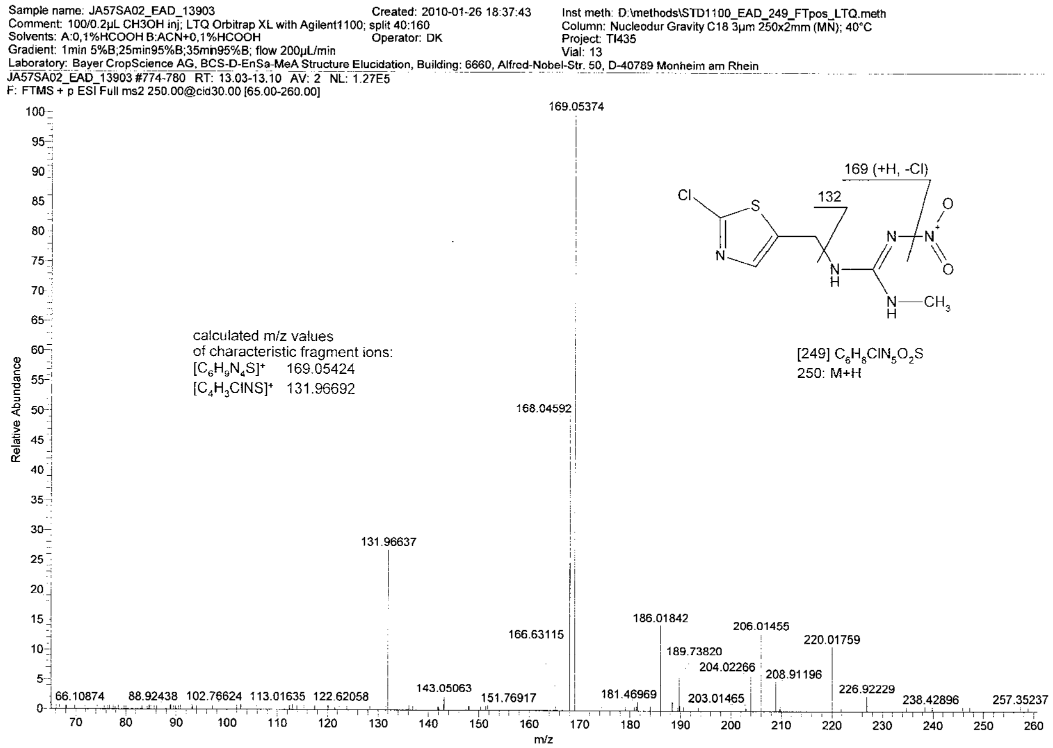
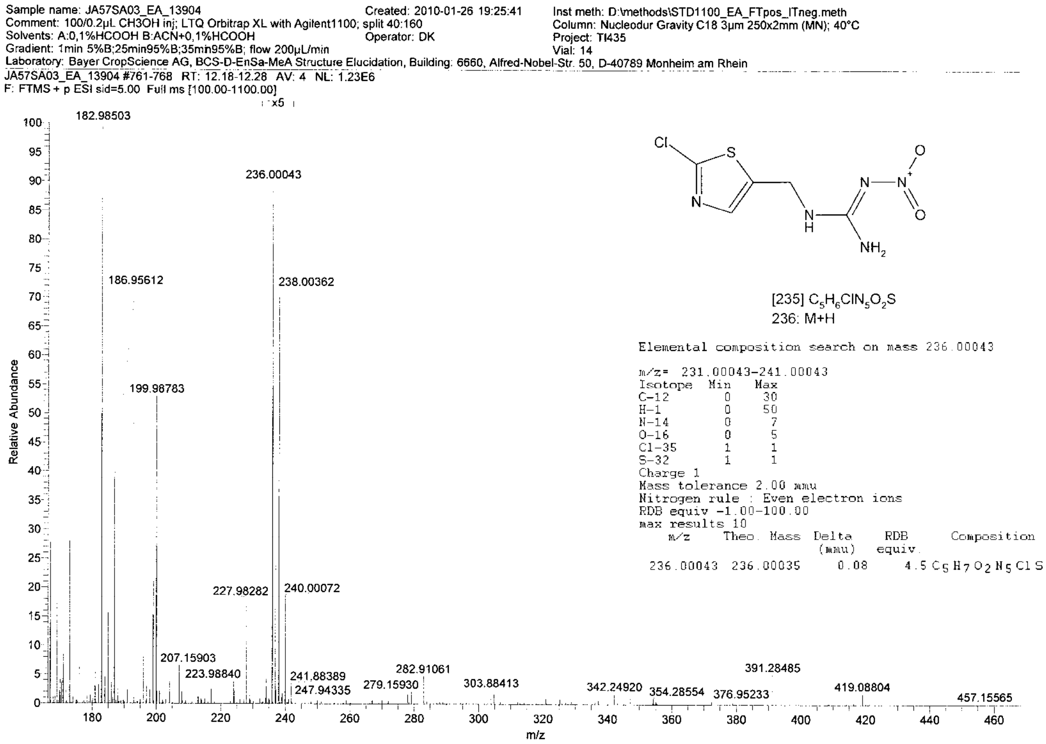


Figure 9: LC-MS and LC-MS/MS Spectra of Relevant Metabolites

Original Samples: Ja57 49 C/D SO

TZNG, Spectrum ID Ja57SA03



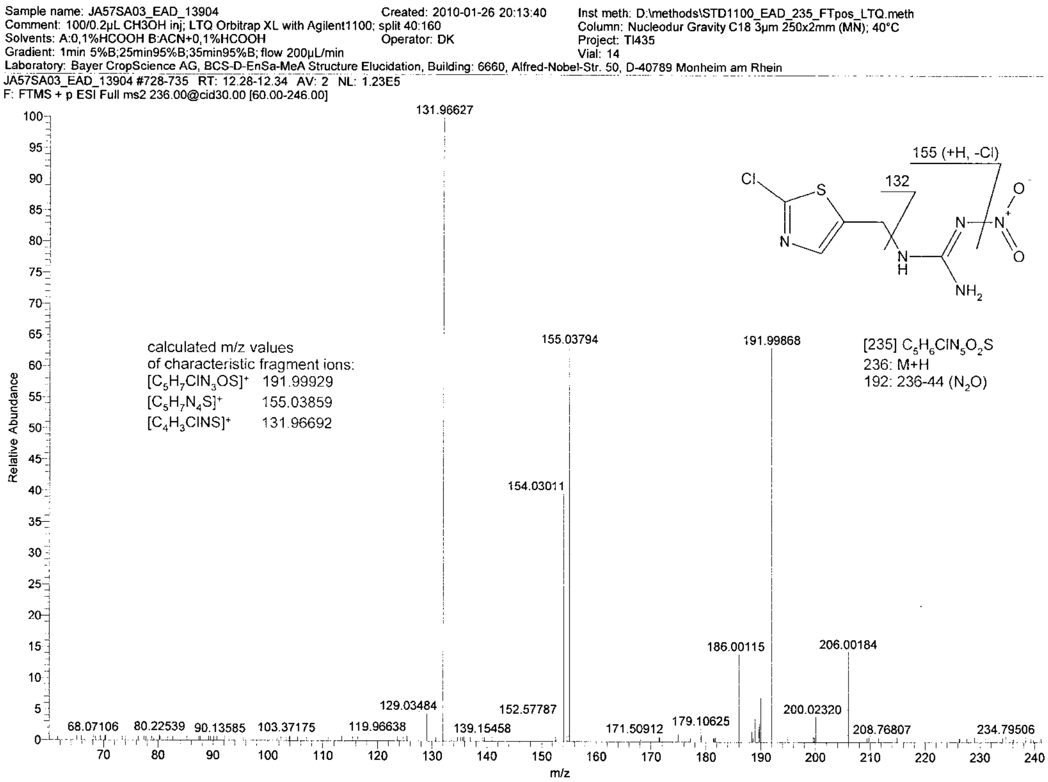
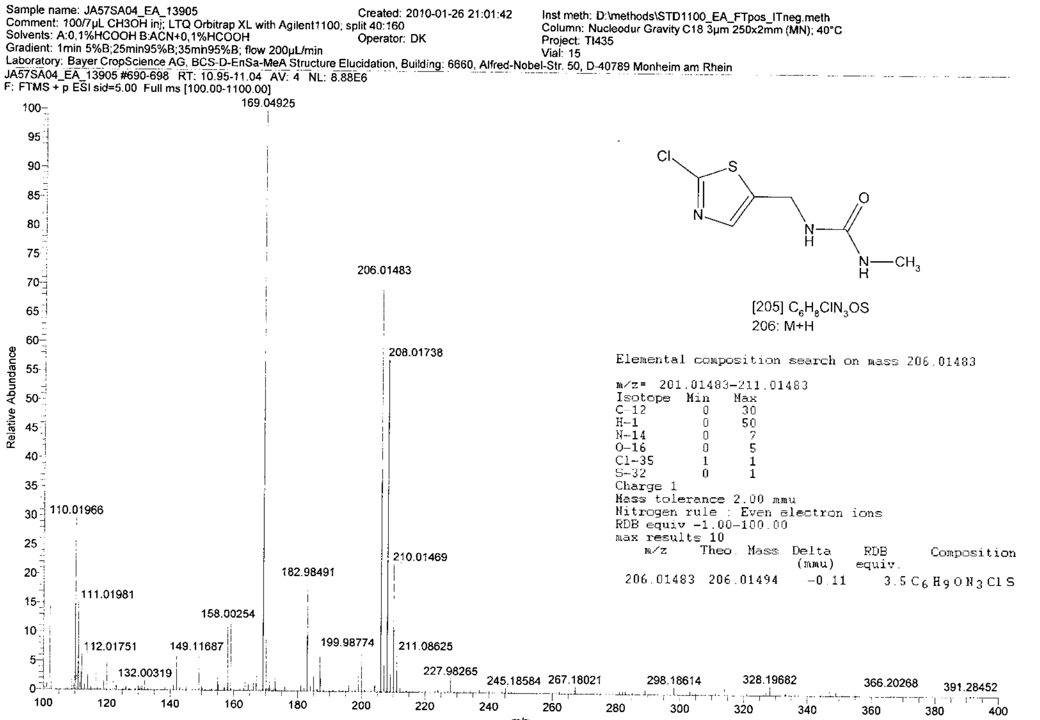


Figure 9: LC-MS and LC-MS/MS Spectra of Relevant Metabolites (continued)

Original Samples: Ja57 49 C/D SO

TZMU, Spectrum ID Ja57SA04



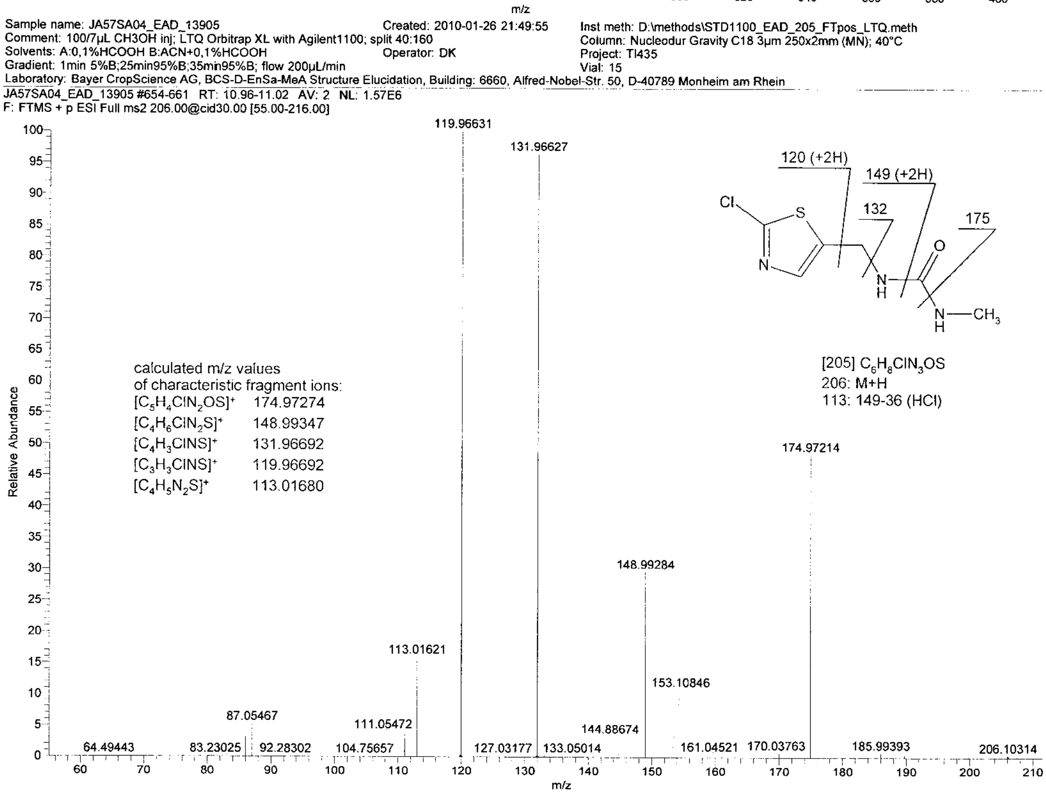
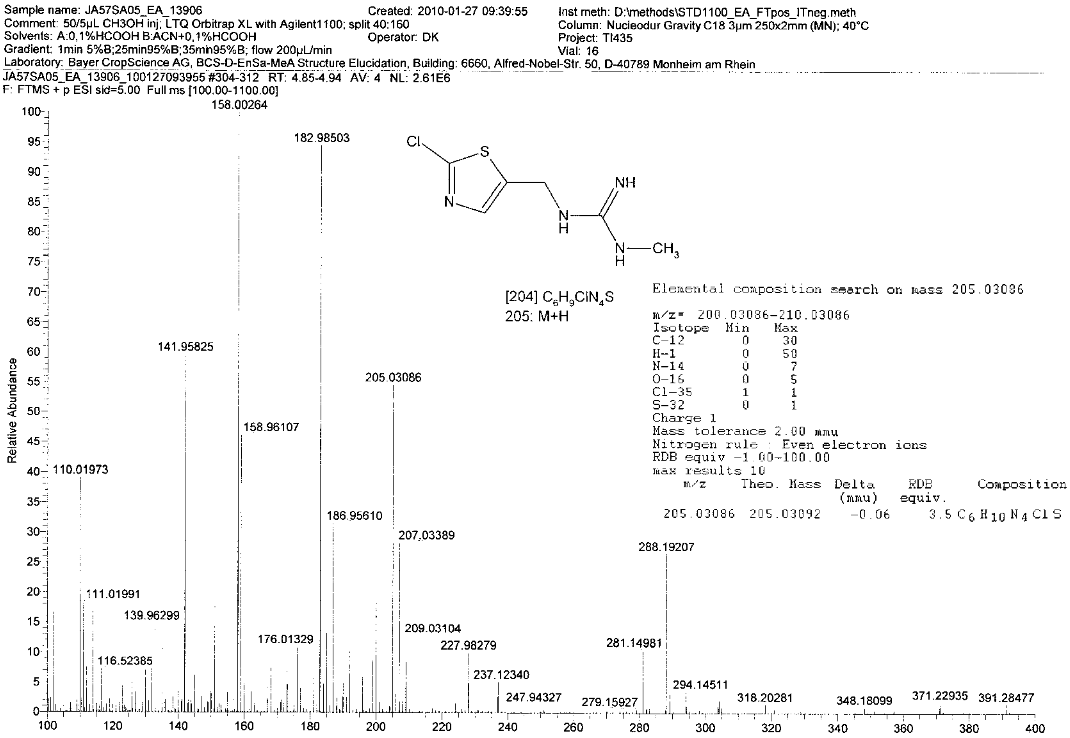


Figure 9: LC-MS and LC-MS/MS Spectra of Relevant Metabolites (continued)

Original Samples: Ja57 49 C/D SO

TMG, Spectrum ID Ja57SA05



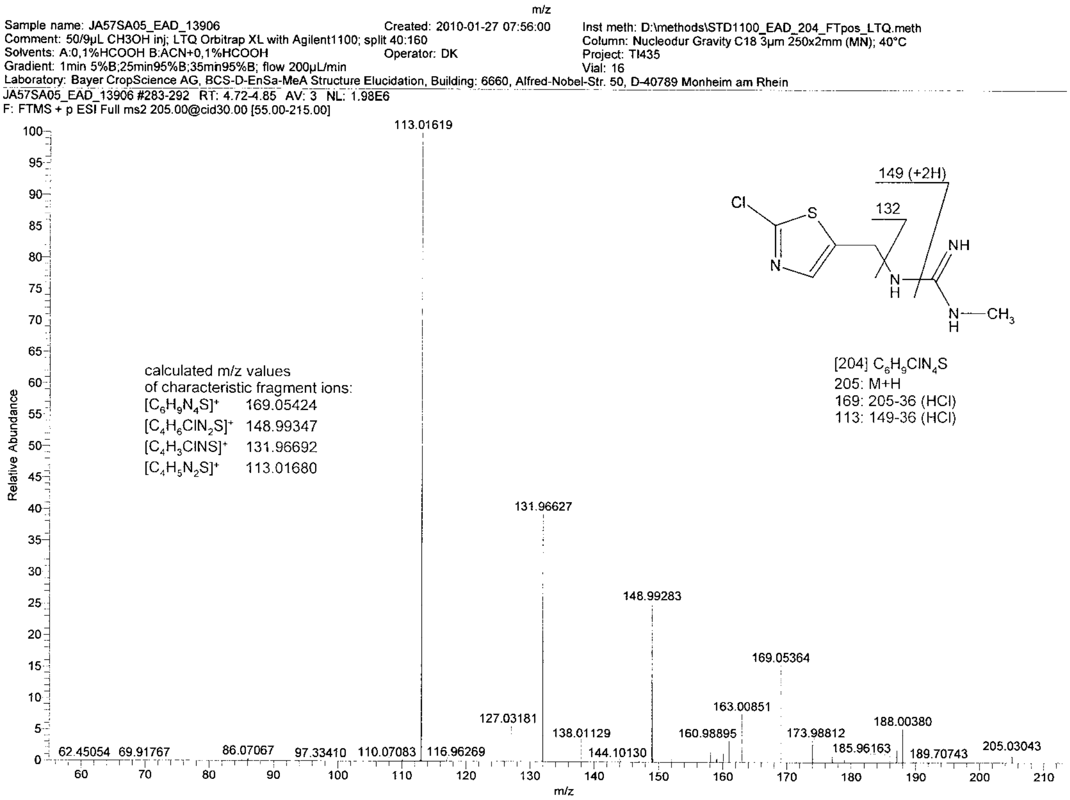
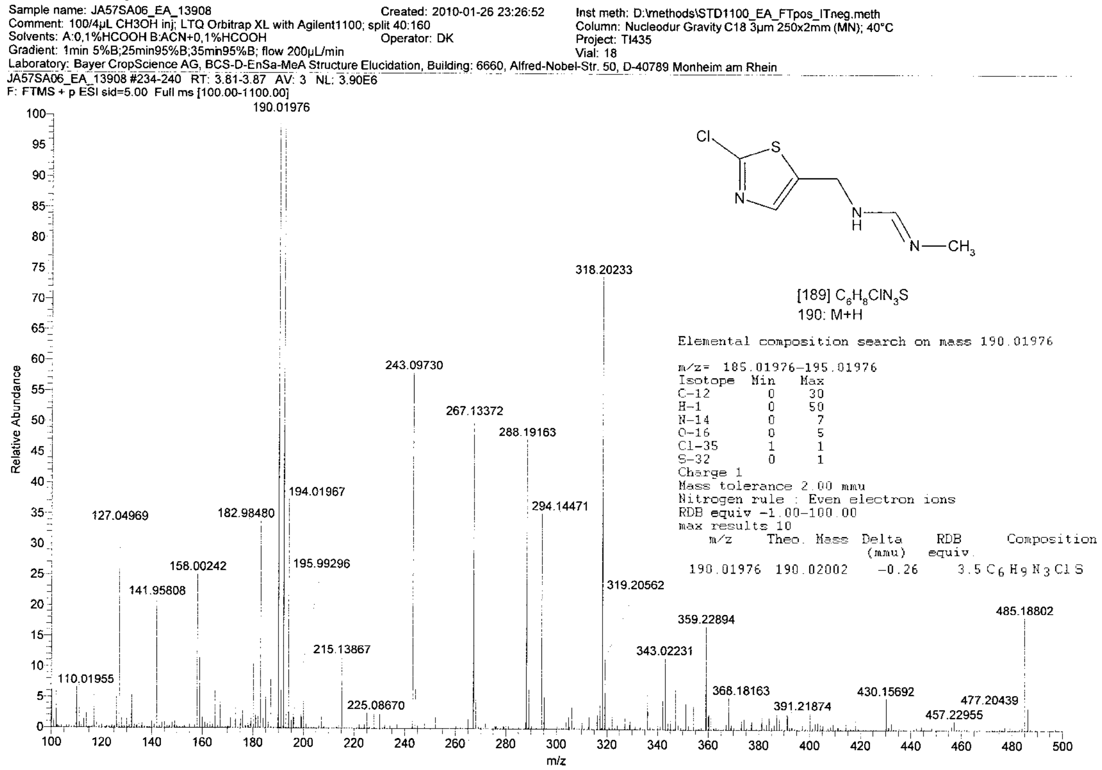


Figure 9: LC-MS and LC-MS/MS Spectra of Relevant Metabolites (continued)

Original Samples: Ja57 35 A/B SO

TZFA, Spectrum ID Ja57SA06



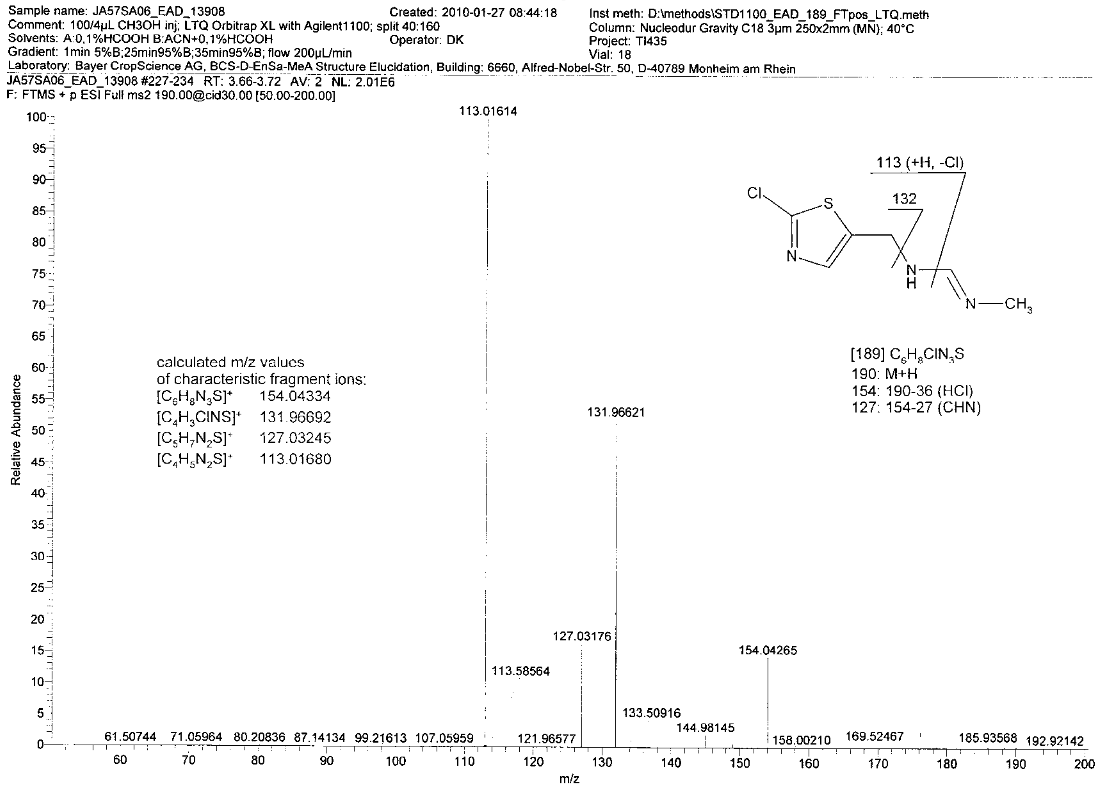


Figure 10: Example for Co-Chromatography

DAT-28, Sample ID: Ja57 28 D SOe (Soil Wellesbourne, Replicate D)

The sample was spiked with 20 µL of the TZNG stock solution (non-labeled)

Integration Q:\D\MEF\Transfer\Unold\M1311911-7\MR218_02\JA57\JA5728.029

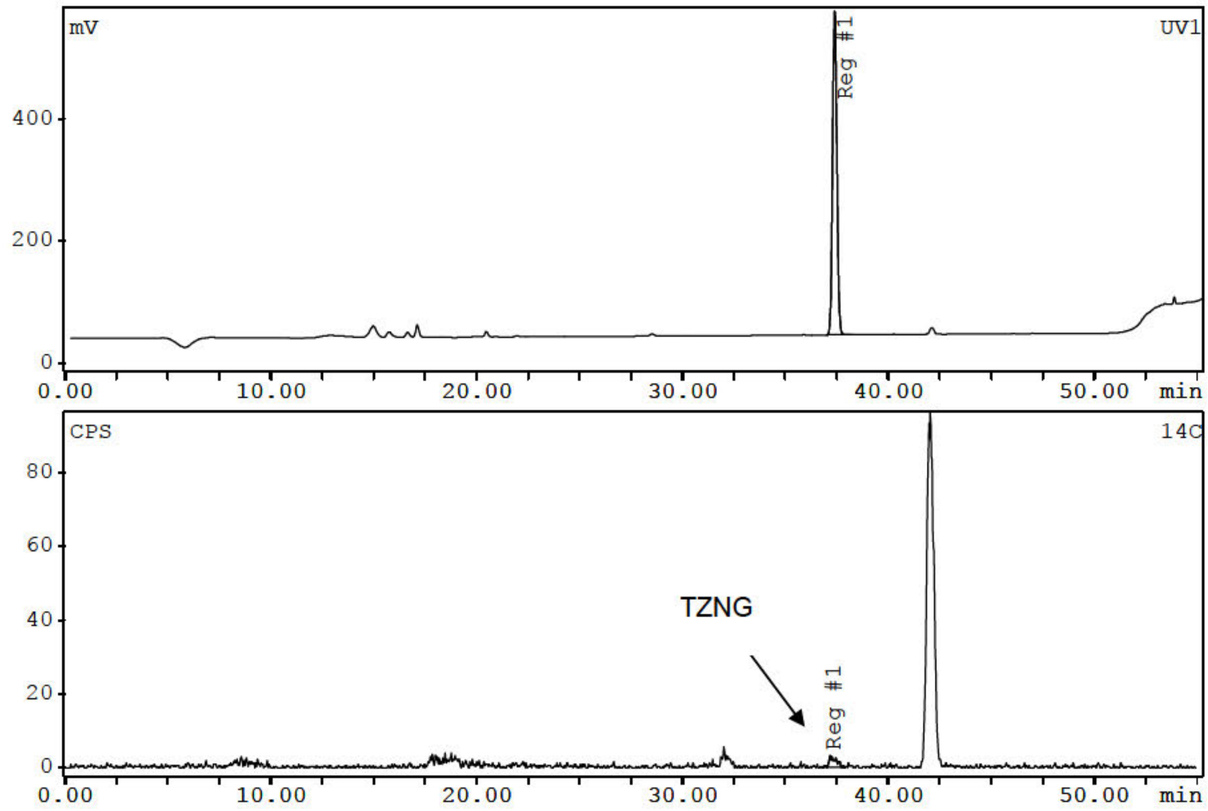
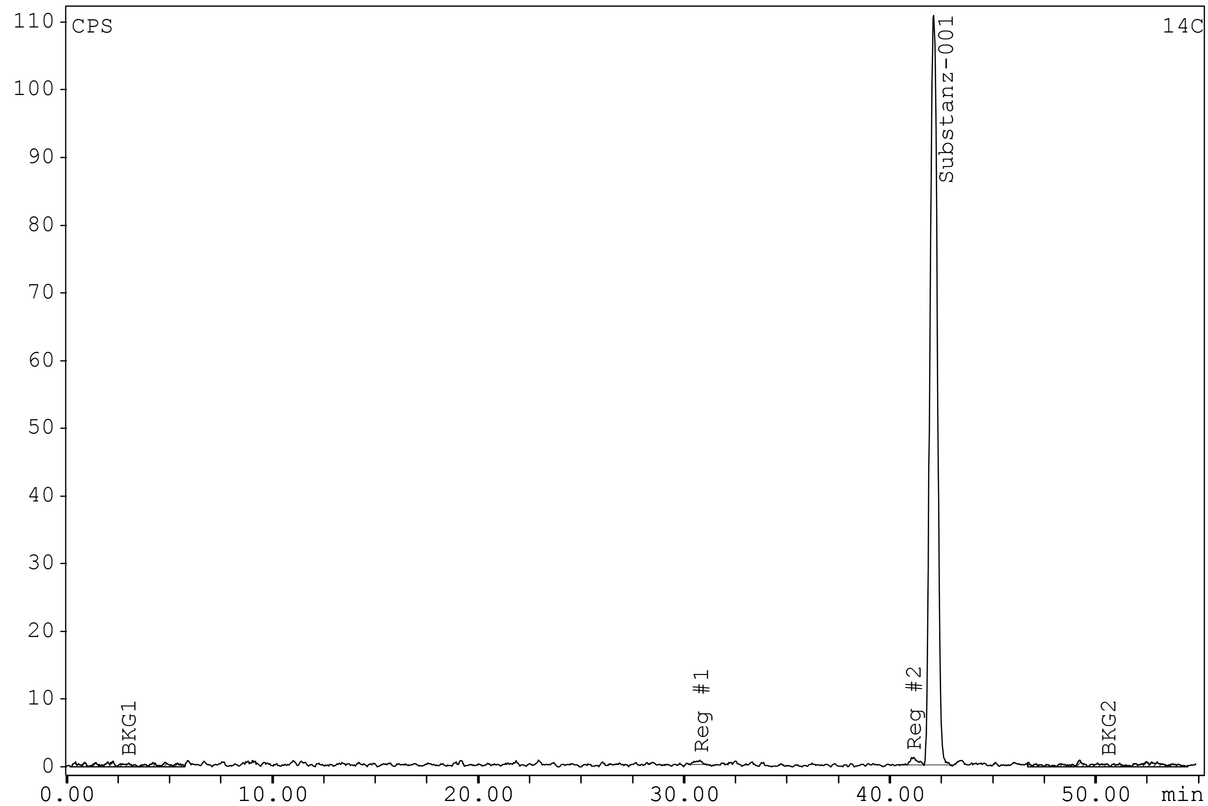


Figure 11: Representative Chromatography of Extracts from Clothianidin Time Dependent Sorption

HPLC of desorption solution ($CaCl_2$-extract)

DAT-0, Sample ID: Ja57 00 C DE (Soil Wellesbourne, Replicate C)

Integration Q:\D\MEF\Transfer\Unold\M1311911-7\MR218_02\JA57\JA5700.003



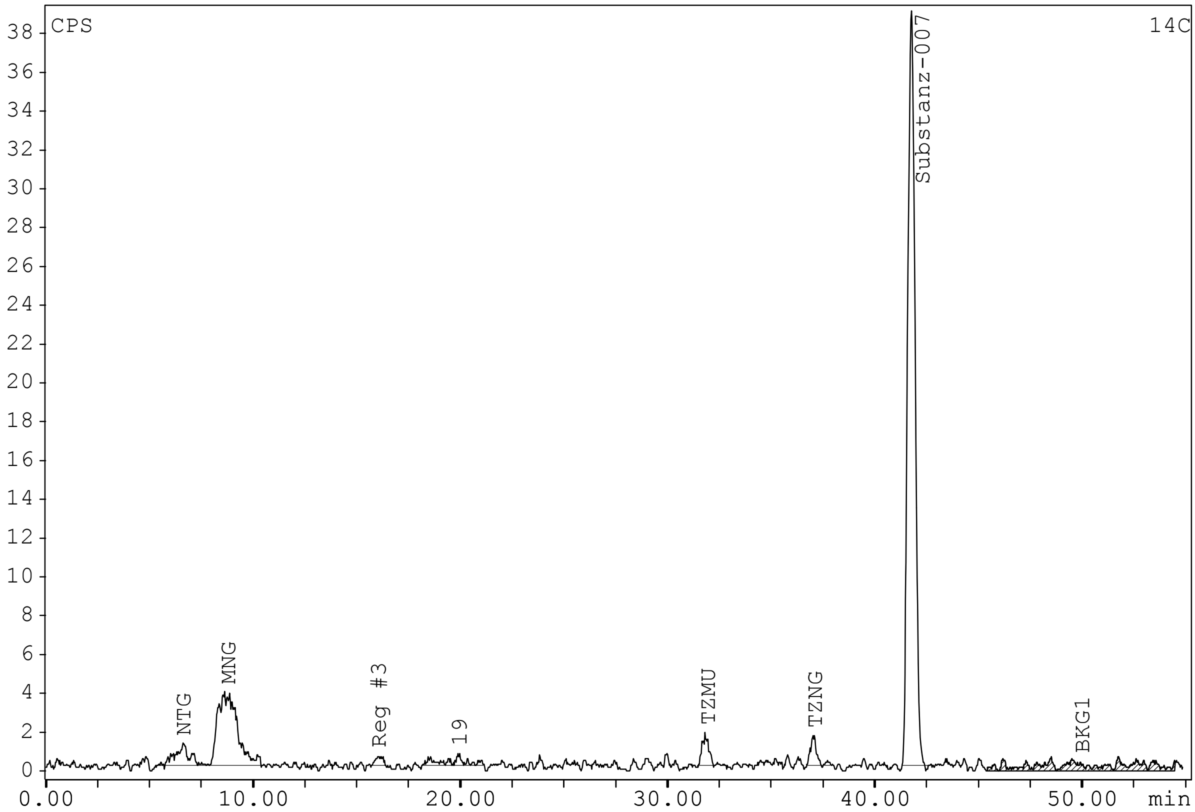
Peak (^{14}C)				
	Ret. time	**Area**	**Area**	**Total**
		Counts	**%**	**%**
Reg #1	30.82	15.663	0.56	0.56
Reg #2	41.13	26.892	0.97	0.96
Clothianidin	42.15	2733.170	98.47	97.61
Sum Peaks		2775.724		
Area		2800.100		
BKG1		0.3284		
BKG2		0.3169		

Figure 11: Representative Chromatography of Extracts from Clothianidin Time Dependent Sorption (continued)

HPLC of desorption solution ($CaCl_2$-extract)

DAT-63, Sample ID: Ja57 63 C DE (Soil Wellesbourne, Replicate C)

Integration Q:\D\MEF\Transfer\Unold\M1311911-7\MR218_02\JA57\JA5763.028



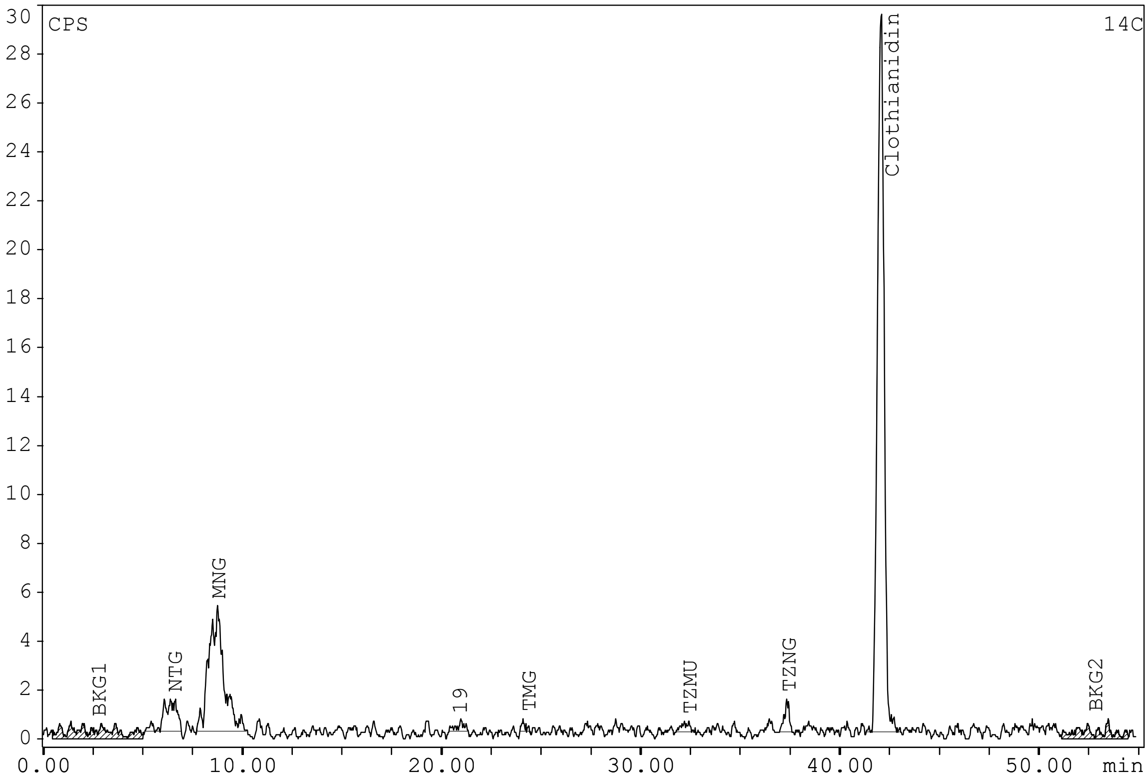
Peak (^{14}C)				
	Ret. time	Area	Area	Total
		Counts	%	%
NTG	6.60	52.9558	3.83	3.80
MNG	8.78	251.1160	18.16	18.03
Reg #3	16.05	12.5985	0.91	0.90
19 (TZFA)	19.92	36.7790	2.66	2.64
TZMU	31.90	36.2615	2.62	2.60
TZNG	37.07	32.8122	2.37	2.36
Clothianidin	41.78	960.4217	69.45	68.97
Sum Peaks		1382.9448		
Area		1392.5985		
BKG1		0.27808		

Figure 11: Representative Chromatography of Extracts from Clothianidin Time Dependent Sorption (continued)

HPLC of desorption solution ($CaCl_2$-extract)

DAT-120, Sample ID: Ja57 12 C DE (Soil Wellesbourne, Replicate C)

Integration Q:\D\MEF\Transfer\Unold\M1311911-7\MR218_02\JA57\JA5712.003



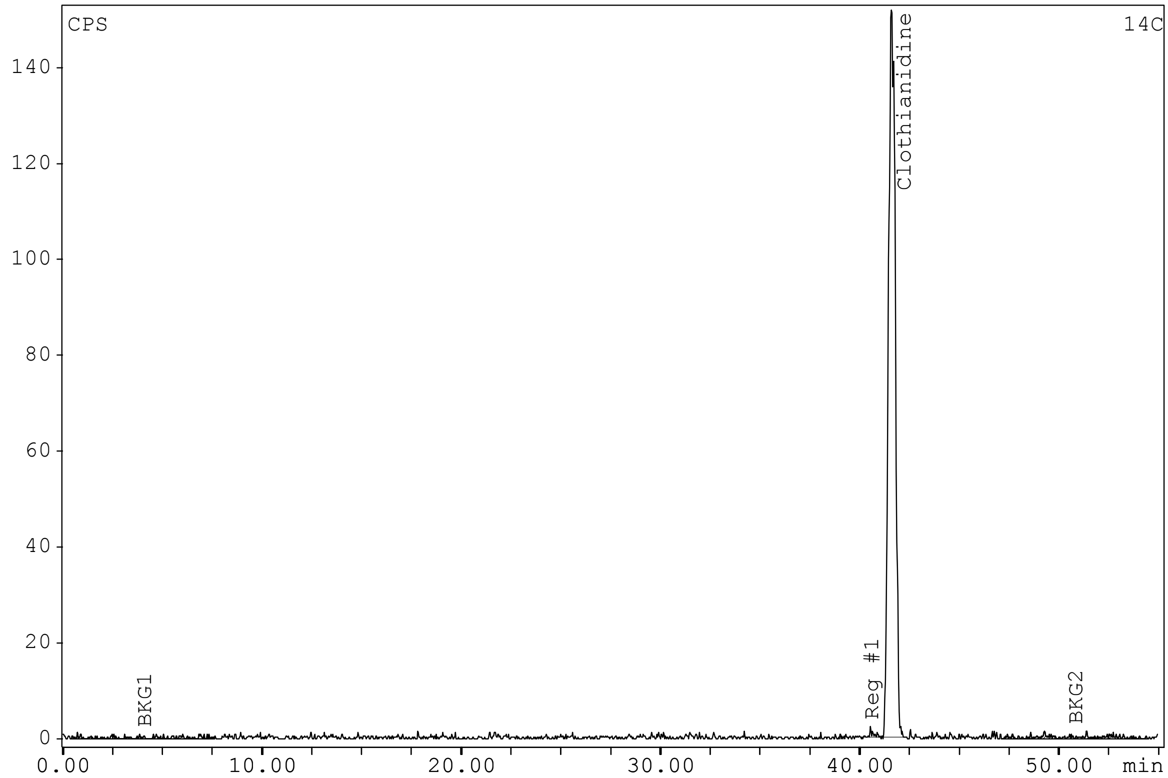
Peak ([14]C)				
	Ret. time	Area	Area	Total
		Counts	%	%
NTG	6.57	60.6672	5.62	5.53
Substanz 002 (MNG)	8.77	268.8946	24.93	24.52
19 (TZFA)	20.90	12.3840	1.15	1.13
TMG	24.33	6.8675	0.64	0.63
TZMU	32.45	11.9792	1.11	1.09
TZNG	37.40	21.7311	2.01	1.98
Clothianidin	42.12	696.2904	64.54	63.50
Sum Peaks		1078.8140		
Area		1096.5251		
BKG1		0.31985		
BKG2		0.29000		

Figure 11: Representative Chromatography of Extracts from Clothianidin Time Dependent Sorption (continued)

HPLC of ambient organic extracts

DAT-0, Sample ID: Ja57 00 C SOe, concentrated (Soil Wellesbourne, Replicate C)

Integration Q:\D\MEF\Transfer\Unold\M1311911-7\MR218_02\JA57\JA5700.011



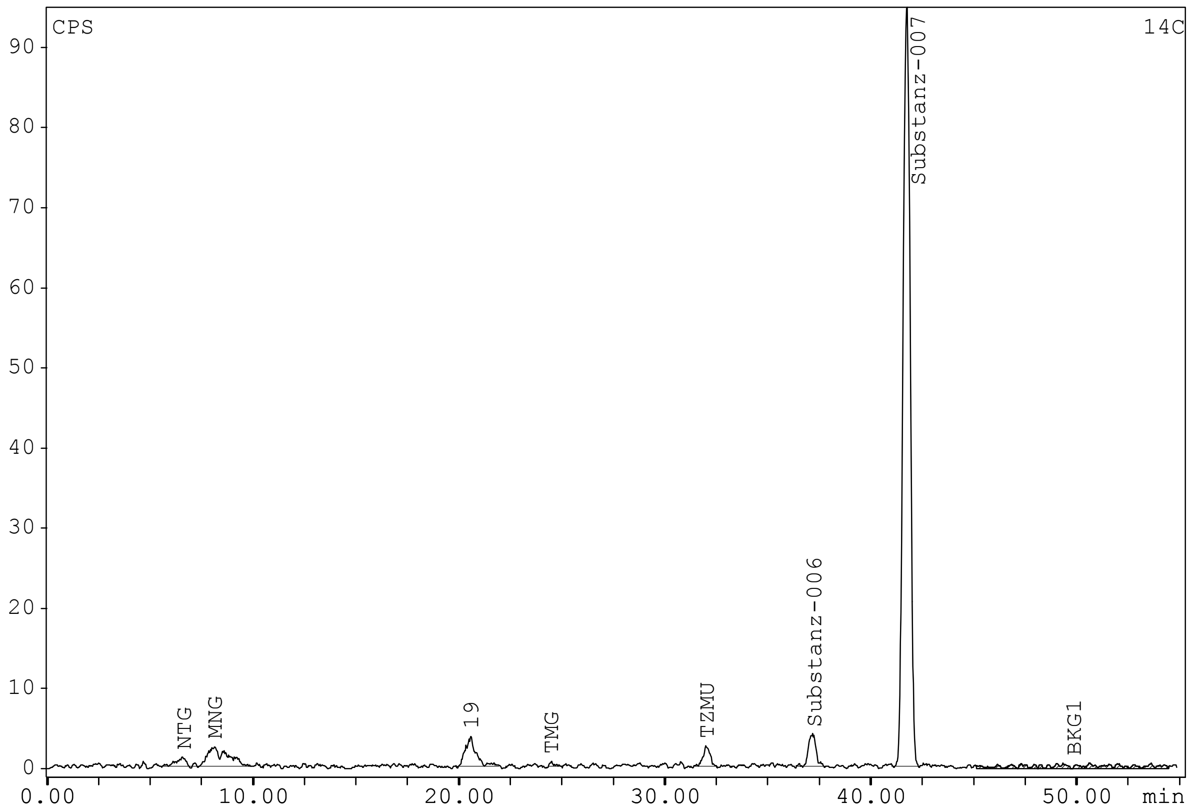
Peak (^{14}C)				
	Ret. time	**Area**	**Area**	**Total**
		Counts	**%**	**%**
Reg#1	40.55	21.912	0.60	0.59
Clothianidin	41.60	3658.819	99.40	98.28
Sum Peaks		3680.731		
Area		3722.751		
BKG1		0.2640		
BKG2		0.3476		

Figure 11: Representative Chromatography of Extracts from Clothianidin Time Dependent Sorption (continued)

HPLC of ambient organic extracts

DAT-63, Sample ID: Ja57 63 C SOe, concentrated (Soil Wellesbourne, Replicate C)

Integration Q:\D\MEF\Transfer\Unold\M1311911-7\MR218_02\JA57\JA5763.036



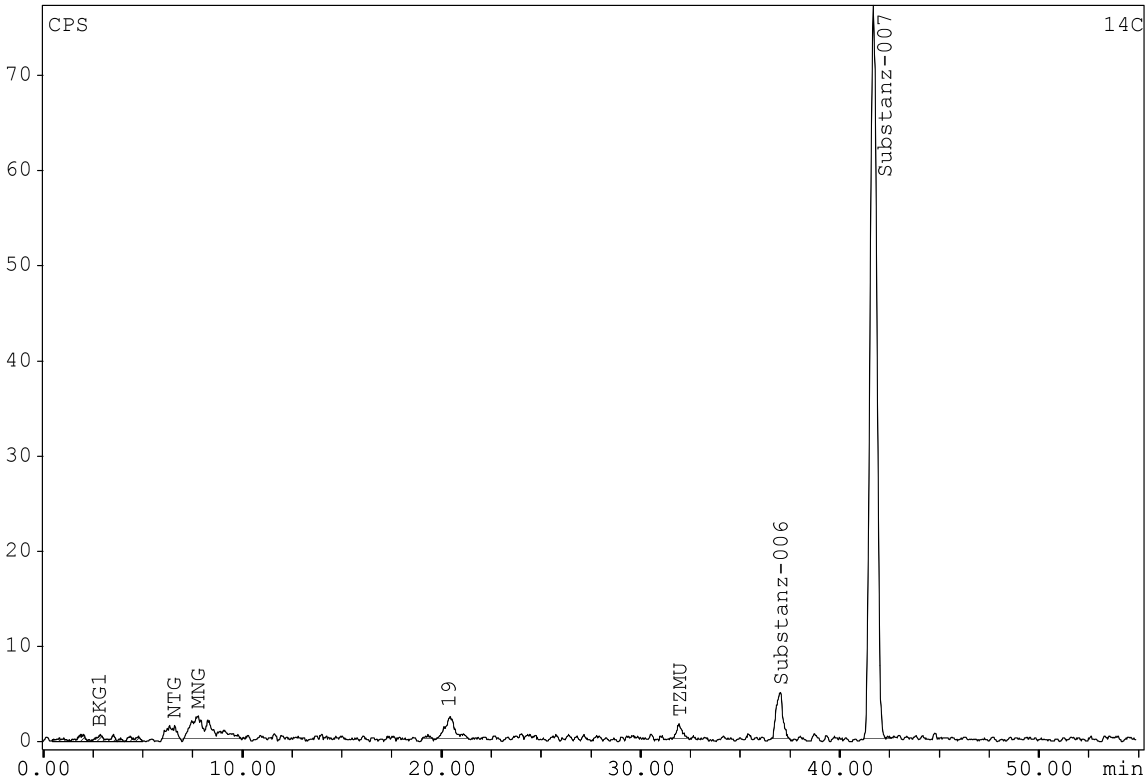
Peak (^{14}C)				
	Ret. time	**Area**	**Area**	**Total**
		Counts	**%**	**%**
NTG	6.62	38.485	1.38	1.36
MNG	8.07	159.424	5.72	5.64
19 (TZFA)	20.58	132.333	4.75	4.68
TMG	24.45	8.788	0.32	0.31
TZMU	32.00	57.788	2.07	2.04
Substanz 006 (TZNG)	37.23	103.242	3.70	3.65
Clothianidin	41.70	2287.152	82.06	80.90
Sum Peaks		2787.212		
Area		2827.303		
BKG1		0.3030		

Figure 11: Representative Chromatography of Extracts from Clothianidin Time Dependent Sorption (continued)

HPLC of ambient organic extracts

DAT-120, Sample ID: Ja57 12 C SOe, concentrated (Soil Wellesbourne, Replicate C)

Integration Q:\D\MEF\Transfer\Unold\M1311911-7\MR218_02\JA57\JA5712.011



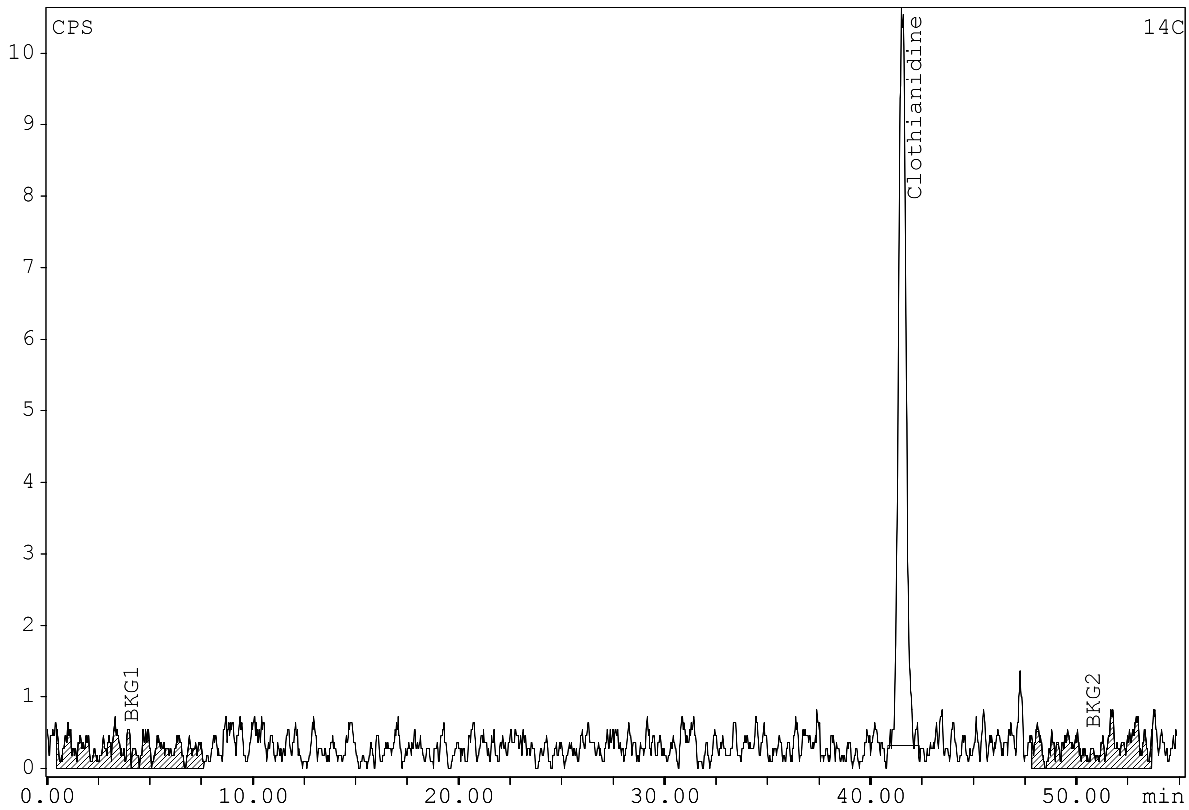
Peak (^{14}C)				
	Ret. time	Area	Area	Total
		Counts	%	%
NTG	6.52	43.912	1.92	1.91
MNG	7.72	167.816	7.35	7.30
19 (TZFA)	20.35	86.287	3.78	3.75
TZMU	31.92	29.404	1.29	1.28
Substanz 006 (TZNG)	37.02	112.213	4.91	4.88
Clothianidin	41.73	1844.375	80.75	80.24
Sum Peaks		2284.007		
Area		2298.449		
BKG1		0.3015		
BKG2		0.3237		

Figure 11: Representative Chromatography of Extracts from Clothianidin Time Dependent Sorption (continued)

HPLC of aggressive organic extracts

DAT-0, Sample ID: Ja57 00 C SHe, concentrated (Soil Wellesbourne, Replicate C)

Integration Q:\D\MEF\Transfer\Unold\M1311911-7\MR218_02\JA57\JA5700.020



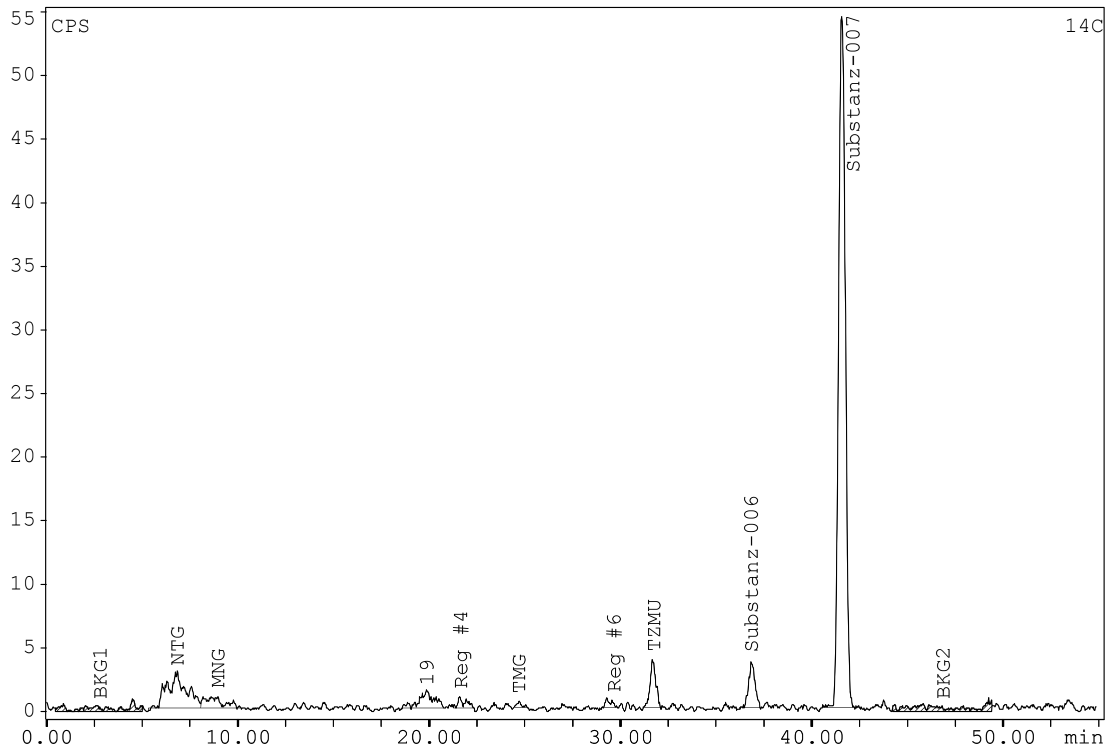
Peak (^{14}C)				
	Ret. time	**Area**	**Area**	**Total**
		Counts	**%**	**%**
Clothianidin	41.65	266.9017	100.00	95.55
Sum Peaks		266.9017		
Area		279.3257		
BKG1		0.28738		
BKG2		0.32764		

Figure 11: Representative Chromatography of Extracts from Clothianidin Time Dependent Sorption (continued)

HPLC of aggressive organic extracts

DAT-63, Sample ID: Ja57 63 C SHe, concentrated (Soil Wellesbourne, Replicate C)

Integration Q:\D\MEF\Transfer\Unold\M1311911-7\MR218_02\JA57\JA5763.044



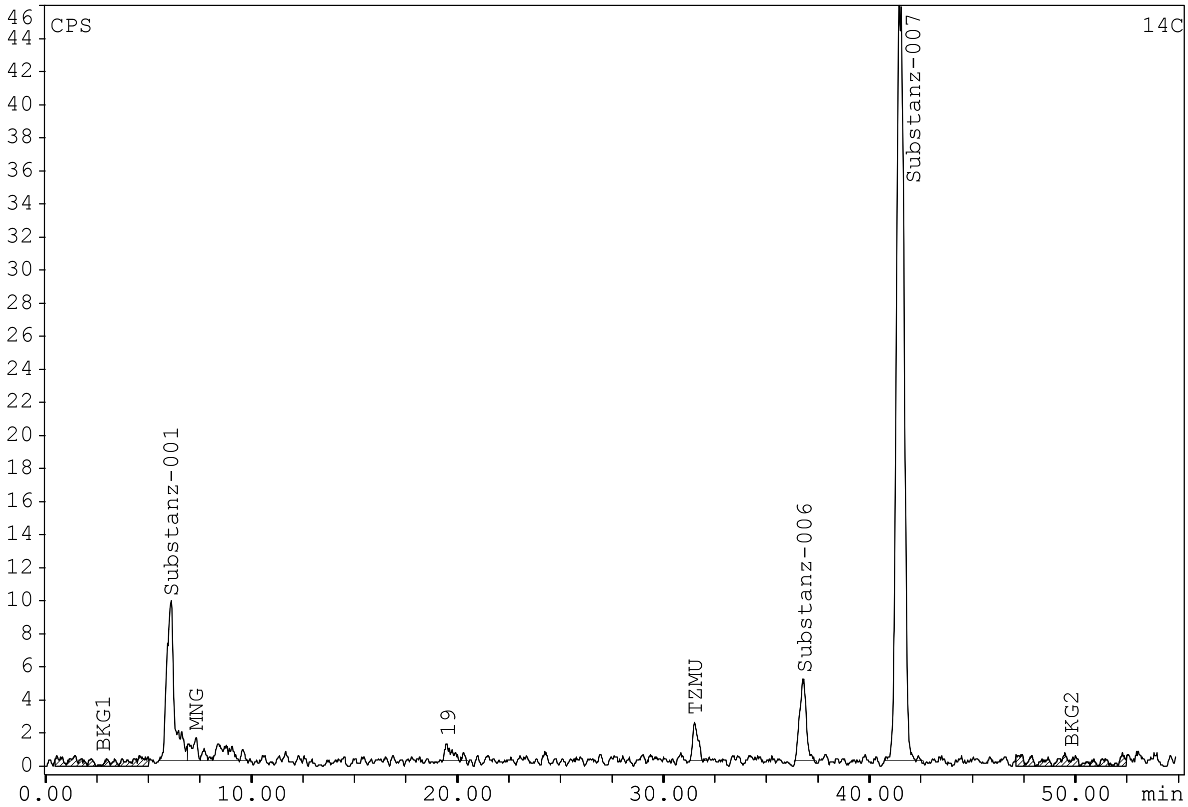
Peak (^{14}C)				
	Ret. time	Area	Area	Total
		Counts	%	%
NTG	6.77	193.738	10.28	10.27
MNG	8.90	60.771	3.23	3.22
19 (TZFA)	19.85	70.538	3.74	3.74
Reg#4	21.62	22.006	1.17	1.17
TMG	24.65	8.958	0.48	0.47
Reg#6	29.60	17.114	0.91	0.91
TZMU	31.70	80.498	4.27	4.27
Substanz 006 (TZNG)	36.82	81.341	4.32	4.31
Clothianidin	41.55	1349.333	71.61	71.51
Sum Peaks		1884.299		
Area		1886.866		
BKG1		0.2647		
BKG2		0.3226		

Figure 11: Representative Chromatography of Extracts from Clothianidin Time Dependent Sorption (continued)

HPLC of aggressive organic extracts

DAT-120, Sample ID: Ja57 12 C SHe, concentrated (Soil Wellesbourne, Replicate C)

Integration Q:\D\MEF\Transfer\Unold\M1311911-7\MR218_02\JA57\JA5712.019

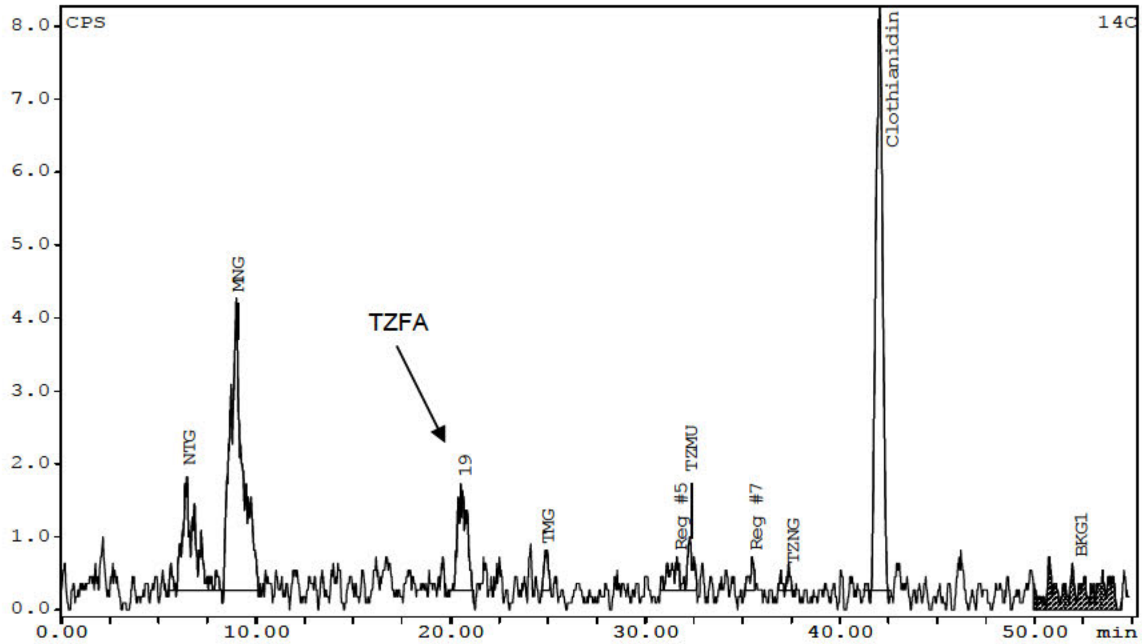


Peak (^{14}C)				
	Ret. time	Area	Area	Total
		Counts	%	%
Substanz 001 (NTG)	6.07	256.785	14.68	14.47
MNG	7.28	97.465	5.57	5.49
19 (TZFA)	19.52	26.452	1.51	1.49
TZMU	31.50	43.802	2.50	2.47
Substanz 006 (TZNG)	36.80	109.898	6.28	6.19
Substanz 007 (Clothianidin)	41.50	1214.271	69.44	68.41
Sum Peaks		1748.673		
Area		1775.010		
BKG1		0.3235		
BKG2		0.3251		

Figure 12: Comparison of Quantification (HPLC) and Confirmation Method (TLC)

1. Quantification method: **HPLC** (extract: Ja57 12 A DE (desorption solution))
Analysis ID: Ja5712.001

Integration Q:\D\MEF\Transfer\Unold\M1311911-7\MR218_02\JA57\JA5712.001



2. Confirmation method: **TLC** (extract: Ja57 12 A DE (desorption solution))
Analysis ID: Ja57DC09

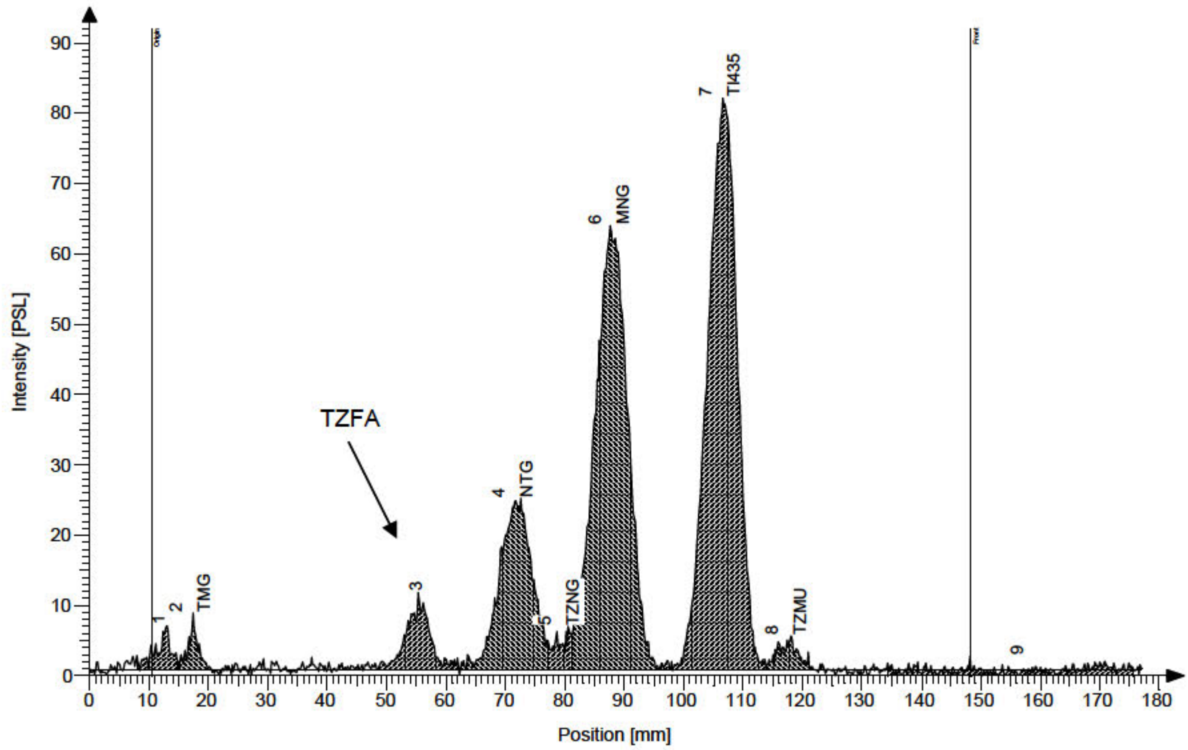
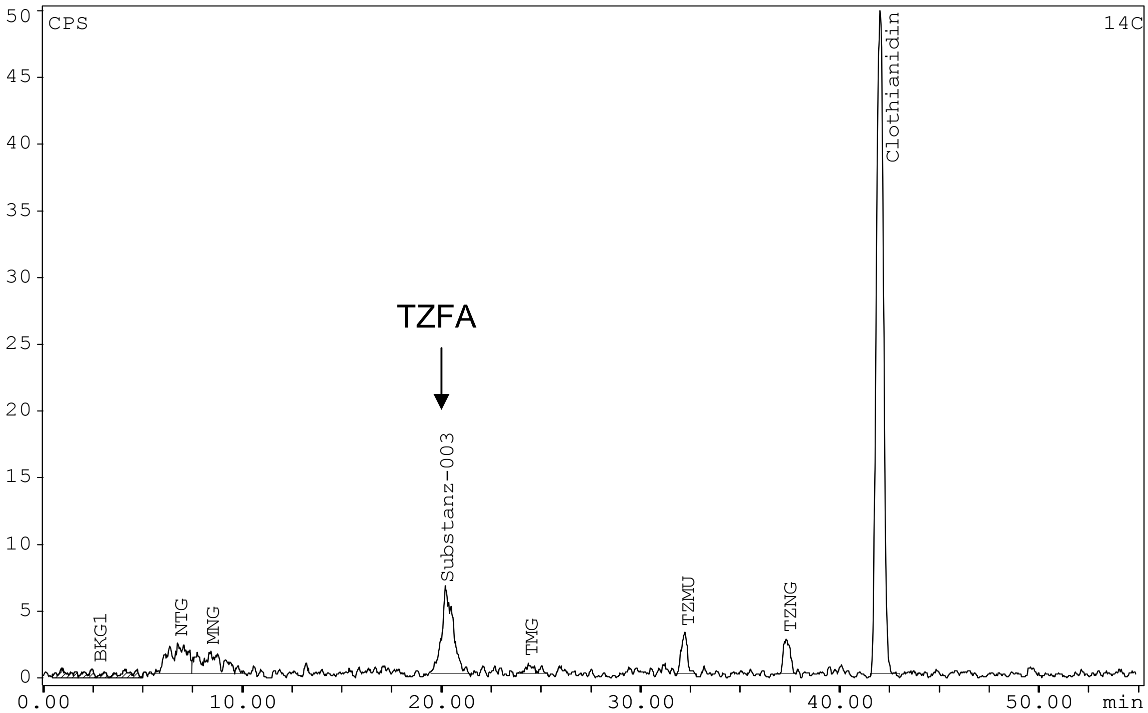


Figure 13: Storage stability of soil extracts sampled on DAT-98 from soil Hoefchen am Hohenseh plot 4011

Ambient Organic –extracts, Sample ID: JA57 98 A SOe

Prior to Storage

Integration Q:\D\MEF\Transfer\Unold\M1311911-7\MR218_02\JA57\JA5798.010



After Storage

Integration Q:\D\MEF\Transfer\Unold\M1311911-7\MR218_02\JA57\JA57LAGERST.013

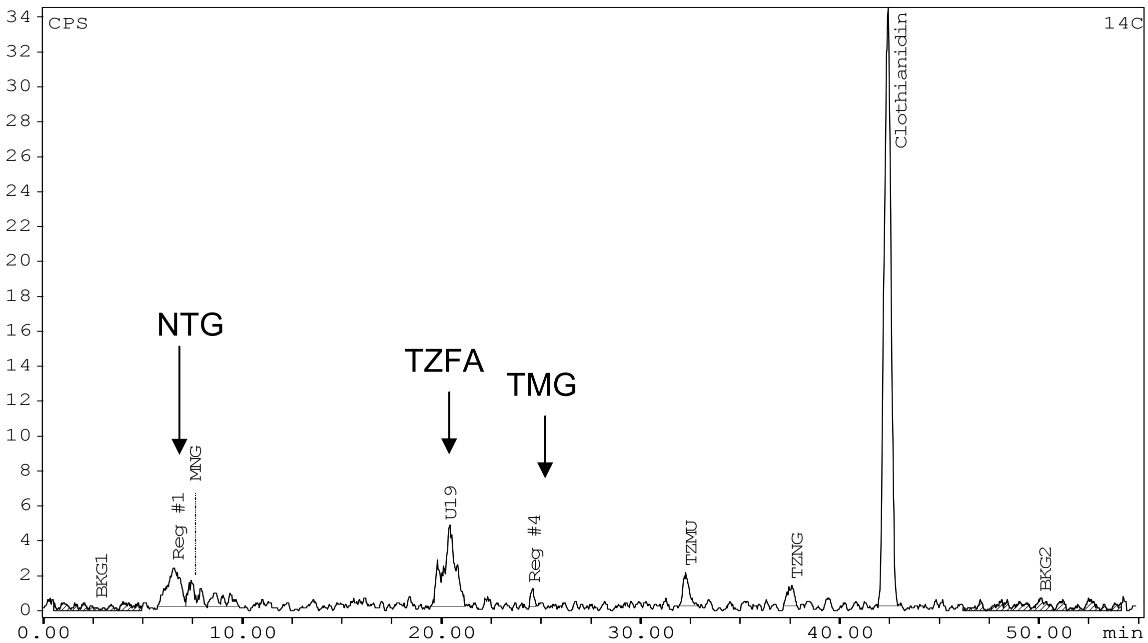
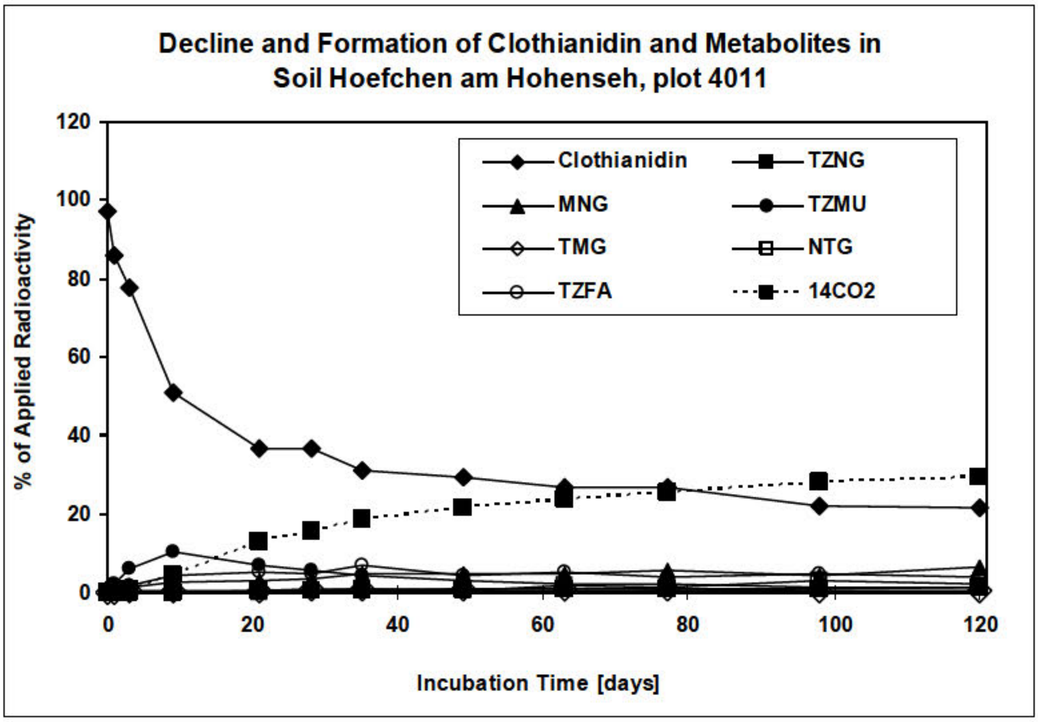


Figure 13: Storage stability of soil extracts sampled on DAT-98 from soil Hoefchen am Hohenseh plot 4011 (continued)

Peak (^{14}C)	**Prior to Storage Total [%]**	**After storage for 108 days Total [%]**
File Name	Ja5798.010	Ja57Lagerst.013
Clothianidin	62.3	63.0
TZNG	3.1	2.2
TZMU	3.2	3.1
MNG	6.6	6.5
NTG	7.6	7.9
TZFA	13.0	15.5
TMG	1.3	1.0

Figure 14: Pattern of Decline of Clothianidin and Formation and Decline of Metabolites

Soil Hoefchen am Hohenseh plot 4011



Soil Wellesbourne

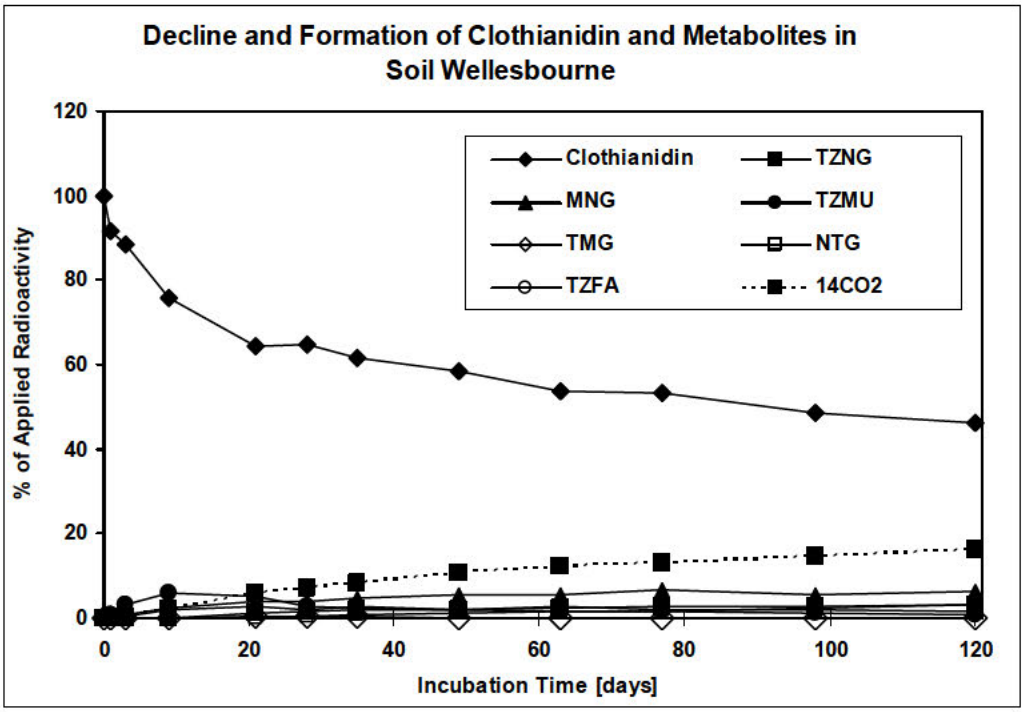
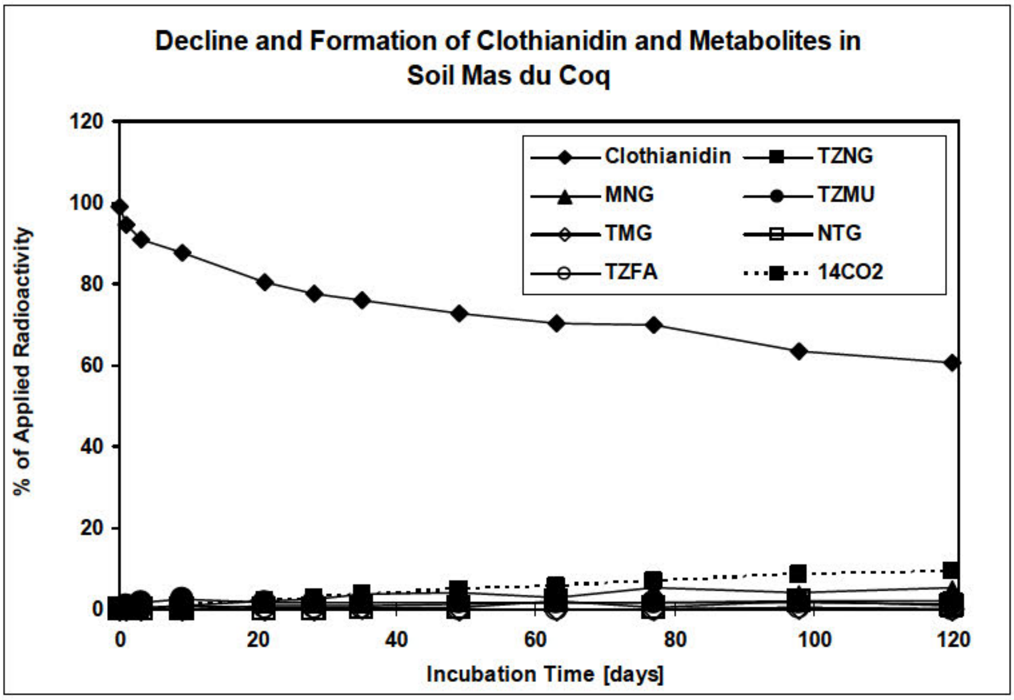


Figure 14: Pattern of Decline of Clothianidin and Formation and Decline of Metabolites (continued)

Soil Mas du Coq



Soil Vilobi d'Onyar

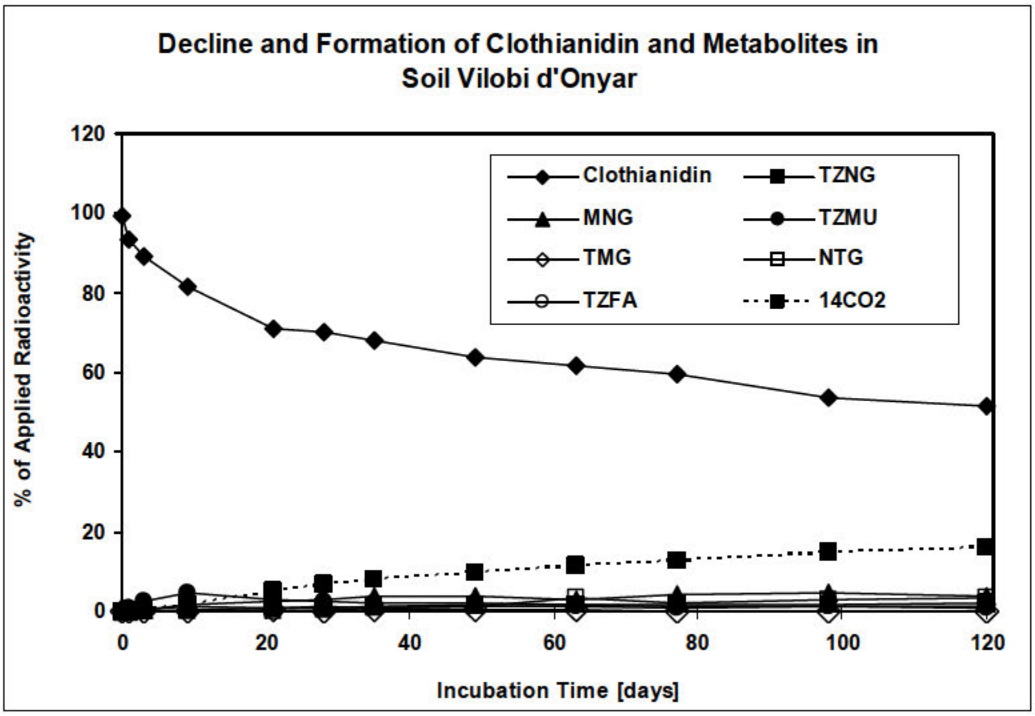


Figure 15: Time-dependent Sorption, Graphical Presentation

Sorption R_{TDS} as a function of time

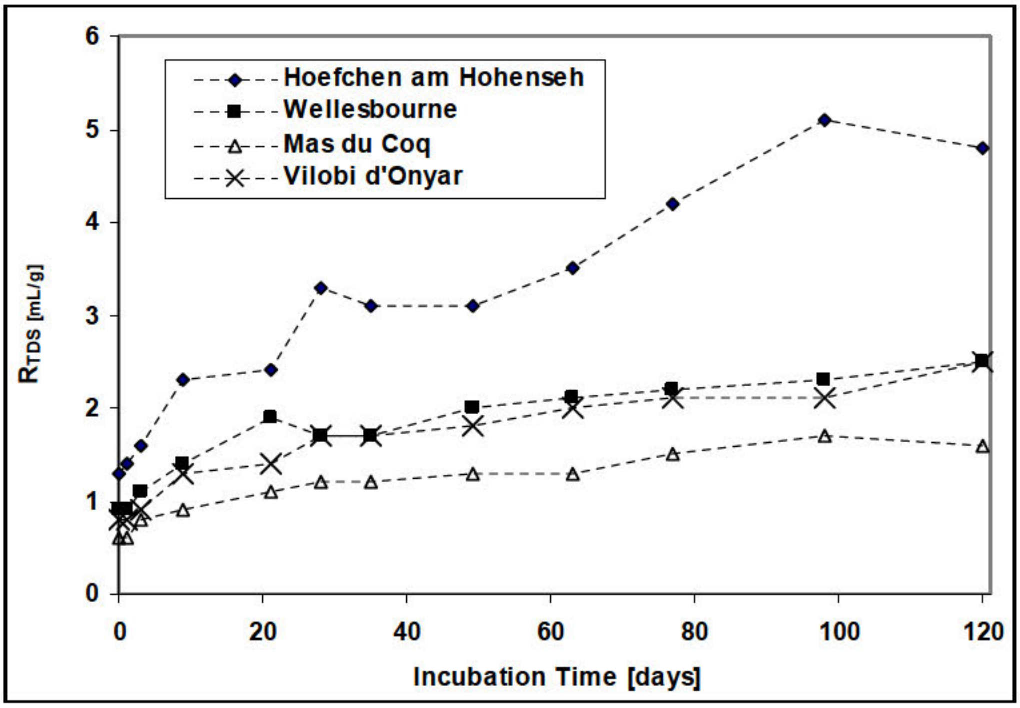


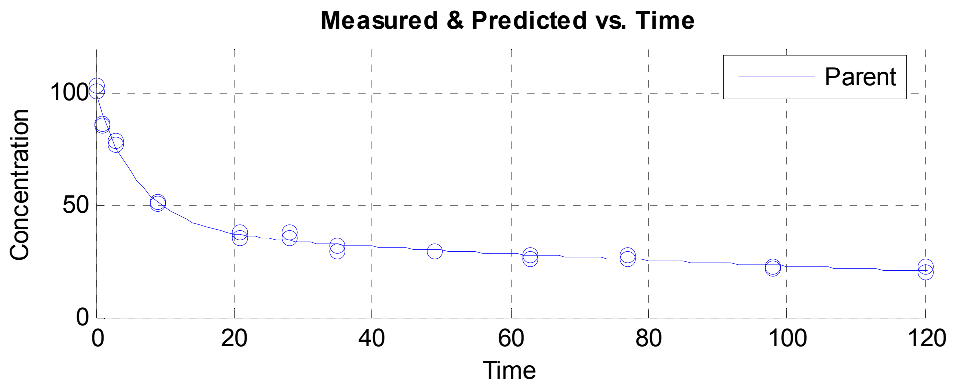
Figure 16: Half-life Determination for Clothianidin in Soil Hoefchen am Hohenseh plot 4011 (Trigger Evaluation)

DFOP

```
Project:             Clothianidin
Testsystem:          TDS on soil
Comment:             Ja57 Hoefchen am Hohenseh 20°C

KinGUI Version: 1.1

Input    Data:                              Q:\D\MEF\Transfer\Unold\M1311911-
7\HoefchenamHohenseh.txt

# =================================
# Results of the kinetic evaluation
# =================================

# ---------------------------------
# Initial values
# ---------------------------------
                     Initial Value    Lower Bound      Upper Bound
        Parent_M(0):   100.0000           0.0000              Inf
          Parent_k1:     0.1000           0.0000              Inf
          Parent_k2:     0.0100           0.0000              Inf
           Parent_g:     0.5000           0.0000              Inf
          Sink_M(0):     0.0000           0.0000              Inf

# ---------------------------------
# Chi2 error estimation
# ---------------------------------
                      Parent      Sink
        Chi2Err%:     3.5791       NaN
   Kinetic Model:       dfop      sink

# ---------------------------------
# Parameter estimation
# ---------------------------------

    Parameter   Estimate     St.Dev   Prob > t
    Parent_k1     0.1606     0.0139   1.3e-010
    Parent_k2     0.0053   7.4e-004   2.7e-007
     Parent_g     0.6089     0.0180   2.0e-019
   Parent_FFS     1.0000
  Parent_M(0)    99.5083     1.2892
    Sink_M(0)     0.0000

# ---------------------------------
# DT50 and DT90 values
# ---------------------------------
                        Parent         Sink
           DT50:        9.6842          NaN
           DT90:      256.4243          NaN
  Kinetic model:          dfop         sink
```

Figure 16: Half-life Determination for Clothianidin in Soil Hoefchen am Hohenseh plot 4011 (Trigger Evaluation) (continued)

```
# --------------------------------
# Measured vs. predicted values
# --------------------------------
   Time               Compartment Parent                 Compartment Sink
          measured predicted  residual  measured predicted  residual
    0.0  100.9000   99.5083    1.3917       NaN    0.0000       NaN
    0.0  103.0000   99.5083    3.4917       NaN    0.0000       NaN
    1.0   85.4000   90.3106   -4.9106       NaN    9.1977       NaN
    1.0   86.6000   90.3106   -3.7106       NaN    9.1977       NaN
    3.0   78.4000   75.7234    2.6766       NaN   23.7849       NaN
    3.0   77.2000   75.7234    1.4766       NaN   23.7849       NaN
    9.0   51.1000   51.3743   -0.2743       NaN   48.1340       NaN
    9.0   51.0000   51.3743   -0.3743       NaN   48.1340       NaN
   21.0   35.6000   36.8846   -1.2846       NaN   62.6237       NaN
   21.0   38.2000   36.8846    1.3154       NaN   62.6237       NaN
   28.0   37.7000   34.2105    3.4895       NaN   65.2978       NaN
   28.0   35.4000   34.2105    1.1895       NaN   65.2978       NaN
   35.0   29.7000   32.5295   -2.8295       NaN   66.9788       NaN
   35.0   32.0000   32.5295   -0.5295       NaN   66.9788       NaN
   49.0   29.3000   30.0150   -0.7150       NaN   69.4933       NaN
   49.0   29.4000   30.0150   -0.6150       NaN   69.4933       NaN
   63.0   26.3000   27.8421   -1.5421       NaN   71.6662       NaN
   63.0   27.5000   27.8421   -0.3421       NaN   71.6662       NaN
   77.0   26.3000   25.8421    0.4579       NaN   73.6662       NaN
   77.0   27.5000   25.8421    1.6579       NaN   73.6662       NaN
   98.0   22.4000   23.1108   -0.7108       NaN   76.3975       NaN
   98.0   22.0000   23.1108   -1.1108       NaN   76.3975       NaN
  120.0   20.4000   20.5587   -0.1587       NaN   78.9496       NaN
  120.0   22.6000   20.5587    2.0413       NaN   78.9496       NaN
```



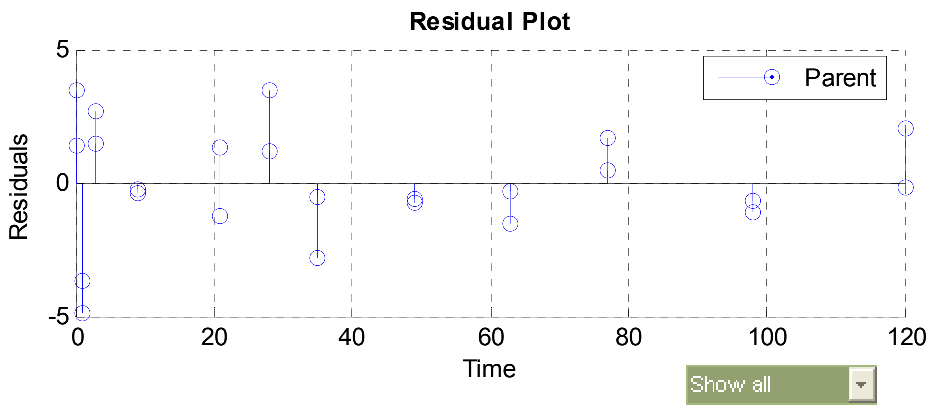


Figure 17: Half-life Determination for Clothianidin in Soil Wellesbourne (Trigger Evaluation)

FOMC

```
Project:             Clothianidin
Testsystem:          TDS on soil
Comment:             Ja57 Wellesbourne 20°C

KinGUI Version: 1.1

Input Data:          Q:\D\MEF\Transfer\Unold\M1311911-7\Wellesbourne.txt

# =================================
# Results of the kinetic evaluation
# =================================

# --------------------------------
# Initial values
# --------------------------------
                    Initial Value    Lower Bound      Upper Bound
        Parent_M(0):   100.0000           0.0000              Inf
       Parent_alpha:     1.0000           0.0000              Inf
        Parent_beta:     1.0000           0.0000              Inf
          Sink_M(0):     0.0000           0.0000              Inf

# --------------------------------
# Chi2 error estimation
# --------------------------------
                     Parent      Sink
        Chi2Err%:    2.2206       NaN
   Kinetic Model:      fomc      sink

# --------------------------------
# Parameter estimation
# --------------------------------

    Parameter   Estimate     St.Dev   Prob > t
 Parent_alpha     0.1909     0.0116   9.0e-014
  Parent_beta     2.3642     0.5273   1.0e-004
   Parent_FFS     1.0000
  Parent_M(0)   102.2162     1.2828
    Sink_M(0)     0.0000

# --------------------------------
# DT50 and DT90 values
# --------------------------------
                       Parent         Sink
          DT50:       86.9035          NaN
          DT90:    >1000.0000          NaN
  Kinetic model:         fomc         sink
```

Figure 17: Half-life Determination for Clothianidin in Soil Wellesbourne (Trigger Evaluation) continued

```
# --------------------------------
# Measured vs. predicted values
# --------------------------------
   Time             Compartment Parent             Compartment Sink
         measured predicted  residual  measured predicted  residual
    0.0  104.5000  102.2162    2.2838       NaN    0.0000       NaN
    0.0  104.0000  102.2162    1.7838       NaN    0.0000       NaN
    1.0   92.0000   95.5601   -3.5601       NaN    6.6561       NaN
    1.0   91.4000   95.5601   -4.1601       NaN    6.6561       NaN
    3.0   87.8000   87.4176    0.3824       NaN   14.7987       NaN
    3.0   89.4000   87.4176    1.9824       NaN   14.7987       NaN
    9.0   75.6000   75.7466   -0.1466       NaN   26.4696       NaN
    9.0   76.1000   75.7466    0.3534       NaN   26.4696       NaN
   21.0   63.0000   66.0106   -3.0106       NaN   36.2057       NaN
   21.0   66.0000   66.0106   -0.0106       NaN   36.2057       NaN
   28.0   65.1000   62.7897    2.3103       NaN   39.4265       NaN
   28.0   64.3000   62.7897    1.5103       NaN   39.4265       NaN
   35.0   61.6000   60.3519    1.2481       NaN   41.8644       NaN
   35.0   61.6000   60.3519    1.2481       NaN   41.8644       NaN
   49.0   58.3000   56.7949    1.5051       NaN   45.4214       NaN
   49.0   58.7000   56.7949    1.9051       NaN   45.4214       NaN
   63.0   52.5000   54.2409   -1.7409       NaN   47.9753       NaN
   63.0   55.2000   54.2409    0.9591       NaN   47.9753       NaN
   77.0   53.2000   52.2683    0.9317       NaN   49.9479       NaN
   77.0   53.3000   52.2683    1.0317       NaN   49.9479       NaN
   98.0   48.0000   49.9777   -1.9777       NaN   52.2385       NaN
   98.0   48.8000   49.9777   -1.1777       NaN   52.2385       NaN
  120.0   46.4000   48.1223   -1.7223       NaN   54.0940       NaN
  120.0   46.0000   48.1223   -2.1223       NaN   54.0940       NaN
```

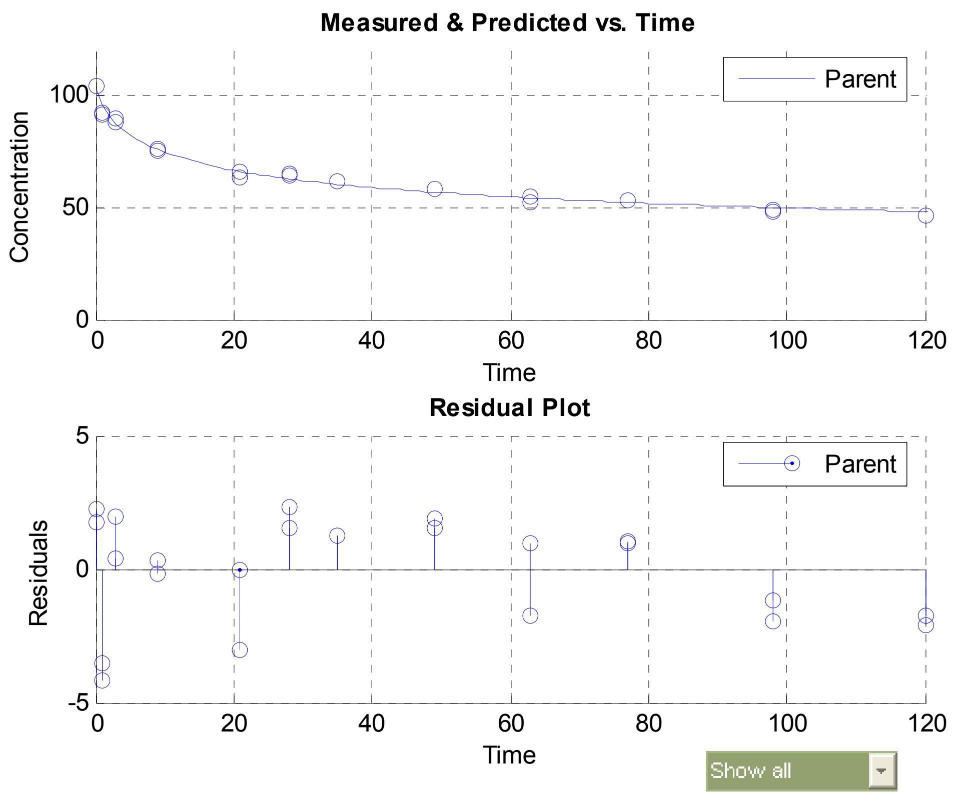


Figure 18: Half-life Determination for Clothianidin in Soil Mas du Coq (Trigger Evaluation)

DFOP

```
Project:             Clothianidin
Testsystem:          TDS on soil
Comment:             Ja57 Mas du Coq 20°C

KinGUI Version: 1.1

Input Data:          Q:\D\MEF\Transfer\Unold\M1311911-7\Mas du Coq.txt

# =================================
# Results of the kinetic evaluation
# =================================

# ---------------------------------
# Initial values
# ---------------------------------
                     Initial Value    Lower Bound      Upper Bound
        Parent_M(0):   100.0000          0.0000              Inf
          Parent_k1:     0.1000          0.0000              Inf
          Parent_k2:     0.0100          0.0000              Inf
           Parent_g:     0.5000          0.0000              Inf
          Sink_M(0):     0.0000          0.0000              Inf

# ---------------------------------
# Chi2 error estimation
# ---------------------------------
                      Parent      Sink
        Chi2Err%:     1.8438       NaN
   Kinetic Model:       dfop      sink

# ---------------------------------
# Parameter estimation
# ---------------------------------

    Parameter   Estimate     St.Dev   Prob > t
    Parent_k1     0.3367     0.0891   5.9e-004
    Parent_k2     0.0030   2.1e-004   2.1e-012
     Parent_g     0.1553     0.0143   4.0e-010
   Parent_FFS     1.0000
  Parent_M(0)   101.6078     1.2816
    Sink_M(0)     0.0000

# ---------------------------------
# DT50 and DT90 values
# ---------------------------------
                        Parent         Sink
           DT50:      174.5501          NaN
           DT90:      710.2405          NaN
  Kinetic model:          dfop         sink
```

Figure 18: Half-life Determination for Clothianidin in Soil Mas du Coq (Trigger Evaluation), continued

```
# ---------------------------------
# Measured vs. predicted values
# ---------------------------------
```

Time	Compartment Parent measured	Compartment Parent predicted	Compartment Parent residual	Compartment Sink measured	Compartment Sink predicted	Compartment Sink residual
0.0	102.6000	101.6078	0.9922	NaN	0.0000	NaN
0.0	104.1000	101.6078	2.4922	NaN	0.0000	NaN
1.0	93.9000	96.8403	-2.9403	NaN	4.7675	NaN
1.0	94.8000	96.8403	-2.0403	NaN	4.7675	NaN
3.0	90.2000	90.8071	-0.6071	NaN	10.8007	NaN
3.0	91.9000	90.8071	1.0929	NaN	10.8007	NaN
9.0	87.2000	84.3043	2.8957	NaN	17.3034	NaN
9.0	88.4000	84.3043	4.0957	NaN	17.3034	NaN
21.0	79.4000	80.5975	-1.1975	NaN	21.0103	NaN
21.0	81.7000	80.5975	1.1025	NaN	21.0103	NaN
28.0	77.1000	78.9083	-1.8083	NaN	22.6994	NaN
28.0	78.2000	78.9083	-0.7083	NaN	22.6994	NaN
35.0	75.6000	77.2650	-1.6650	NaN	24.3428	NaN
35.0	76.0000	77.2650	-1.2650	NaN	24.3428	NaN
49.0	73.4000	74.0824	-0.6824	NaN	27.5254	NaN
49.0	71.9000	74.0824	-2.1824	NaN	27.5254	NaN
63.0	70.8000	71.0310	-0.2310	NaN	30.5768	NaN
63.0	69.6000	71.0310	-1.4310	NaN	30.5768	NaN
77.0	70.8000	68.1052	2.6948	NaN	33.5025	NaN
77.0	69.3000	68.1052	1.1948	NaN	33.5025	NaN
98.0	62.7000	63.9410	-1.2410	NaN	37.6667	NaN
98.0	64.1000	63.9410	0.1590	NaN	37.6667	NaN
120.0	61.3000	59.8514	1.4486	NaN	41.7564	NaN
120.0	60.2000	59.8514	0.3486	NaN	41.7564	NaN

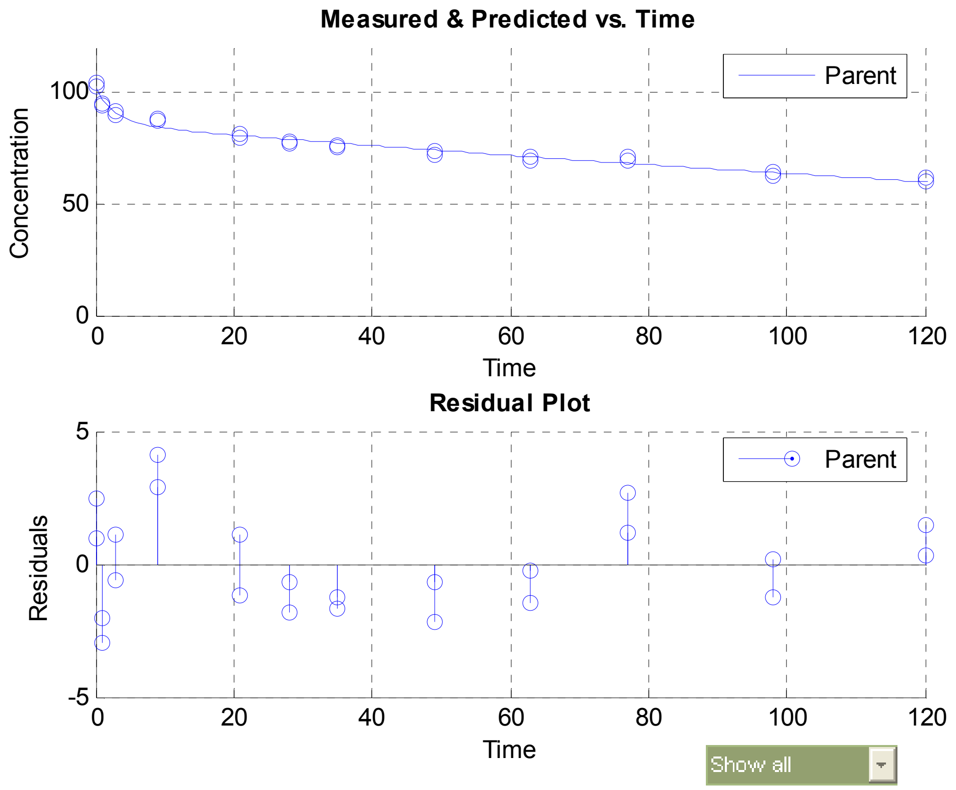


Figure 19: Half-life Determination for Clothianidin in Soil Vilobi d'Onyar (Trigger Evaluation)

DFOP

```
Project:             Clothianidin
Testsystem:          TDS on soil
Comment:             Ja57 Vilobi d'Onyar 20°C

KinGUI Version: 1.1

Input Data:          Q:\D\MEF\Transfer\Unold\M1311911-7\Vilobi d Onyar.txt

# =================================
# Results of the kinetic evaluation
# =================================

# ---------------------------------
# Initial values
# ---------------------------------
                    Initial Value    Lower Bound      Upper Bound
        Parent_M(0):   100.0000          0.0000              Inf
          Parent_k1:     0.1000          0.0000              Inf
          Parent_k2:     0.0100          0.0000              Inf
           Parent_g:     0.5000          0.0000              Inf
          Sink_M(0):     0.0000          0.0000              Inf

# ---------------------------------
# Chi2 error estimation
# ---------------------------------
                     Parent      Sink
        Chi2Err%:    1.7996       NaN
   Kinetic Model:      dfop      sink

# ---------------------------------
# Parameter estimation
# ---------------------------------

    Parameter   Estimate     St.Dev   Prob > t
    Parent_k1     0.1749     0.0295   4.2e-006
    Parent_k2     0.0035   2.5e-004   4.4e-012
     Parent_g     0.2365     0.0136   7.7e-014
   Parent_FFS     1.0000
  Parent_M(0)   100.5908     1.0411
    Sink_M(0)     0.0000

# ---------------------------------
# DT50 and DT90 values
# ---------------------------------
                       Parent         Sink
          DT50:     121.8891          NaN
          DT90:     585.3275          NaN
  Kinetic model:        dfop         sink
```

Figure 19: Half-life Determination for Clothianidin in Soil Vilobi d'Onyar (Trigger Evaluation), continued

```
# ---------------------------------
# Measured vs. predicted values
# ---------------------------------
   Time            Compartment Parent                Compartment Sink
         measured predicted  residual  measured predicted  residual
    0.0  102.3000  100.5908    1.7092       NaN    0.0000       NaN
    0.0  103.9000  100.5908    3.3092       NaN    0.0000       NaN
    1.0   94.1000   96.5063   -2.4063       NaN    4.0845       NaN
    1.0   92.8000   96.5063   -3.7063       NaN    4.0845       NaN
    3.0   90.8000   90.0801    0.7199       NaN   10.5107       NaN
    3.0   87.8000   90.0801   -2.2801       NaN   10.5107       NaN
    9.0   81.0000   79.3645    1.6355       NaN   21.2264       NaN
    9.0   82.4000   79.3645    3.0355       NaN   21.2264       NaN
   21.0   71.3000   72.0027   -0.7027       NaN   28.5881       NaN
   21.0   71.0000   72.0027   -1.0027       NaN   28.5881       NaN
   28.0   70.5000   69.8616    0.6384       NaN   30.7292       NaN
   28.0   70.2000   69.8616    0.3384       NaN   30.7292       NaN
   35.0   67.2000   68.0628   -0.8628       NaN   32.5281       NaN
   35.0   68.6000   68.0628    0.5372       NaN   32.5281       NaN
   49.0   64.3000   64.7876   -0.4876       NaN   35.8032       NaN
   49.0   63.6000   64.7876   -1.1876       NaN   35.8032       NaN
   63.0   61.4000   61.7091   -0.3091       NaN   38.8817       NaN
   63.0   61.6000   61.7091   -0.1091       NaN   38.8817       NaN
   77.0   60.2000   58.7803    1.4197       NaN   41.8106       NaN
   77.0   58.8000   58.7803    0.0197       NaN   41.8106       NaN
   98.0   53.4000   54.6460   -1.2460       NaN   45.9448       NaN
   98.0   53.6000   54.6460   -1.0460       NaN   45.9448       NaN
  120.0   51.1000   50.6265    0.4735       NaN   49.9644       NaN
  120.0   52.1000   50.6265    1.4735       NaN   49.9644       NaN
```

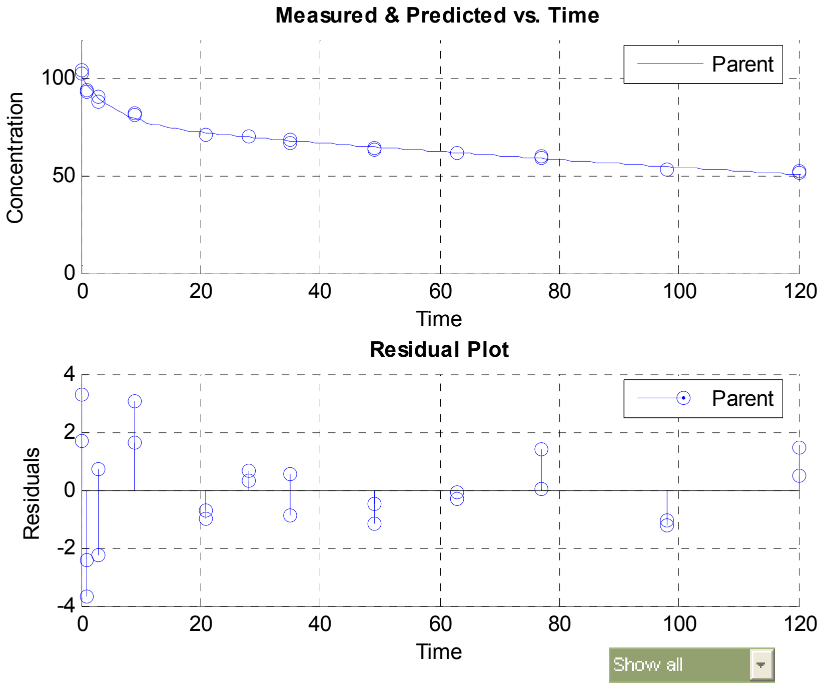
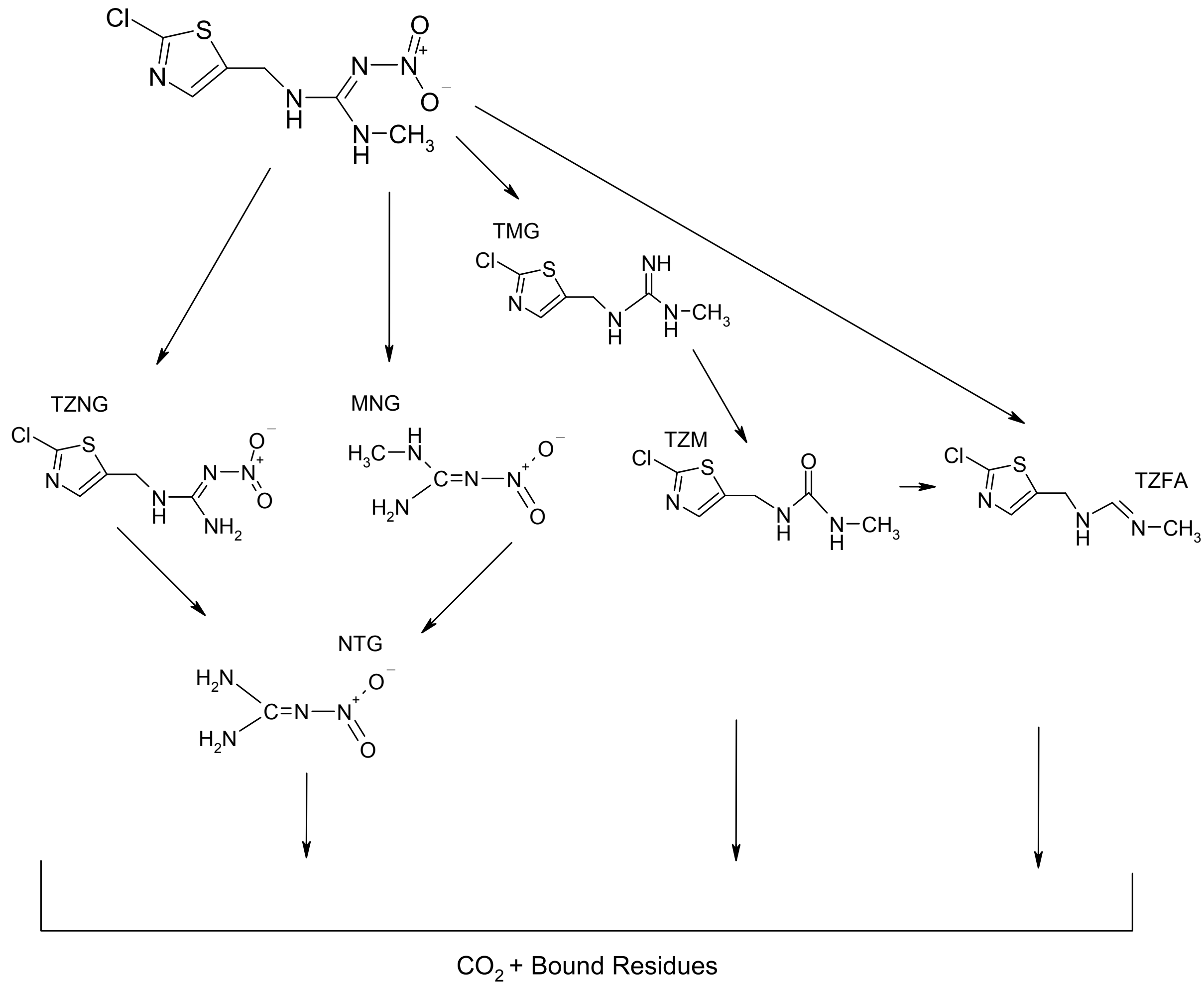


Figure 20: Proposed Metabolic Pathway of Clothianidin in Soil under Aerobic Conditions



List of Appendices

Appendix 1: Reagents and Equipment

Article	**Supplier**	**Type**
Analytical balance	Mettler-Toledo	XP205
Analytical balance	Sartorius	1712 004
Centrifuge	Eppendorf	5415R
Centrifuge	Beckman	Avanti J-26 XP
Climatic chamber	Bayer CropScience	Room 163 of test facility
Cotton wool	Amicus	167307
Data logger	Grant Instruments	Squirrel 1206
Folded filter	Schleicher & Schüll	----------
Halogen Moisture Analyzer	Mettler Toledo	HB43
Halogen Moisture Printer	Mettler Toledo	GA42
HPLC	Agilent	1100
HPLC column	Merck	Purospher STAR
HPLC detector	Raytest	Ramona Star
Laboratory balance	Sartorius	1507
Liquid scintillation counter	Beckman	LS 6500
Liquid scintillation counter	Beckman	LS 6000 LL
Liquid scintillation counter	Perkin-Elmer Live Science	LKB 1219 Spectral
Lyophilization	Christ	Alpha 2-4 LD
Micro wave extractor	MLS GmbH	ETHOS 1600
Oxidizer	Peerless	Robotic autosampler
Oxidizer	Zinsser Analytic	OX 500
Pipette 100-5000	Eppendorf	Researchpro
Pipette 20-300	Eppendorf	Researchpro
Pipette 50-1000	Eppendorf	Researchpro
Planetary ball mill	Retsch	PM 4000
Polyvails	Zinsser Analytic	20 mL
Polyvails	Zinsser Analytic	6 mL
Shaker	Bühler	SM25
Sieve (2 mm)	Retsch	DIN ISO 3310-1
Sieving maschine	Retsch	AS 400
Temperature sensor	Grant Instruments	Thermistor
TLC - radioactivty - scanner	Fuji	Image analyzer, Fujix BAS 2000
TLC plate spotter	Camag	Linomat ATS4
TLC plates	Merck	Si60 / F254
Ultra sonic bath	Bransonic	220
Vacuum concentrator	Christ	Alpha RVC 2-33

Appendix 1: Reagents and Equipment (continued)

Article	Supplier	Order Number
Acetonitrile	Merck	1.00030.9010
Acetonitrile	Krämer+Martin	17-225
Barium chloride dihydrate	Sigma-Aldrich	31125
Ethyl acetate	Merck	1.09623
Formic acid	Merck	1.00264
Hydrochloric acid	Kreamer & Martin	02-740
Methanol	Merck	1.06009.1011
Propanol -2- (Isopropanol)	Merck	1.09634.1000
Scintillator Carbosorb E	Perkin-Elmer-Live-Science	6013729
Scintillator Permafluor E+	Perkin-Elmer-Live-Science	6013187
Scintillator Quicksafe A	Zinsser Analytic	1008000
Scintillator Rotaszint eco plus	Roth	0016.2
Soda lime	Riedel	31474
Sodium hydroxide	Merck	1.06498.1000
Szintillator Oxysolve C400	Zinsser Analytic	1691400
Triethylamine	Merck	8.08352.2500
Water, demineralized	Bayer AG	---
Water, demineralized	TKA	08.2206

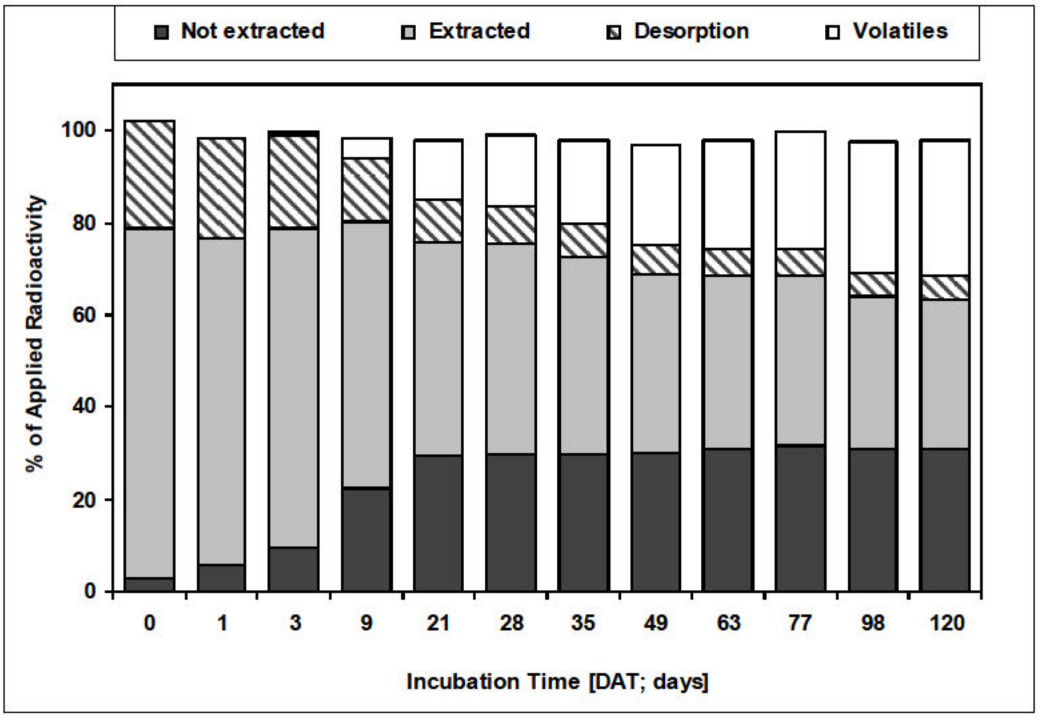
Appendix 2: Description of Soil Collection and Storage (DER Table 1)

Parameter	Hoefchen am Hohenseh plot 4011	Wellesbourne	Mas du Coq	Vilobi d'Onyar
Geographic location City: State: Country: GPS coordinates: Field plot:	 Burscheid NRW* Germany N/A plot 4011	 Warwick HRI, Wellesbourne Warwickshire United Kingdom N/A N/A	 St. Etienné du Gres N/A France N 43° 48'19 E 004° 43' 10 Tarascon Mas du Coq	 Vilobi d' Onyar N/A Spain WGS 84/ UTM x479104 y 463666 plot 54
Site Description	soil dissipation field, grassland	stuble	Grassland	Usual crops: Grass, corn, cereals
Pesticide use history at the collection site	No information	No use for at least 3 years	R2006 0857/8 AE 1170437 (500SC) (A1 le 15/05/2006)	No information
Collection date	2009-09-03	2009-09-18	2009-08-31	2009-09-18
Collection procedures	With a spade from 4 different points of the plot	Samples taken with a spade and placed in plastic bag	With a spade from 10 different spots, stored in plastic bags	Sampling device with tractor
Sampling depth (cm)	0 – 20 cm	0 – 20 cm	0 – 20 cm	0 – 20 cm
Shipping date	2009-09-03	N/A	2009-09-04 (soil delivery)	2009-09-18
Shipping conditions	By car at ambient temperature	With DHL	Truck transport at ambient conditions	With DHL
Storage conditions at the receiving facility	After sieving to > 2 mm, the soils were stored at ambient temperature			
Storage length prior to use	< 1 month	About 10 days	about 1 month	About 10 days
Soil preparation prior to use	Soil was successively sieved to ≤ 20, 10, 5, 3.35 and 2 mm			

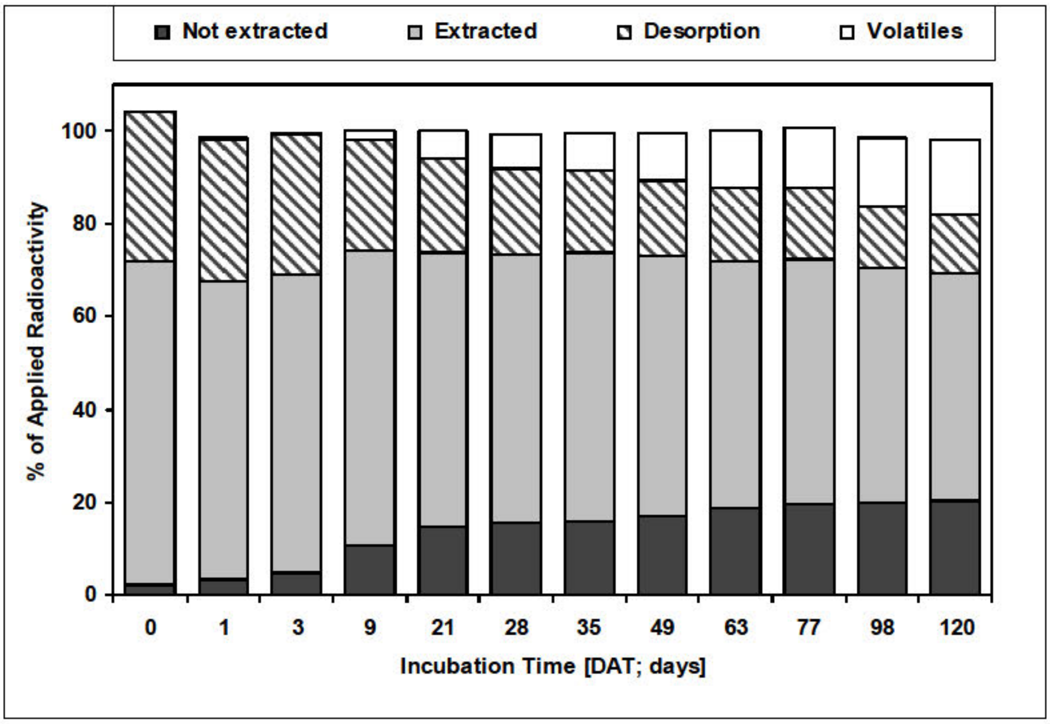
*) NRW = Northrhine Westphalia

Appendix 3: Material Balance of Radioactivity (graphic presentation, expressed as percent of applied radiocarbon)

Soil Hoefchen am Hohenseh plot 4011

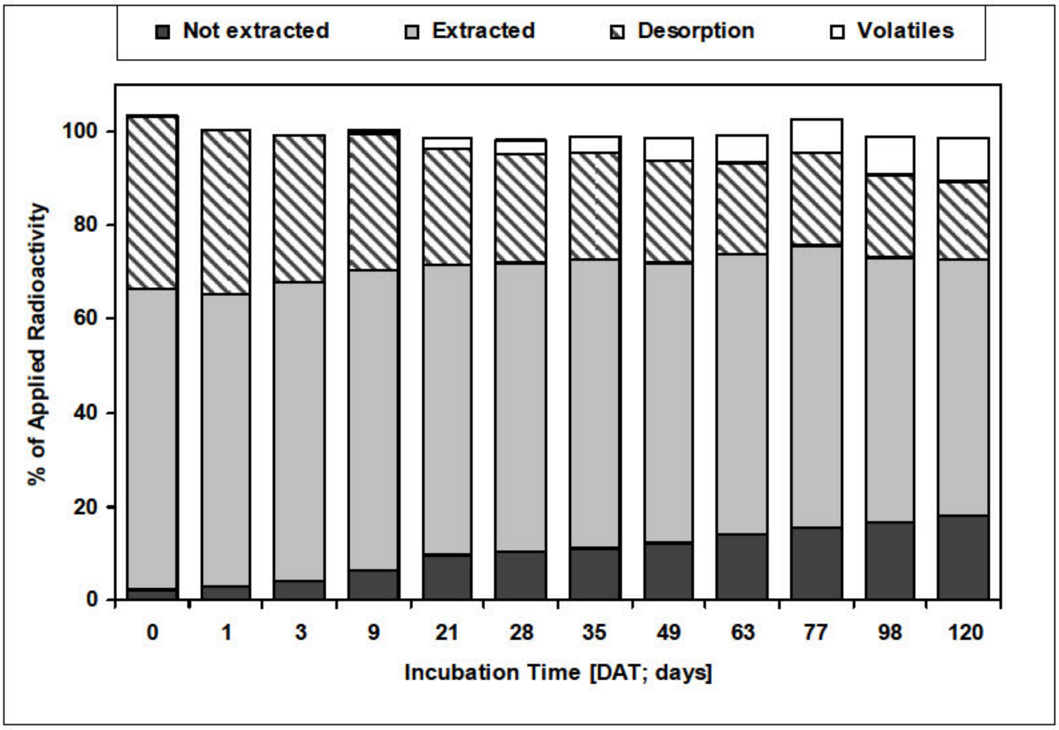


Soil Wellesbourne

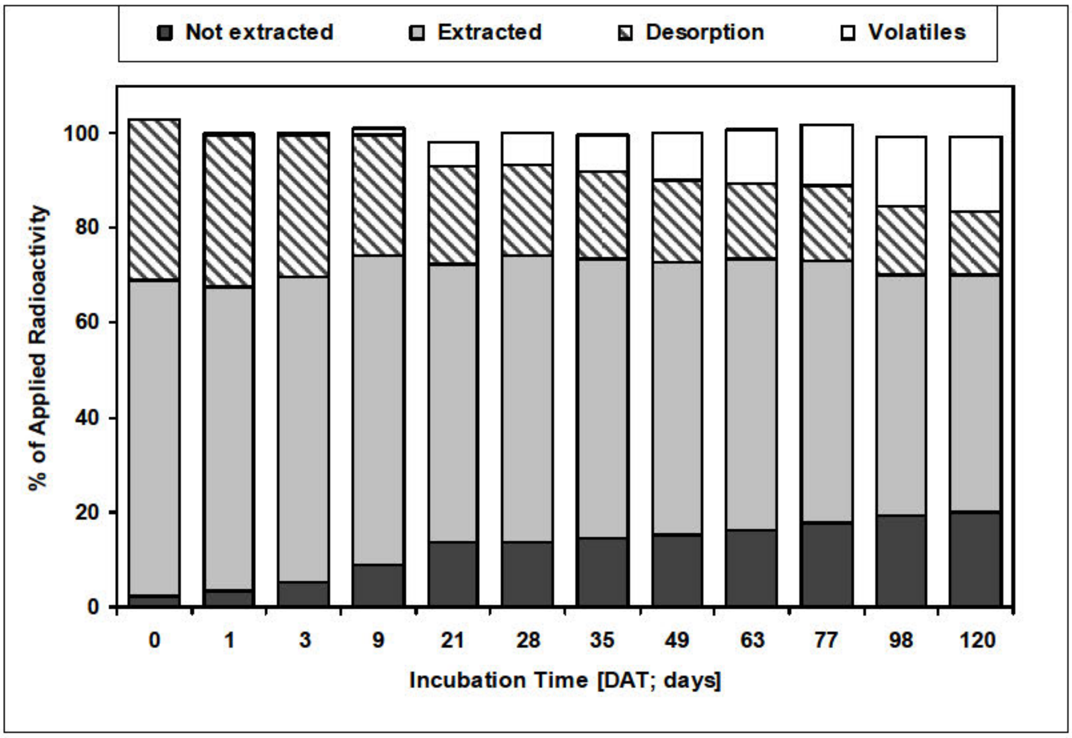


Appendix 3: Material Balance of Radioactivity (graphic presentation, expressed as percent of applied radiocarbon), continued

Soil Mas du Coq



Soil Viloby d'Onyar



Appendix 4: Biotransformation of Clothianidin, Expressed as Percentage of Applied Radioactivity, in Soil Hoefchen am Hohenseh plot 4011 (Replicate A, B)

	Replicate	DAT											
Compound	**No.**	**0**	**1**	**3**	**9**	**21**	**28**	**35**	**49**	**63**	**77**	**98**	**120**
Clothianidin	A	96.3	85.4	78.4	51.1	35.6	37.7	29.7	29.3	26.3	26.3	22.4	20.4
	B	97.5	86.6	77.2	51.0	38.2	35.4	32.0	29.4	27.5	27.5	22.0	22.6
	Mean	**96.9**	**86.0**	**77.8**	**51.1**	**36.9**	**36.6**	**30.9**	**29.4**	**26.9**	**26.9**	**22.2**	**21.5**
TZNG	A	n.d.	n.d.	n.d.	n.d.	0.5	0.6	0.4	0.8	0.9	1.0	1.1	1.1
	B	n.d.	n.d.	0.1	0.2	0.5	0.5	0.7	0.7	0.8	1.0	1.0	1.1
	Mean			**0.0**	**0.1**	**0.5**	**0.5**	**0.6**	**0.8**	**0.8**	**1.0**	**1.0**	**1.1**
MNG	A	n.d.	0.6	1.5	2.4	3.0	3.5	4.5	4.5	5.1	5.9	3.7	6.4
	B	n.d.	0.4	1.1	2.6	3.1	3.6	4.8	5.2	4.5	5.4	5.2	6.4
	Mean		**0.5**	**1.3**	**2.5**	**3.1**	**3.6**	**4.7**	**4.8**	**4.8**	**5.6**	**4.5**	**6.4**
TZMU	A	0.5	2.3	6.4	10.6	7.3	5.4	4.3	3.0	2.5	2.1	1.4	1.1
	B	0.5	2.2	5.9	10.5	6.5	5.5	4.0	2.8	1.9	2.1	1.4	1.2
	Mean	**0.5**	**2.3**	**6.2**	**10.6**	**6.9**	**5.4**	**4.2**	**2.9**	**2.2**	**2.1**	**1.4**	**1.2**
TMG	A	n.d.	0.3	0.3	0.6	0.6	0.8	0.9	0.9	0.6	0.7	0.4	0.6
	B	n.d.	n.d.	0.3	0.6	0.7	0.8	0.6	0.4	0.7	0.9	0.7	0.4
	Mean		**0.1**	**0.3**	**0.6**	**0.6**	**0.8**	**0.8**	**0.7**	**0.6**	**0.8**	**0.6**	**0.5**
NTG	A	n.d.	0.1	<LOQ	n.d.	0.2	0.2	0.8	0.6	1.4	1.3	3.4	2.3
	B	n.d.	0.1	n.d.	0.1	0.2	0.2	0.8	0.7	1.6	1.7	2.2	2.1
	Mean		**0.1**		**0.0**	**0.2**	**0.2**	**0.8**	**0.6**	**1.5**	**1.5**	**2.8**	**2.2**
TZFA	A	n.d.	0.8	1.8	3.7	5.2	4.1	6.8	3.7	4.5	4.0	4.3	3.9
	B	n.d.	0.7	1.8	4.7	5.3	5.8	6.6	5.0	5.5	4.1	4.9	3.6
	Mean		**0.8**	**1.8**	**4.2**	**5.3**	**4.9**	**6.7**	**4.3**	**5.0**	**4.0**	**4.6**	**3.8**
Unidentified radioactivity	A	1.2	2.3	1.7	2.2	2.5	1.8	1.4	2.0	1.2	0.8	1.7	1.2
	B	1.9	2.1	1.9	2.6	1.3	1.6	1.3	1.4	1.8	0.7	1.1	0.9
	Mean	**1.6**	**2.2**	**1.8**	**2.4**	**1.9**	**1.7**	**1.3**	**1.7**	**1.5**	**0.8**	**1.4**	**1.1**
Total extractable residues	A	98.0	91.8	90.2	70.5	54.8	54.1	48.9	44.7	42.6	42.0	38.4	37.0
	B	99.9	92.2	88.2	72.2	55.7	53.4	50.8	45.7	44.2	43.4	38.5	38.4
	Mean	**99.0**	**92.0**	**89.2**	**71.4**	**55.2**	**53.8**	**49.8**	**45.2**	**43.4**	**42.7**	**38.4**	**37.7**
$^{14}CO_2$	A	n.a.	0.2	0.8	4.3	12.8	15.1	18.5	21.6	23.6	25.4	28.2	29.4
	B	n.a.	0.3	0.9	4.5	13.5	15.9	18.2	21.3	23.8	25.7	28.1	29.2
	Mean		**0.2**	**0.8**	**4.4**	**13.1**	**15.5**	**18.3**	**21.5**	**23.7**	**25.5**	**28.2**	**29.3**
Volatile organics	A	n.a.	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	0.3
	B	n.a.	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1
	Mean												**0.2**
Non-extractable residue	A	2.9	6.3	9.1	22.6	30.5	29.5	30.2	30.0	30.9	31.2	30.5	30.8
	B	3.0	5.8	10.1	22.3	28.8	29.7	29.4	30.0	30.7	31.9	31.0	30.7
	Mean	**3.0**	**6.1**	**9.6**	**22.5**	**29.6**	**29.6**	**29.8**	**30.0**	**30.8**	**31.5**	**30.7**	**30.8**
Total % recovery	A	100.9	98.4	100.1	97.4	98.0	98.8	97.5	96.3	97.2	98.6	97.1	97.5
	B	103.0	98.3	99.2	99.0	97.9	99.0	98.5	97.0	98.7	101.0	97.6	98.3
	Mean	**101.9**	**98.3**	**99.6**	**98.2**	**98.0**	**98.9**	**98.0**	**96.7**	**97.9**	**99.8**	**97.3**	**97.9**

n.d.: not detected, n.a.: not analyzed, DAT: day after treatment
See Table 5 for Material balance.

Appendix 5: Biotransformation of Clothianidin, Expressed as Percentage of Applied Radioactivity, in Soil Wellesbourne (Replicate C and D)

	Replicate	DAT											
Compound	**No.**	**0**	**1**	**3**	**9**	**21**	**28**	**35**	**49**	**63**	**77**	**98**	**120**
Clothianidin	C	100.4	92.0	87.8	75.6	63.0	65.1	61.6	58.3	52.5	53.2	48.0	46.4
	D	99.1	91.4	89.4	76.1	66.0	64.3	61.6	58.7	55.2	53.3	48.8	46.0
	Mean	**99.8**	**91.7**	**88.6**	**75.8**	**64.5**	**64.7**	**61.6**	**58.5**	**53.8**	**53.2**	**48.4**	**46.2**
TZNG	C	n.d.	n.d.	n.d.	0.1	1.0	1.6	2.0	2.0	2.3	2.8	2.8	2.8
	D	n.d.	0.1	0.1	0.3	1.2	1.7	2.0	2.2	2.3	2.7	3.0	3.3
	Mean		**0.0**	**0.0**	**0.2**	**1.1**	**1.7**	**2.0**	**2.1**	**2.3**	**2.7**	**2.9**	**3.0**
MNG	C	n.d.	0.8	0.8	2.2	4.3	4.2	4.7	5.6	5.6	7.3	6.2	6.5
	D	n.d.	0.3	0.7	2.2	3.7	3.6	4.8	5.9	5.3	6.4	5.1	6.2
	Mean		**0.6**	**0.8**	**2.2**	**4.0**	**3.9**	**4.7**	**5.7**	**5.4**	**6.8**	**5.6**	**6.4**
TZMU	C	n.d.	0.9	2.7	6.2	6.0	3.3	2.6	2.1	1.6	1.5	1.0	0.9
	D	n.d.	0.8	3.2	5.7	4.2	2.6	2.5	2.1	1.8	1.5	1.0	1.1
	Mean		**0.9**	**3.0**	**5.9**	**5.1**	**3.0**	**2.5**	**2.1**	**1.7**	**1.5**	**1.0**	**1.0**
TMG	C	n.d.	n.d.	0.3	0.2	0.2	0.3	0.4	<LOQ	0.2	n.d.	0.2	0.1
	D	n.d.	0.1	n.d.	n.d.	0.2	0.3	0.3	0.2	<LOQ	n.d.	0.1	0.1
	Mean		**0.0**	**0.1**	**0.1**	**0.2**	**0.3**	**0.3**	**0.1**	**0.1**		**0.2**	**0.1**
NTG	C	n.d.	n.d.	0.1	0.1	0.1	0.2	0.7	1.0	1.9	1.3	2.6	2.7
	D	n.d.	n.d.	n.d.	n.d.	0.1	0.5	0.6	1.3	1.5	1.5	2.5	3.5
	Mean			**0.1**	**0.1**	**0.1**	**0.3**	**0.6**	**1.2**	**1.7**	**1.4**	**2.5**	**3.1**
TZFA	C	n.d.	n.d.	0.8	2.0	3.2	2.4	2.9	1.8	2.8	1.8	1.7	1.8
	D	n.d.	n.d.	0.1	1.9	2.4	1.9	2.6	2.3	2.4	2.1	2.1	1.3
	Mean			**0.4**	**1.9**	**2.8**	**2.1**	**2.7**	**2.0**	**2.6**	**1.9**	**1.9**	**1.5**
Unidentified radioactivity	C	2.0	1.4	0.9	1.2	1.5	0.4	0.7	0.5	1.0	0.4	1.1	0.6
	D	2.6	1.7	2.0	1.1	1.6	0.7	1.2	0.4	1.5	1.0	1.4	0.4
	Mean	**2.3**	**1.6**	**1.5**	**1.1**	**1.5**	**0.5**	**1.0**	**0.4**	**1.3**	**0.7**	**1.3**	**0.5**
Total extractable residues	C	102.4	95.1	93.4	87.5	79.4	77.5	75.4	71.3	67.9	68.3	63.5	61.8
	D	101.6	94.5	95.6	87.2	79.4	75.6	75.5	73.0	69.9	68.5	64.1	61.9
	Mean	**102.0**	**94.8**	**94.5**	**87.4**	**79.4**	**76.5**	**75.5**	**72.1**	**68.9**	**68.4**	**63.8**	**61.8**
$^{14}CO_2$	C	n.a.	0.2	0.3	2.0	5.8	7.3	8.2	10.4	12.2	13.2	15.0	16.1
	D	n.a.	0.2	0.4	2.0	6.0	7.2	8.2	10.9	12.3	12.9	14.5	16.4
	Mean		**0.2**	**0.3**	**2.0**	**5.9**	**7.3**	**8.2**	**10.6**	**12.3**	**13.1**	**14.8**	**16.2**
Volatile organics	C	n.a.	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1
	D	n.a.	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1
	Mean												
Non-extractable residue	C	2.1	3.6	4.6	10.8	14.4	15.1	15.9	17.236	18.8	19.5	20.0	20.3
	D	2.3	3.3	4.9	10.9	14.8	16.0	16.2	16.9	18.8	19.3	19.8	20.1
	Mean	**2.2**	**3.5**	**4.7**	**10.8**	**14.6**	**15.5**	**16.0**	**17.0**	**18.8**	**19.4**	**19.9**	**20.2**
Total % recovery	C	104.5	98.9	98.3	100.3	99.6	99.9	99.6	98.9	98.9	101.0	98.5	98.1
	D	104.0	98.0	100.8	100.1	100.3	98.8	99.9	100.7	101.0	100.6	98.4	98.4
	Mean	**104.2**	**98.4**	**99.6**	**100.2**	**99.9**	**99.3**	**99.7**	**99.8**	**100.0**	**100.8**	**98.5**	**98.2**

n.d.: not detected, n.a.: not analyzed, DAT: day after treatment
See Table 6 for Material Balance.

Appendix 6: Biotransformation of Clothianidin, Expressed as Percentage of Applied Radioactivity, in Soil Mas du Coq (Replicate E and F)

Compound	Replicate No.	DAT 0	1	3	9	21	28	35	49	63	77	98	120
Clothianidin	E	98.9	93.9	90.2	87.2	79.4	77.1	75.6	73.4	70.8	70.8	62.7	61.3
	F	99.5	94.8	91.9	88.4	81.7	78.2	76.0	71.9	69.6	69.3	64.1	60.2
	Mean	**99.2**	**94.4**	**91.0**	**87.8**	**80.6**	**77.6**	**75.8**	**72.6**	**70.2**	**70.1**	**63.4**	**60.8**
TZNG	E	n.d.	n.d.	n.d.	0.1	0.8	1.0	0.9	1.1	1.6	1.6	2.2	2.2
	F	n.d.	n.d.	n.d.	0.2	0.6	1.0	0.9	1.6	1.5	2.0	1.8	2.0
	Mean				**0.1**	**0.7**	**1.0**	**0.9**	**1.3**	**1.6**	**1.8**	**2.0**	**2.1**
MNG	E	n.d.	0.2	0.6	0.9	2.3	2.9	3.4	4.0	2.9	5.9	4.3	5.2
	F	n.d.	0.5	n.d.	1.0	2.1	2.0	3.6	4.0	2.8	4.5	3.4	5.5
	Mean		**0.4**	**0.3**	**0.9**	**2.2**	**2.5**	**3.5**	**4.0**	**2.9**	**5.2**	**3.9**	**5.3**
TZMU	E	n.d.	0.7	1.8	2.0	2.0	1.8	1.8	1.7	1.6	1.4	1.3	1.0
	F	n.d.	1.0	1.8	2.5	1.6	1.8	1.8	1.8	1.5	1.6	1.6	1.4
	Mean		**0.8**	**1.8**	**2.2**	**1.8**	**1.8**	**1.8**	**1.7**	**1.6**	**1.5**	**1.5**	**1.2**
TMG	E	n.d.	n.d.	n.d.	0.1	n.d.	0.1	0.1	n.d.	n.d.	0.3	0.3	0.1
	F	n.d.	n.d.	0.1	n.d.	0.1	<LOQ	0.3	0.1	0.4	<LOQ	0.3	0.2
	Mean			**0.0**	**0.0**	**0.1**	**0.0**	**0.2**	**0.1**	**0.2**	**0.1**	**0.3**	**0.2**
NTG	E	n.d.	n.d.	0.1	0.1	n.d.	0.3	0.4	0.4	2.1	0.3	1.9	0.9
	F	n.d.	0.3	n.d.	0.2	n.d.	n.d.	n.d.	0.1	2.2	0.1	2.2	0.9
	Mean		**0.2**	**0.1**	**0.1**		**0.2**	**0.2**	**0.2**	**2.1**	**0.2**	**2.1**	**0.9**
TZFA	E	n.d.	n.d.	0.1	0.8	0.6	0.5	0.7	0.2	n.d.	n.d.	0.4	<LOQ
	F	n.d.	n.d.	n.d.	0.6	0.3	0.4	0.5	0.2	0.2	n.d.	0.2	0.1
	Mean			**0.1**	**0.7**	**0.4**	**0.4**	**0.6**	**0.2**	**0.1**		**0.3**	**0.0**
Unidentified radioactivity	E	1.6	1.0	2.0	1.4	1.3	1.7	0.8	0.9	0.5	0.9	0.9	0.8
	F	2.0	2.1	1.6	1.0	0.8	1.0	1.8	1.8	1.3	1.5	0.8	0.7
	Mean	**1.8**	**1.6**	**1.8**	**1.2**	**1.0**	**1.4**	**1.3**	**1.3**	**0.9**	**1.2**	**0.8**	**0.8**
Total extractable residues	E	100.5	95.9	94.9	92.7	86.3	85.3	83.7	81.7	79.6	81.3	74.0	71.4
	F	101.5	98.7	95.2	93.7	87.3	84.4	84.8	81.4	79.4	79.0	74.5	71.0
	Mean	**101.0**	**97.3**	**95.0**	**93.2**	**86.8**	**84.8**	**84.2**	**81.6**	**79.5**	**80.1**	**74.2**	**71.2**
$^{14}CO_2$	E	n.a.	0.1	0.2	0.7	2.0	3.2	3.5	4.8	5.8	6.9	8.2	9.1
	F	n.a.	0.1	0.2	0.7	2.1	2.9	3.5	4.9	5.8	6.8	8.4	9.6
	Mean		**0.1**	**0.2**	**0.7**	**2.0**	**3.0**	**3.5**	**4.8**	**5.8**	**6.8**	**8.3**	**9.4**
Volatile organics	E	n.a.	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1
	F	n.a.	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1
	Mean												
Non-extractable residue	E	2.2	2.9	4.3	6.4	9.6	10.2	11.2	12.2	13.8	15.4	16.5	18.0
	F	2.6	3.0	4.1	6.4	9.7	10.3	11.2	12.2	14.0	15.5	16.5	18.1
	Mean	**2.4**	**2.9**	**4.2**	**6.4**	**9.7**	**10.2**	**11.2**	**12.2**	**13.9**	**15.5**	**16.5**	**18.1**
Total % recovery	E	102.6	98.8	99.4	99.8	97.9	98.6	98.4	98.6	99.1	103.6	98.8	98.6
	F	104.1	101.8	99.5	100.8	99.1	97.5	99.5	98.6	99.3	101.2	99.4	98.7
	Mean	**103.4**	**100.3**	**99.4**	**100.3**	**98.5**	**98.1**	**99.0**	**98.6**	**99.2**	**102.4**	**99.1**	**98.7**

n.d.: not detected, n.a.: not analyzed, DAT: day after treatment
See Table 7 for material balance.

Appendix 7: Biotransformation of Clothianidin, Expressed as Percentage of Applied Radioactivity, in Soil Viloby d'Onyar (Replicate G and H)

	Replicate	DAT											
Compound	**No.**	**0**	**1**	**3**	**9**	**21**	**28**	**35**	**49**	**63**	**77**	**98**	**120**
Clothianidin	G	98.3	94.1	90.8	81.0	71.3	70.5	67.2	64.3	61.4	60.2	53.4	51.1
	H	100.0	92.8	87.8	82.4	71.0	70.2	68.6	63.6	61.6	58.8	53.6	52.1
	Mean	**99.1**	**93.5**	**89.3**	**81.7**	**71.1**	**70.3**	**67.9**	**64.0**	**61.5**	**59.5**	**53.5**	**51.6**
TZNG	G	n.d.	n.d.	n.d.	0.2	0.6	1.0	1.2	1.5	1.6	1.9	1.7	1.7
	H	n.d.	0.1	<LOQ	0.2	0.7	0.8	1.0	1.9	1.4	1.9	1.6	2.1
	Mean		**0.0**		**0.2**	**0.7**	**0.9**	**1.1**	**1.7**	**1.5**	**1.9**	**1.7**	**1.9**
MNG	G	n.d.	0.3	0.6	1.6	2.5	2.9	4.0	3.8	2.6	4.3	4.6	3.9
	H	n.d.	0.1	0.6	1.5	2.3	3.0	3.3	4.1	3.2	4.3	4.4	3.7
	Mean		**0.2**	**0.6**	**1.5**	**2.4**	**3.0**	**3.6**	**4.0**	**2.9**	**4.3**	**4.5**	**3.8**
TZMU	G	n.d.	0.7	2.1	4.7	2.6	2.5	1.9	2.1	2.0	1.4	1.2	1.2
	H	0.2	0.7	3.0	4.4	3.1	2.3	2.3	1.9	1.4	1.5	1.1	0.9
	Mean	**0.1**	**0.7**	**2.5**	**4.5**	**2.9**	**2.4**	**2.1**	**2.0**	**1.7**	**1.5**	**1.2**	**1.1**
TMG	G	n.d.	n.d.	n.d.	0.2	0.2	0.3	0.3	0.3	0.3	<LOQ	0.1	0.2
	H	n.d.	n.d.	0.1	n.d.	0.2	0.1	0.3	0.4	0.4	n.d.	0.1	0.1
	Mean			**0.1**	**0.1**	**0.2**	**0.2**	**0.3**	**0.3**	**0.4**		**0.1**	**0.2**
NTG	G	n.d.	n.d.	n.d.	n.d.	0.1	0.4	1.0	1.4	3.0	2.0	2.8	3.5
	H	n.d.	n.d.	0.1	0.1	n.d.	0.3	0.7	1.2	3.3	2.2	2.9	3.4
	Mean			**0.1**	**0.0**	**0.1**	**0.4**	**0.9**	**1.3**	**3.2**	**2.1**	**2.8**	**3.5**
TZFA	G	n.d.	n.d.	0.2	1.3	0.9	1.1	1.0	1.1	1.3	0.7	1.3	0.8
	H	n.d.	n.d.	0.3	0.9	1.1	1.3	1.0	1.5	1.0	0.7	1.1	1.0
	Mean			**0.3**	**1.1**	**1.0**	**1.2**	**1.0**	**1.3**	**1.2**	**0.7**	**1.2**	**0.9**
Unidentified radioactivity	G	1.4	2.0	1.1	1.4	0.9	1.6	0.6	0.4	0.4	1.1	0.6	0.5
	H	1.8	2.2	1.9	1.6	0.7	1.4	0.6	0.6	0.8	1.8	0.3	0.9
	Mean	**1.6**	**2.1**	**1.5**	**1.5**	**0.8**	**1.5**	**0.6**	**0.5**	**0.6**	**1.5**	**0.4**	**0.7**
Total extractable residues	G	99.6	97.1	94.8	90.4	79.1	80.3	77.2	74.9	72.6	71.6	65.6	63.0
	H	102.1	96.0	93.8	90.9	79.1	79.4	77.8	75.3	73.2	71.1	65.2	64.2
	Mean	**100.9**	**96.5**	**94.3**	**90.7**	**79.1**	**79.8**	**77.5**	**75.1**	**72.9**	**71.4**	**65.4**	**63.6**
$^{14}CO_2$	G	n.a.	0.2	0.3	1.7	5.0	6.6	7.9	9.7	11.3	12.8	14.7	15.6
	H	n.a.	0.2	0.3	1.7	5.2	6.7	7.9	10.1	11.6	13.0	14.6	16.2
	Mean		**0.2**	**0.3**	**1.7**	**5.1**	**6.6**	**7.9**	**9.9**	**11.4**	**12.9**	**14.7**	**15.9**
Volatile organics	G	n.a.	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1
	H	n.a.	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1
	Mean												
Non-extractable residue	G	2.7	3.3	5.0	9.1	14.0	13.7	14.1	15.5	16.3	17.6	18.8	20.0
	H	1.9	3.1	5.6	8.6	13.5	13.3	14.6	14.8	16.2	17.7	19.4	19.7
	Mean	**2.3**	**3.2**	**5.3**	**8.8**	**13.8**	**13.5**	**14.3**	**15.1**	**16.3**	**17.7**	**19.1**	**19.8**
Total % recovery	G	102.3	100.5	100.2	101.3	98.2	100.7	99.1	100.0	100.2	102.0	99.2	98.6
	H	103.9	99.2	99.8	101.2	97.9	99.3	100.3	100.2	101.0	101.8	99.2	100.1
	Mean	**103.1**	**99.9**	**100.0**	**101.2**	**98.0**	**100.0**	**99.7**	**100.1**	**100.6**	**101.9**	**99.2**	**99.3**

n.d.: not detected, n.a.: not analyzed, DAT: day after treatment
See Table 8 for material balance.

Appendix 8: Example Calculations

- Percent of applied radioactivity: Calculation was based on applied radioactivity which was set as 100%.

  Applied radioactivity = 86167 Bq (3.4.1.1)

- Percent of AR in the soil extracts and PU foam extracts (volatile organics)

$$\text{Portion}\left[\%\ \text{applied}\right] = 100 * \frac{Bq_{aliquot} * \frac{mL_{total}}{mL_{aliquot}}}{Bq_{applied}}$$

  Sample of soil Wellesbourne, DAT-0, desorption solution (Ja57 00 C-DE, $CaCl_2$-solution, Appendix 13):
  100 x 229.52 Bq x 59 mL / 0.5 mL / 86167 Bq = 31.4% (Table 6).

- Amount of non-extractable radioactivity in soil

$$\text{Non extractable residues } [\%\text{ofapplied}] = 100 \text{ x } \frac{Bq_{aliquot} \frac{g_{\text{dry soil total}}}{g_{\text{dry soil aliquot}}} + Bq_{filter}}{Bq_{applied}}$$

  Sample of soil Wellesbourne, DAT-0, bound residue (JR57 00 C, soil not extracted + filter, Appendix 13):

  100 x [(7.65 Bq x 99.6 g/ 1 g)+1013)/ 86167 Bq = 2.1 % (Table 6).

- Recovery

$$\text{Recovery } [\%\text{ of applied}] = \sum\ \%\text{applied}_{\text{soil extractables + bound residues + volatiles}}$$

  for sample of soil Wellesbourne, DAT-0 (Table 6):
  102.4 +2.1% + 0% = 104.5%.

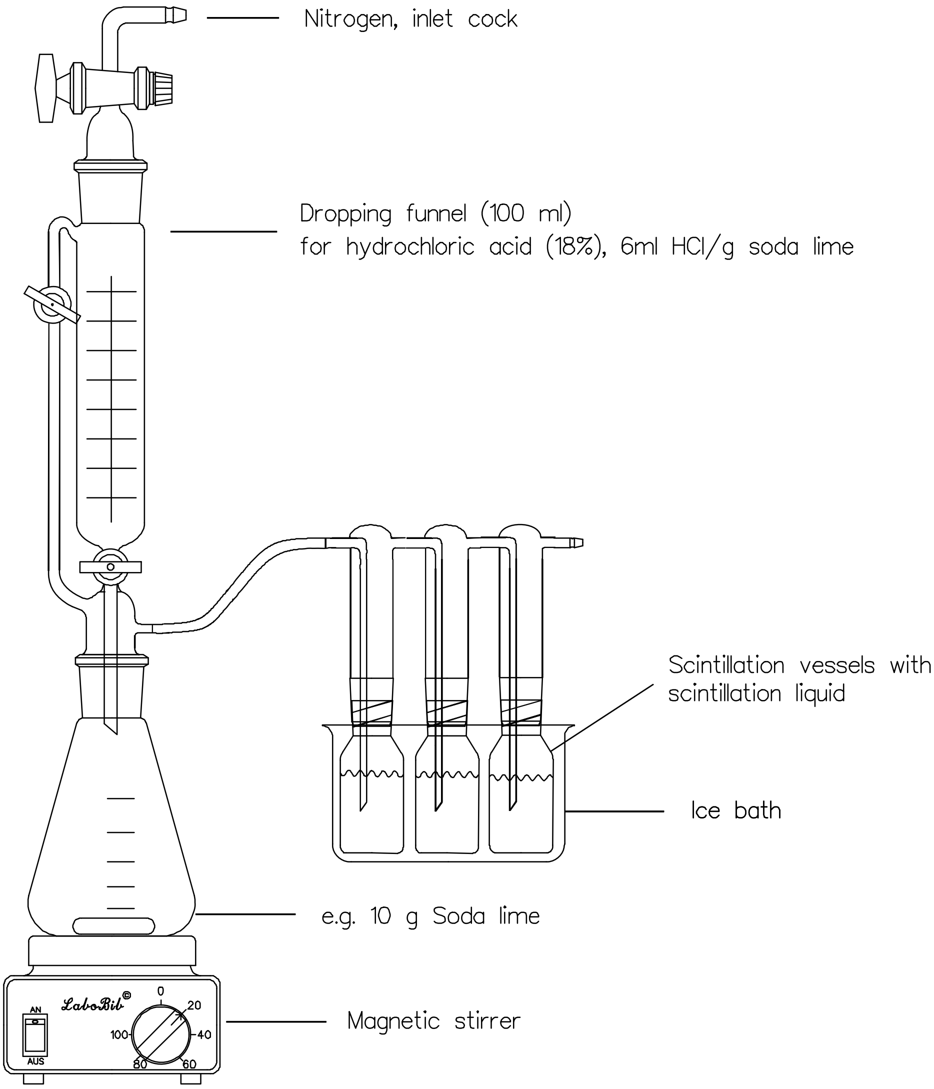
- Concentration of test item / metabolite in a certain phase

$$\text{Test item/metabolite}_{phase}\left[\%\text{applied}\right] = \frac{\left(\%_{\text{peak area}}\right) * \left(\%\text{applied}_{phase}\right)}{100}$$

  for sample of soil Wellesbourne, DAT-1, $CaCl_2$ desorption solution and organic extracts (JR57 00 C-DE, JR57 00 C-SO, JR57 00 C-SA Appendix 14, Table 6):

test item:	97.61% (HPLC) x 31.4% / 100 =	30.65%
	98.28% (HPLC) x 69.9% / 100 =	68.70%
	95.55% (HPLC) x 1.1% / 100 =	<u>1.05%</u>
	total test item:	100.4%

Appendix 9: Apparatus for Liberation of $^{14}CO_2$ Absorbed by Soda Lime



Appendix 10: Time of storage between first and second HPLC-analysis

Sample		1st Analysis (Preliminary Method)		2nd analysis (Evaluation Method)	
Code	Date	Date*	Days	Date*	Days
Ja57 00	2009-09-28	2009-10-01	4		
Ja57 01	2009-09-29	2009-10-02	4	2009-12-19	82
Ja57 03	2009-10-01	2009-10-06	6	2009-12-21	82
Ja57 09	2009-10-07	2009-10-10	4	2010-01-05	91
Ja57 21	2009-10-19	2009-10-22	4	2010-01-07	81
Ja57 28	2009-10-26	2009-10-29	4	2010-01-09	76
Ja57 35**	2009-11-02	2009-11-05	4	2010-01-10	70
Ja57 49	2009-11-16	2009-11-19	4	2010-01-11	57
Ja57 63	2009-11-30	2009-12-02	3	2010-01-12	44
Ja57 77	2009-12-14	2009-12-18	5		
Ja57 98	2010-01-04	2010-01-06	3		
Ja57 120	2010-01-26	2010-01-29	4		

* last chromatogram of sample series (24 vials)

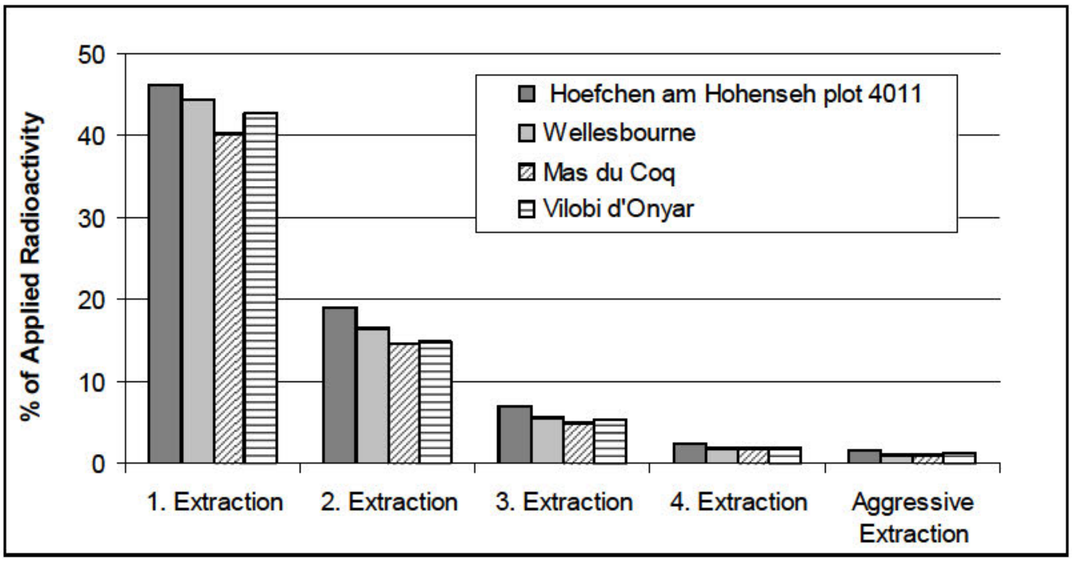
** additional check of LSC and concentration recovery

Appendix 11: Distribution of Radioactivity in individual ambient organic extracts sampled on DAT-0

Sample	Volumes LSC Aliquot [mL]	Total [mL]	Radioactivity LSC Aliquot [Bq]	Total [Bq]	[%] of AR
Soil Hoefchen plot 4011, Ja57 00 A					
1. Extraction	0.5	77	259.35	39940	46.4
2. Extraction	0.5	79	103.29	16320	18.9
3. Extraction	0.5	78	38.43	5995	7.0
4. Extraction	0.5	77	14.24	2193	2.5
			Sum	64448	74.8
Soil Wellesbourne, Ja57 00 C					
1. Extraction	0.5	78	245.4	38282	44.4
2. Extraction	0.5	79	90.23	14256	16.5
3. Extraction	0.5	78	30.4	4742	5.5
4. Extraction	0.5	78	10.25	1599	1.9
			Sum	58880	68.3
Soil Mas du Coq, Ja57 00 E					
1. Extraction	0.5	77	224.88	34632	40.2
2. Extraction	0.5	79	80.55	12727	14.8
3. Extraction	0.5	78	27.65	4313	5.0
4. Extraction	0.5	77	10.02	1543	1.8
			Sum	53215	61.8
Soil Vilobi d'Onyar, Ja57 00 G					
1. Extraction	0.5	77	239.72	36917	42.8
2. Extraction	0.5	78	82.7	12901	15.0
3. Extraction	0.5	79	28.8	4550	5.3
4. Extraction	0.5	78	10.67	1665	1.9
			Sum	56033	65.0

Appendix 11: Distribution of Radioactivity in individual ambient organic extracts sampled on DAT-0 (continued)

Graphical presentation including the amount of radioactivity extracted with the aggressive extraction



Appendix 12: Identification of $^{14}CO_2$ by Barium Carbonate Precipitation

Sample	**Ja57 12 A**	**Ja 57 12 C**	**Ja57 12 E**	**Ja57 12 G**
Radioactivity before liberation for identification procedure	12075 Bq	13684 Bq	7759 Bq	13239 Bq
Trapped carbon dioxide in sodium hydroxide solution after liberation	10488.59 Bq	12296.305 Bq	7791 Bq	11602.75 Bq
Recovery of carbon dioxide after liberation with acetic acid	86.9%	89.9%	100.4%	87.6%
Radioactivity used for barium carbonate precipitation	9191.79 Bq (38 mL of sodium hydroxide solution)	10910.305 Bq (38 mL of sodium hydroxide solution)	7558.2 Bq (38 mL of sodium hydroxide solution)	10266.8 Bq (38 mL of sodium hydroxide solution)
Radioactivity of supernatant after barium carbonate precipitation	42.56 Bq (0.4%) (indicating that 99.6% of the radioactivity were precipitated as $BaCO_3$)	8.36 Bq (0.1%) (indicating that 99.9% of the radioactivity were precipitated as $BaCO_3$)	15.2 Bq (0.2%) (indicating that 99.8% of the radioactivity were precipitated as $BaCO_3$)	12.16 Bq (0.1%) (indicating that 99.9% of the radioactivity were precipitated as $BaCO_3$)

Appendix 13: Raw Data of Distribution of Radioactivity in Extracts

Soil Hoefchen am Hohenseh plot 4011

DAT	Sample ID	Calcium chloride solution				Ambient organic extract				Aggressive Organic Extract			
		V_T	V_A	LS	Subtotal	V_T	V_A	LS	Subtotal	V_T	V_A	LS	Subtotal
[days]		[mL]	[mL]	[Bq]	[Bq]	[mL]	[mL]	[Bq]	[Bq]	[mL]	[mL]	[Bq]	[Bq]
0	Ja57 00 A	52	0.5	188.12	**19564**	300	0.5	105.77	**63462**	76	0.5	9.58	**1456**
	Ja57 00 B	54	0.5	187.29	**20227**	305	0.5	105.58	**64404**	75	0.5	9.77	**1466**
1	Ja57 01 A	53	0.5	175.87	**18642**	295	0.5	99.47	**58687**	69	0.5	12.82	**1769**
	Ja57 01 B	53	0.5	173.54	**18395**	295	0.5	100.37	**59218**	68	0.5	13.59	**1848**
3	Ja57 03 A	56	0.5	161.86	**18128**	305	0.5	92.47	**56407**	74	0.5	21.65	**3204**
	Ja57 03 B	52	0.5	159.97	**16637**	305	0.5	92.27	**56285**	75	0.5	20.61	**3092**
9	Ja57 09 A	53	0.5	109.11	**11566**	305	0.5	74.62	**45518**	74	0.5	24.99	**3699**
	Ja57 09 B	53	0.5	111.38	**11806**	305	0.5	76.70	**46787**	74	0.5	24.69	**3654**
21	Ja57 21 A	52	0.5	71.02	**7386**	295	0.5	59.35	**35017**	71	0.5	34.00	**4828**
	Ja57 21 B	53	0.5	74.67	**7915**	295	0.5	59.67	**35205**	72	0.5	33.59	**4837**
28	Ja57 28 A	53	0.5	64.71	**6859**	310	0.5	56.42	**34980**	72	0.5	33.47	**4820**
	Ja57 28 B	54	0.5	62.19	**6717**	310	0.5	55.38	**34336**	72	0.5	34.37	**4949**
35	Ja57 35 A	53	0.5	58.35	**6185**	300	0.5	52.07	**31242**	72	0.5	32.41	**4667**
	Ja57 35 B	53	0.5	60.21	**6382**	305	0.5	53.71	**32763**	73	0.5	31.95	**4665**
49	Ja57 49 A	52	0.5	52.04	**5412**	300	0.5	46.70	**28020**	74	0.5	34.23	**5066**
	Ja57 49 B	53	0.5	51.55	**5464**	300	0.5	47.92	**28752**	73	0.5	35.15	**5132**
63	Ja57 63 A	53	0.5	47.53	**5038**	300	0.5	43.50	**26100**	72	0.5	38.96	**5610**
	Ja57 63 B	54	0.5	48.73	**5263**	300	0.5	45.18	**27108**	75	0.5	38.18	**5727**
77	Ja57 77 A	54	0.5	46.76	**5050**	295	0.5	42.79	**25246**	73	0.5	40.33	**5888**
	Ja57 77 B	54	0.5	45.88	**4955**	300	0.5	44.03	**26418**	71	0.5	42.45	**6028**
98	Ja57 98 A	53	0.5	43.47	**4608**	305	0.5	37.71	**23003**	72	0.5	38.06	**5481**
	Ja57 98 B	54	0.5	40.62	**4387**	305	0.5	37.97	**23162**	73	0.5	38.50	**5621**
120	Ja57 12 A	51	0.5	41.49	**4232**	305	0.5	35.68	**21765**	75	0.5	39.02	**5853**
	Ja57 12 B	53	0.5	41.27	**4375**	305	0.5	36.78	**22436**	74	0.5	42.32	**6263**

DAT	Sample ID	Soil not extracted				Volatiles				
		Soil			Filter	Soda lime	PU-foam			
		M_T	LS	Subtotal	Subtotal	Subtotal	V_T	V_A	LS	Subtotal
[days]		[g]	[Bq/g]	[Bq]	[Bq]	[Bq]	[mL]	[mL]	[Bq]	[Bq]
0	Ja57 00 A	98.1	14.68	**1440**	**1047**					
	Ja57 00 B	97.7	14.74	**1440**	**1177**					
1	Ja57 01 A	100.0	38.53	**3853**	**1601**	**207**	50	5	0.20	**2**
	Ja57 01 B	98.7	36.17	**3570**	**1413**	**219**	50	5	0.15	**2**
3	Ja57 03 A	98.7	66.12	**6526**	**1306**	**704**	50	5	0.44	**4**
	Ja57 03 B	98.4	76.24	**7502**	**1233**	**742**	50	5	0.18	**2**
9	Ja57 09 A	99.7	184.49	**18394**	**1053**	**3675**	50	5	0.17	**2**
	Ja57 09 B	99.5	181.81	**18090**	**1165**	**3845**	50	5	0.24	**2**
21	Ja57 21 A	101.1	245.91	**24862**	**1382**	**11012**	50	5	0.19	**2**
	Ja57 21 B	100.8	234.95	**23683**	**1096**	**11610**	50	5	0.14	**1**
28	Ja57 28 A	101.7	239.58	**24365**	**1066**	**13001**	50	5	0.58	**6**
	Ja57 28 B	101.6	242.40	**24628**	**985**	**13691**	50	5	0.33	**3**
35	Ja57 35 A	103.1	241.92	**24942**	**1088**	**15907**	50	5	0.21	**2**
	Ja57 35 B	102.6	237.23	**24340**	**1034**	**15713**	50	5	0.41	**4**
49	Ja57 49 A	100.4	249.88	**25088**	**771**	**18638**	50	5	0.31	**3**
	Ja57 49 B	100.4	249.94	**25094**	**749**	**18394**	50	5	0.13	**1**
63	Ja57 63 A	100.5	256.53	**25781**	**873**	**20320**	50	5	0.70	**7**
	Ja57 63 B	100.2	253.64	**25415**	**1068**	**20468**	50	5	0.33	**3**
77	Ja57 77 A	100.1	260.03	**26029**	**838**	**21897**	50	5	0.51	**5**
	Ja57 77 B	100.1	265.90	**26617**	**876**	**22128**	50	5	0.34	**3**
98	Ja57 98 A	99.1	258.26	**25594**	**665**	**24309**	50	5	0.21	**2**
	Ja57 98 B	99.6	261.40	**26035**	**673**	**24218**	50	5	0.42	**4**
120	Ja57 12 A	99.9	258.95	**25869**	**710**	**25302**	50	5	29.71	**297**
	Ja57 12 B	100.1	256.30	**25656**	**807**	**25164**	50	5	0.64	**6**

V_T = Total volume
V_A = Volume of aliquot
LS = Liquid scintillation counting
M_T = Total soil weight before exhaustive extraction (dry weight)
DAT= Day after treatment

Appendix 13: Raw Data of Distribution of Radioactivity in Extracts (continued)

Soil Wellesbourne

DAT	Sample ID	Calcium chloride solution				Ambient organic extract				Agressive Organic extract			
		V_T	V_A	LS	Subtotal	V_T	V_A	LS	Subtotal	V_T	V_A	LS	Subtotal
[days]		[mL]	[mL]	[Bq]	[Bq]	[mL]	[mL]	[Bq]	[Bq]	[mL]	[mL]	[Bq]	[Bq]
0	Ja57 00 C	59	0.5	229.52	**27083**	305	0.5	98.73	**60225**	75	0.5	6.40	**960**
	Ja57 00 D	61	0.5	232.08	**28314**	310	0.5	94.19	**58398**	74	0.5	5.84	**864**
1	Ja57 01 C	61	0.5	215.59	**26302**	300	0.5	90.55	**54330**	55	0.5	11.85	**1304**
	Ja57 01 D	62	0.5	213.67	**26495**	300	0.5	89.12	**53472**	71	0.5	10.40	**1477**
3	Ja57 03 C	69	0.5	189.95	**26213**	310	0.5	83.77	**51937**	74	0.5	15.86	**2347**
	Ja57 03 D	61	0.5	211.33	**25782**	310	0.5	87.35	**54157**	75	0.5	16.25	**2438**
9	Ja57 09 C	61	0.5	168.24	**20525**	310	0.5	83.28	**51634**	80	0.5	20.15	**3224**
	Ja57 09 D	61	0.5	171.30	**20899**	310	0.5	83.20	**51584**	68	0.5	19.65	**2672**
21	Ja57 21 C	60	0.5	144.77	**17372**	300	0.5	77.93	**46758**	73	0.5	29.59	**4320**
	Ja57 21 D	61	0.5	144.65	**17647**	300	0.5	77.25	**46350**	76	0.5	29.10	**4423**
28	Ja57 28 C	61	0.5	130.87	**15966**	310	0.5	74.34	**46091**	68	0.5	34.67	**4715**
	Ja57 28 D	61	0.5	129.88	**15845**	310	0.5	73.16	**45359**	55	0.5	35.78	**3936**
35	Ja57 35 C	60	0.5	125.61	**15073**	305	0.5	74.33	**45341**	69	0.5	33.25	**4589**
	Ja57 35 D	60	0.5	127.68	**15322**	305	0.5	74.19	**45256**	68	0.5	32.76	**4455**
49	Ja57 49 C	61	0.5	114.73	**13997**	305	0.5	69.19	**42206**	69	0.5	38.14	**5263**
	Ja57 49 D	61	0.5	115.10	**14042**	320	0.5	67.87	**43437**	74	0.5	36.51	**5403**
63	Ja57 63 C	62	0.5	105.86	**13127**	305	0.5	64.92	**39601**	74	0.5	39.07	**5782**
	Ja57 63 D	64	0.5	108.23	**13853**	305	0.5	66.70	**40687**	71	0.5	40.39	**5735**
77	Ja57 77 C	63	0.5	103.99	**13103**	300	0.5	66.14	**39684**	71	0.5	42.63	**6053**
	Ja57 77 D	64	0.5	104.69	**13400**	300	0.5	65.41	**39246**	74	0.5	42.81	**6336**
98	Ja57 98 C	60	0.5	94.92	**11390**	310	0.5	59.73	**37033**	71	0.5	44.59	**6332**
	Ja57 98 D	60	0.5	96.24	**11549**	305	0.5	60.40	**36844**	77	0.5	44.19	**6805**
120	Ja57 12 C	60	0.5	91.06	**10927**	310	0.5	56.49	**35024**	79	0.5	45.99	**7266**
	Ja57 12 D	59	0.5	90.93	**10730**	305	0.5	57.34	**34977**	81	0.5	46.93	**7603**

DAT	Sample ID	Soil not extracted				Volatiles				
		Soil			Filter	Soda lime	PU-foam			
		M_T	LS	Subtotal	Subtotal	Subtotal	V_T	V_A	LS	Subtotal
[days]		[g]	[Bq/g]	[Bq]	[Bq]	[Bq]	[mL]	[mL]	[Bq]	[Bq]
0	Ja57 00 C	99.6	7.65	**762**	**1013**					
	Ja57 00 D	99 2	7.32	**726**	**1287**					
1	Ja57 01 C	99.4	14.29	**1420**	**1714**	**138**	50	5	0.18	**2**
	Ja57 01 D	100.3	13.61	**1365**	**1507**	**135**	50	5	0.16	**2**
3	Ja57 03 C	97.6	29.46	**2875**	**1066**	**298**	50	5	0.31	**3**
	Ja57 03 D	98.6	28.74	**2834**	**1376**	**305**	50	5	0.20	**2**
9	Ja57 09 C	99.4	81.51	**8102**	**1203**	**1730**	50	5	0.25	**3**
	Ja57 09 D	99.8	78.48	**7832**	**1546**	**1687**	50	5	0.19	**2**
21	Ja57 21 C	101.1	105.29	**10645**	**1772**	**4986**	50	5	0.27	**3**
	Ja57 21 D	100.8	109.31	**11018**	**1739**	**5211**	50	5	0.20	**2**
28	Ja57 28 C	100.5	113.26	**11383**	**1631**	**6311**	50	5	0.13	**1**
	Ja57 28 D	100.6	113.58	**11426**	**2325**	**6212**	50	5	0.52	**5**
35	Ja57 35 C	100.7	122.59	**12345**	**1324**	**7108**	50	5	0.48	**5**
	Ja57 35 D	100.7	122.61	**12347**	**1581**	**7099**	50	5	0.17	**2**
49	Ja57 49 C	100.0	133.47	**13347**	**1505**	**8965**	50	5	0.11	**1**
	Ja57 49 D	100.3	135.27	**13568**	**953**	**9360**	50	5	0.27	**3**
63	Ja57 63 C	100.1	148.12	**14827**	**1398**	**10523**	50	5	0.34	**3**
	Ja57 63 D	100.1	147.78	**14793**	**1397**	**10598**	50	5	0.25	**3**
77	Ja57 77 C	99.8	155.46	**15515**	**1302**	**11412**	50	5	0.23	**2**
	Ja57 77 D	99.8	155.67	**15536**	**1072**	**11106**	50	5	0.22	**2**
98	Ja57 98 C	99 0	162.94	**16131**	**1090**	**12934**	50	5	2.13	**21**
	Ja57 98 D	99.6	161.34	**16069**	**1025**	**12535**	50	5	0.34	**3**
120	Ja57 12 C	99.8	162.47	**16215**	**1241**	**13844**	50	5	0.53	**5**
	Ja57 12 D	99.6	163.06	**16241**	**1045**	**14153**	50	5	0.51	**5**

V_T = Total volume
V_A = Volume of aliquot
LS = Liquid scintillation counting
M_T = Total soil weight before exhaustive extraction (dry weight)
DAT= Day after treatment

Appendix 13: Raw Data of Distribution of Radioactivity in Extracts (continued)

Soil Mas du Coq

DAT	Sample ID	Calcium chloride solution				Ambient organic extract				Aggressive organic extract			
		V_T	V_A	LS	Subtotal	V_T	V_A	LS	Subtotal	V_T	V_A	LS	Subtotal
[days]		[mL]	[mL]	[Bq]	[Bq]	[mL]	[mL]	[Bq]	[Bq]	[mL]	[mL]	[Bq]	[Bq]
0	Ja57 00 E	58	0.5	274.82	**31879**	305	0.5	88.13	**53759**	74	0.5	6.29	**931**
	Ja57 00 F	59	0.5	271.29	**32012**	305	0.5	89.36	**54510**	72	0.5	6.62	**953**
1	Ja57 01 E	58	0.5	255.83	**29676**	300	0.5	86.06	**51636**	64	0.5	10.19	**1304**
	Ja57 01 F	58	0.5	264.48	**30680**	300	0.5	88.40	**53040**	68	0.5	9.70	**1319**
3	Ja57 03 E	58	0.5	235.12	**27274**	305	0.5	85.37	**52076**	72	0.5	16.61	**2392**
	Ja57 03 F	57	0.5	236.56	**26967**	305	0.5	86.47	**52747**	72	0.5	16.27	**2343**
9	Ja57 09 E	59	0.5	210.53	**24843**	305	0.5	85.14	**51935**	72	0.5	21.55	**3103**
	Ja57 09 F	60	0.5	211.82	**25418**	305	0.5	85.45	**52125**	73	0.5	21.88	**3194**
21	Ja57 21 E	58	0.5	186.37	**21619**	295	0.5	82.31	**48563**	71	0.5	29.38	**4172**
	Ja57 21 F	58	0.5	185.41	**21508**	295	0.5	83.67	**49365**	71	0.5	30.56	**4340**
28	Ja57 28 E	58	0.5	172.28	**19984**	305	0.5	79.96	**48776**	69	0.5	34.30	**4733**
	Ja57 28 F	58	0.5	172.41	**20000**	305	0.5	78.99	**48184**	70	0.5	32.34	**4528**
35	Ja57 35 E	57	0.5	169.45	**19317**	300	0.5	80.16	**48096**	72	0.5	32.87	**4733**
	Ja57 35 F	58	0.5	171.68	**19915**	305	0.5	79.67	**48599**	72	0.5	31.38	**4519**
49	Ja57 49 E	60	0.5	154.96	**18595**	305	0.5	76.91	**46915**	70	0.5	34.74	**4864**
	Ja57 49 F	60	0.5	155.61	**18673**	305	0.5	76.48	**46653**	69	0.5	35.15	**4851**
63	Ja57 63 E	56	0.5	150.92	**16903**	305	0.5	74.67	**45549**	72	0.5	42.39	**6104**
	Ja57 63 F	56	0.5	152.57	**17088**	305	0.5	74.11	**45207**	74	0.5	41.51	**6143**
77	Ja57 77 E	59	0.5	149.84	**17681**	300	0.5	75.62	**45372**	73	0.5	48.03	**7012**
	Ja57 77 F	59	0.5	144.30	**17027**	300	0.5	74.27	**44562**	72	0.5	45.01	**6481**
98	Ja57 98 E	57	0.5	129.84	**14802**	310	0.5	67.92	**42110**	72	0.5	47.46	**6834**
	Ja57 98 F	59	0.5	131.73	**15544**	310	0.5	68.06	**42197**	72	0.5	44.64	**6428**
120	Ja57 12 E	56	0.5	127.22	**14249**	305	0.5	66.91	**40815**	71	0.5	45.97	**6528**
	Ja57 12 F	57	0.5	126.76	**14451**	305	0.5	65.96	**40236**	72	0.5	45.07	**6490**

DAT	Sample ID	Soil not extracted				Volatiles				
		Soil			Filter	Soda lime	PU-foam			
		M_T	LS	Subtotal	Subtotal	Subtotal	V_T	V_A	LS	Subtotal
[days]		[g]	[Bq/g]	[Bq]	[Bq]	[Bq]	[mL]	[mL]	[Bq]	[Bq]
0	Ja57 00 E	98.5	6.87	**677**	**1203**					
	Ja57 00 F	98.1	6.98	**685**	**1560**					
1	Ja57 01 E	98.3	13.21	**1299**	**1170**	**47**	50	5	0.23	**2**
	Ja57 01 F	99.7	12.24	**1220**	**1374**	**49**	50	5	0.19	**2**
3	Ja57 03 E	98.0	21.74	**2131**	**1596**	**149**	50	5	0.69	**7**
	Ja57 03 F	98.6	21.72	**2142**	**1390**	**158**	50	5	0.27	**3**
9	Ja57 09 E	98.8	40.76	**4027**	**1445**	**622**	50	5	0.31	**3**
	Ja57 09 F	98.1	40.12	**3936**	**1570**	**595**	50	5	0.39	**4**
21	Ja57 21 E	100.4	59.26	**5950**	**2352**	**1700**	50	5	0.34	**3**
	Ja57 21 F	100.4	59.46	**5970**	**2392**	**1831**	50	5	0.13	**1**
28	Ja57 28 E	100.1	71.70	**7177**	**1600**	**2716**	50	5	0.14	**1**
	Ja57 28 F	100.8	71.32	**7189**	**1678**	**2490**	50	5	0.44	**4**
35	Ja57 35 E	100.2	81.71	**8187**	**1436**	**3043**	50	5	0.15	**2**
	Ja57 35 F	100.6	78.92	**7939**	**1730**	**3020**	50	5	0.29	**3**
49	Ja57 49 E	100.2	91.53	**9171**	**1298**	**4110**	50	5	0.23	**2**
	Ja57 49 F	100.3	92.37	**9265**	**1289**	**4191**	50	5	0.65	**7**
63	Ja57 63 E	99.7	101.86	**10155**	**1722**	**4968**	50	5	0.26	**3**
	Ja57 63 F	100.7	102.32	**10304**	**1797**	**5016**	50	5	0.32	**3**
77	Ja57 77 E	99.7	118.93	**11857**	**1451**	**5907**	50	5	0.47	**5**
	Ja57 77 F	100.0	118.12	**11812**	**1556**	**5841**	50	5	0.14	**1**
98	Ja57 98 E	99.5	132.74	**13208**	**1046**	**7101**	50	5	0.29	**3**
	Ja57 98 F	99.5	131.93	**13127**	**1081**	**7253**	50	5	0.22	**2**
120	Ja57 12 E	99.8	139.39	**13911**	**1616**	**7875**	50	5	0.52	**5**
	Ja57 12 F	99.6	141.63	**14106**	**1527**	**8273**	50	5	0.71	**7**

V_T = Total volume
V_A = Volume of aliquot
LS = Liquid scintillation counting
M_T = Total soil weight before exhaustive extraction (dry weight)
DAT= Day after treatment

Appendix 13: Raw Data of Distribution of Radioactivity in Extracts (continued)

Soil Vilobi d'Onyar

DAT	Sample ID	Calcium chloride solution				Ambient organic extract				Aggressive organic extract			
		V_T	V_A	LS	Subtotal	V_T	V_A	LS	Subtotal	V_T	V_A	LS	Subtotal
[days]		[mL]	[mL]	[Bq]	[Bq]	[mL]	[mL]	[Bq]	[Bq]	[mL]	[mL]	[Bq]	[Bq]
0	Ja57 00 G	61	0.5	241.24	**29431**	300	0.5	92.64	**55584**	56	0.5	7.56	**847**
	Ja57 00 H	60	0.5	247.09	**29651**	305	0.5	93.62	**57108**	62	0.5	9.52	**1180**
1	Ja57 01 G	60	0.5	230.42	**27650**	295	0.5	92.42	**54528**	62	0.5	11.92	**1478**
	Ja57 01 H	60	0.5	229.74	**27569**	295	0.5	90.88	**53619**	61	0.5	12.35	**1507**
3	Ja57 03 G	60	0.5	219.46	**26335**	305	0.5	86.94	**53033**	54	0.5	21.76	**2350**
	Ja57 03 H	59	0.5	214.18	**25273**	305	0.5	87.64	**53460**	54	0.5	19.77	**2135**
9	Ja57 09 G	60	0.5	182.12	**21854**	300	0.5	87.42	**52452**	67	0.5	26.84	**3597**
	Ja57 09 H	61	0.5	180.94	**22075**	300	0.5	87.60	**52560**	70	0.5	26.67	**3734**
21	Ja57 21 G	58	0.5	152.36	**17674**	285	0.5	81.50	**46455**	55	0.5	36.94	**4063**
	Ja57 21 H	59	0.5	148.48	**17521**	290	0.5	79.53	**46127**	64	0.5	35.38	**4529**
28	Ja57 28 G	60	0.5	141.04	**16925**	305	0.5	76.85	**46879**	70	0.5	38.71	**5419**
	Ja57 28 H	60	0.5	136.36	**16363**	305	0.5	76.36	**46580**	72	0.5	37.74	**5435**
35	Ja57 35 G	59	0.5	133.28	**15727**	300	0.5	74.97	**44982**	75	0.5	38.53	**5780**
	Ja57 35 H	58	0.5	137.11	**15905**	300	0.5	75.32	**45192**	76	0.5	39.20	**5958**
49	Ja57 49 G	60	0.5	122.48	**14698**	305	0.5	72.33	**44121**	65	0.5	43.70	**5681**
	Ja57 49 H	61	0.5	124.76	**15221**	305	0.5	72.67	**44329**	69	0.5	38.35	**5292**
63	Ja57 63 G	58	0.5	113.07	**13116**	305	0.5	68.10	**41541**	83	0.5	47.46	**7878**
	Ja57 63 H	58	0.5	120.32	**13957**	305	0.5	68.64	**41870**	79	0.5	46.06	**7277**
77	Ja57 77 G	62	0.5	112.69	**13974**	295	0.5	67.79	**39996**	74	0.5	52.60	**7785**
	Ja57 77 H	62	0.5	110.36	**13685**	295	0.5	67.37	**39748**	76	0.5	51.79	**7872**
98	Ja57 98 G	59	0.5	104.71	**12356**	305	0.5	59.29	**36167**	76	0.5	52.79	**8024**
	Ja57 98 H	60	0.5	102.67	**12320**	310	0.5	59.03	**36599**	67	0.5	54.02	**7239**
120	Ja57 12 G	57	0.5	95.69	**10909**	305	0.5	58.08	**35429**	74	0.5	53.73	**7952**
	Ja57 12 H	59	0.5	99.51	**11742**	305	0.5	58.51	**35691**	78	0.5	50.68	**7906**

DAT	Sample ID	Soil not extracted				Volatiles				
		Soil			Filter	Soda lime	PU-foam			
		M_T	LS	Subtotal	Subtotal	Subtotal	V_T	V_A	LS	Subtotal
[days]		[g]	[Bq/g]	[Bq]	[Bq]	[Bq]	[mL]	[mL]	[Bq]	[Bq]
0	Ja57 00 G	98.2	6.70	**658**	**1656**					
	Ja57 00 H	98.5	7 59	**748**	**876**					
1	Ja57 01 G	99.6	12.19	**1214**	**1628**	**141**	50	5	0.15	**2**
	Ja57 01 H	98.4	13 31	**1310**	**1363**	**142**	50	5	0.25	**3**
3	Ja57 03 G	98.1	26.43	**2593**	**1754**	**291**	50	5	0.26	**3**
	Ja57 03 H	99.3	32.18	**3195**	**1662**	**299**	50	5	0.15	**2**
9	Ja57 09 G	98.5	60.75	**5984**	**1876**	**1486**	50	5	0.26	**3**
	Ja57 09 H	97.5	58.47	**5701**	**1690**	**1466**	50	5	0.24	**2**
21	Ja57 21 G	100.6	86.65	**8717**	**3361**	**4349**	50	5	0.26	**3**
	Ja57 21 H	100.3	88.42	**8869**	**2777**	**4499**	50	5	0.14	**1**
28	Ja57 28 G	99.9	99.48	**9938**	**1859**	**5715**	50	5	0.28	**3**
	Ja57 28 H	100.1	97 94	**9804**	**1663**	**5738**	50	5	0.28	**3**
35	Ja57 35 G	100.0	103.69	**10369**	**1779**	**6793**	50	5	0.33	**3**
	Ja57 35 H	100.8	109.10	**10997**	**1550**	**6826**	50	5	0.16	**2**
49	Ja57 49 G	99.5	115.23	**11465**	**1861**	**8355**	50	5	0.20	**2**
	Ja57 49 H	100.0	115.24	**11524**	**1244**	**8705**	50	5	0.11	**1**
63	Ja57 63 G	100.2	121.71	**12195**	**1863**	**9745**	50	5	0.22	**2**
	Ja57 63 H	99.8	123.00	**12275**	**1700**	**9962**	50	5	0.24	**2**
77	Ja57 77 G	99.9	132.75	**13262**	**1896**	**11014**	50	5	0.18	**2**
	Ja57 77 H	99.6	136.84	**13629**	**1644**	**11178**	50	5	0.48	**5**
98	Ja57 98 G	99.2	148.82	**14763**	**1470**	**12682**	50	5	0.21	**2**
	Ja57 98 H	99.0	153.10	**15157**	**1600**	**12575**	50	5	0.18	**2**
120	Ja57 12 G	99.7	157.10	**15663**	**1577**	**13442**	50	5	0.52	**5**
	Ja57 12 H	99.7	156.88	**15641**	**1305**	**13926**	50	5	0.77	**8**

V_T = Total volume
V_A = Volume of aliquot
LS = Liquid scintillation counting
M_T = Total soil weight before exhaustive extraction (dry weight)
DAT= Day after treatment

Appendix 14: Raw Data of HPLC Analysis (Evaluation Method)

Soi: Wellesbourne

DAT (Replicate)	Extract	HPLC ID	Chlothia-nidin	TZNG	MNG	TZMU	TMG	NTG	TZU	TZFA	41	31	30	Diffuse radioactivity	Unidentified radioactivity	Total
0 C	$CaCl_2$ solution	Ja5700.003	97.61								0.96			1.45	2.41	100.0
	Ambient org. extract	Ja5700.011	98.28								0.59			1.13	1.72	100.0
	Agressive org. extract	Ja5700.020	95.55											4.45	4.45	100.0
0 D	$CaCl_2$ solution	Ja5700.004	96.33								0.99			2.68	3.67	100.0
	Ambient org. extract	Ja5700.012	98.08								0.64			1.28	1.92	100.0
	Agressive org. extract	Ja5700.021	94.74											5.26	5.26	100.0
1 C	$CaCl_2$ solution	Ja5701.027	94.73		2.11	1 56					0.85	0.66		0.09	1.60	100.0
	Ambient org. extract	Ja5701.035	97.86		0.23	0.72					0.57		0 32	0.30	1.19	100.0
	Agressive org. extract	Ja5701.043	90.33											9.67	9.67	100.0
1 D	$CaCl_2$ solution	Ja5701.028	94.83		0.50	1 94					0.73		0.73	1.27	2.73	100.0
	Ambient org. extract	Ja5701.036	97.83	0.16	0.28	0 39	0.11				0.38		0 24	0.61	1.23	100.0
	Agressive org. extract	Ja5701.044	91.62											8.38	8.38	100.0
3 C	$CaCl_2$ solution	Ja5703.035	92.16		1.49	4.46		0.49		0.53			0 33	0.54	0.87	100.0
	Ambient org. extract	Ja5703.043	94.72		0.62	2 27	0.42			1.02			0 68	0.27	0.95	100.0
	Agressive org. extract	Ja5703.051	96.42											3.58	3.58	100.0
3 D	$CaCl_2$ solution	Ja5703.036	89.81		2.32	4.79				0.38			0 58	2.12	2.70	100.0
	Ambient org. extract	Ja5703.044	95.55	0.12		2.45					0.48	0.48		0.92	1.88	100.0
	Agressive org. extract	Ja5703.053	88.56			9.42								2.02	2.02	100.0
9 C	$CaCl_2$ solution	Ja5709.030	80.50		4.54	11.84		0.49		1.87		0.66		0.10	0.76	100.0
	Ambient org. extract	Ja5709.042	88.74	0.13	1.59	5.11	0 33			2.54		0.50	0 27	0.79	1.56	100.0
	Agressive org. extract	Ja5709.050	85.66	1.11	4.21	7 27								1.75	1.75	100.0
9 D	$CaCl_2$ solution	Ja5709.031	81.24		5.24	10.59				1.04		0.66		1.23	1.89	100.0
	Ambient org. extract	Ja5709.043	90.12	0.42	1.33	4.76				2.53		0.40	0.18	0.26	0.84	100.0
	Agressive org. extract	Ja5709.051	78.70		5.17	7 80				3.35				4.98	4.98	100.0
21 C	$CaCl_2$ solution	Ja5721.029	56.74	0.49	13 35	16.75				8.15				4.52	4.52	100.0
	Ambient org. extract	Ja5721.037	88.20	1.55	2.33	4 30	0 35			2.48			0 54	0.25	0.79	100.0
	Agressive org. extract	Ja5721.045	74.49	1.45	7.73	6.76	0 81	1.56		4.09			1.45	1.66	3.11	100.0
21 D	$CaCl_2$ solution	Ja5721.030	80.62	0.68	9.00	7 38		0.37		0.82				1.13	1.13	100.0
	Ambient org. extract	Ja5721.038	85.61	1.65	2.64	4 28	0.42			3.50		0.21	0.41	1.28	1.90	100.0
	Agressive org. extract	Ja5721.046	66.19	2.51	9.19	8 33		1.15		6.47				6.16	6.16	100.0
28 C	$CaCl_2$ solution	Ja5728.043	79.05	1.08	10.74	6 59	0 38			1.79				0.37	0.37	100.0
	Ambient org. extract	Ja5728.051	86.46	2.33	3.34	3 25	0.42	0.23		3.78				0.19	0.19	100.0
	Agressive org. extract	Ja5728.0 59	76.32	3.59	7.83	6 29	0 61	0.75		1.28				3.33	3.33	100.0
28 D	$CaCl_2$ solution	Ja5728.044	78.55	2.08	10 02	4 92		1.23		1.64				1.56	1.56	100.0
	Ambient org. extract	Ja5728.052	87.62	2.27	3.01	2 84	0 55	0.37		2.82			0 39	0.13	0.52	100.0
	Agressive org. extract	Ja5728.060	82.50	2.45	3.92	4 67	0.43	2.36		1.40			0 61	1.66	2.27	100.0
35 C	$CaCl_2$ solution	Ja5735.029	76.68	1.50	11 63	4.45		1.47		1.41				2.86	2.86	100.0
	Ambient org. extract	Ja5735.037	83.86	3.05	4.19	2 84	0 68	0.55		4.70				0.13	0.13	100.0
	Agressive org. extract	Ja5735.045	75.36	2.38	7.82	5 80		2.05		3.27				3.32	3.32	100.0
35 D	$CaCl_2$ solution	Ja5735.030	78.35	1.15	11 67	5 04		1.38		1.35				1.06	1.06	100.0
	Ambient org. extract	Ja5735.038	83.38	3.21	4.13	2 57	0.48	0.46		4.18				1.59	1.59	100.0
	Agressive org. extract	Ja5735.046	74.11	2.19	9.80	4.77	0 34	2.20		2.38				4.21	4.21	100.0
49 C	$CaCl_2$ solution	Ja5749.029	74.29	1.72	15 27	3 51		2.49		1.75				0.97	0.97	100.0
	Ambient org. extract	Ja5749.037	85.57	3.02	4.99	2.48		0.92		2.77				0.25	0.25	100.0
	Agressive org. extract	Ja5749.045	71.17	4.45	10 27	4 81	0 55	2.27		3.00		0.75	1.09	1.64	3.48	100.0
49 D	$CaCl_2$ solution	Ja5749.030	74.07	1.65	14 93	3 84	0 80	2.90		1.26				0.55	0.55	100.0
	Ambient org. extract	Ja5749.038	83.40	3.20	5.62	2.48		1.38		3.86				0.06	0.06	100.0
	Agressive org. extract	Ja5749.046	72.80	4.33	9.92	4.12	0.46	2.87		1.80			0.99	2.71	3.70	100.0
63 C	$CaCl_2$ solution	Ja5763.028	68.97	2.36	18 03	2 60		3.80		2.64				1.60	1.60	100.0
	Ambient org. extract	Ja5763.036	80.90	3.65	5.64	2 04	0 31	1.36		4.68				1.42	1.42	100.0
	Agressive org. extract	Ja5763.044	71.51	4.31	3.22	4 27	0.47	10.27		3.74			0.91	1.30	2 21	100.0

DAT: days after treatment, The sum of HPLC peaks (assigned peaks) and unidentified radioactivity (sum of minor peaks and background signal) is 100%.

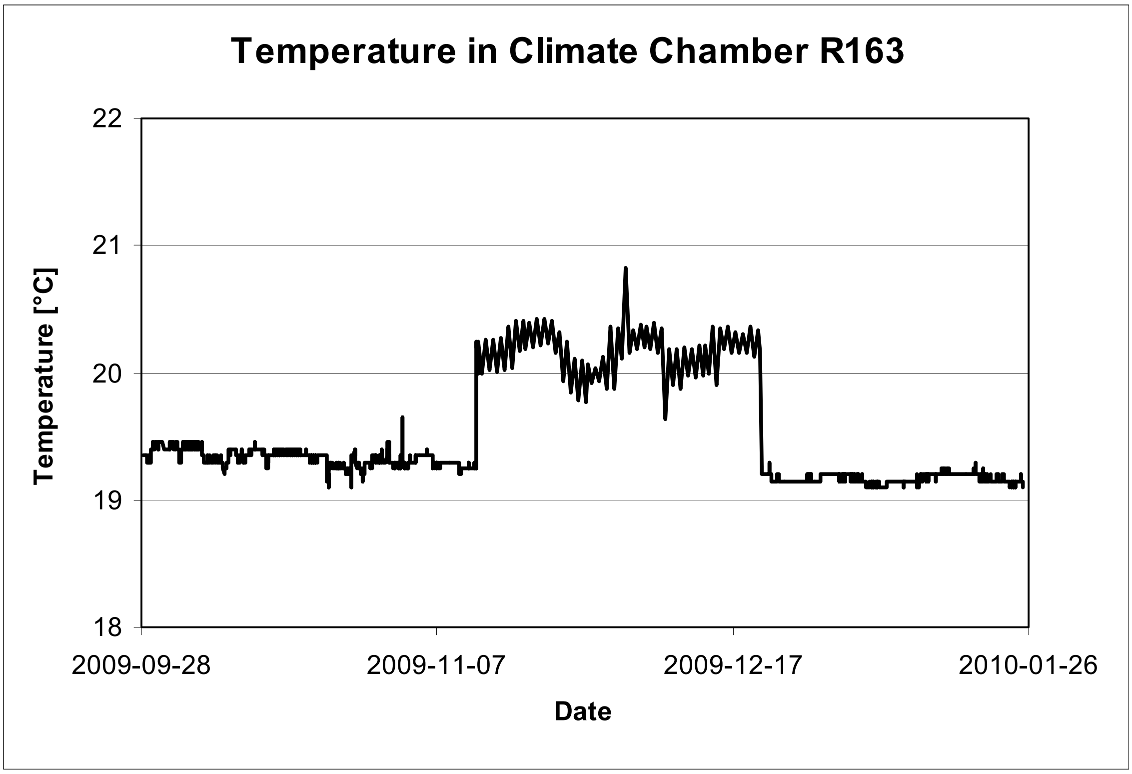
Appendix 14: Raw Data of HPLC Analysis (Evaluation Method), continued

Soil Wellesbourne

DAT (Replicate)	Extract	HPLC ID	Chlothia-nidin	TZNG	MNG	TZMU	TMG	NTG	TZU	TZFA	41	31	30	Diffuse radioactivity	Unidentified radioactivity	Total
63 D	$CaCl_2$ solution	Ja5763.029	70.43	1.49	16.79	2 60		3.30		2 05				3.34	3.34	100 0
	Ambient org. extract	Ja5763.037	82.92	3.65	4 85	2 22		0.72		4 03				1.61	1.61	100 0
	Aggressive org. extract	Ja5763.045	70.69	4.62	4 02	4 50	0.42	9.51		2.72				3.52	3.52	100 0
77 C	$CaCl_2$ solution	Ja5777.006	70.19	1.97	18.19	3 08		3.86		1 51				1 20	1.20	100 0
	Ambient org. extract	Ja5777.014	81.14	4.65	7.71	1 85		1.14		3.19				0.32	0.32	100 0
	Aggressive org. extract	Ja5777.022	72.81	5 35	13.54	3 24		2.97		1.46				0.63	0.63	100 0
77 D	$CaCl_2$ solution	Ja5777.007	67.81	2.33	20.06	2 99		4.13		1.48				1 20	1.20	100 0
	Ambient org. extract	Ja5777.015	82.31	4.25	5 37	1 82		1.21		3.76				1.28	1.28	100 0
	Aggressive org. extract	Ja5777.023	72.17	5.14	11.55	2.70		4.09		1 51				2.84	2.84	100 0
98 C	$CaCl_2$ solution	Ja5798.004	64.16	2.81	19.90	2 03	0.72	6.42		1.14				2 82	2.82	100 0
	Ambient org. extract	Ja5798.012	80.65	4.52	7.46	1 27	0 26	1.74		3 24		0.26		0.60	0.86	100 0
	Aggressive org. extract	Ja5798.020	66.14	6 33	4.48	2 68	0 24	13.50		1 56				5.07	5.07	100 0
98 D	$CaCl_2$ solution	Ja5798.005	66.75	2.09	20.26	1 20		5.71		1.41				2 58	2.58	100 0
	Ambient org. extract	Ja5798.013	80.03	5.34	5.13	1.43	0.19	1.58		4.18				2.12	2.12	100 0
	Aggressive org. extract	Ja5798.021	70.98	6.12	2 39	3 26	0.40	12.86		1 57		0.51		1.91	2.42	100 0
120 C	$CaCl_2$ solution	Ja5712.003	63.50	1.98	24.52	1 09	0 63	5.53		1.13				1 62	1.62	100 0
	Ambient org. extract	Ja5712.011	80.24	4.88	7 30	1 28		1.91		3.75				0.64	0.64	100 0
	Aggressive org. extract	Ja5712.019	68.41	6.19	5.49	2.47		14.47		1.49				1.48	1.48	100 0
120 D	$CaCl_2$ solution	Ja5712.004	61.04	3.12	21.69	1.77	1 05	9.15		1 03				1.15	1.15	100 0
	Ambient org. extract	Ja5712.012	79.24	5.81	7 62	1.47		2.60		2 90				0.36	0.36	100 0
	Aggressive org. extract	Ja5712.020	70.10	6 53	4 92	3 25		14.43						0.77	0.77	100 0

DAT: days after treatment,
The sum of HPLC peaks (assigned peaks) and unidentified radioactivity (sum of minor peaks) is 100%.

Appendix 15: Temperature Record During Incubation



Data analysis start time (start of test incubation)	2009-09-28
Data analysis stop time (termination of test incubation)	2010-01-26
Minimum [°C]	19.1
Maximum [°C]	20.8
Mean [°C]	19.3
Standard deviation [°C]	0.2

Appendix 16: Time-Dependent Sorption Evaluation for Soil Hoefchen am Hohenseh plot 4011, Exemplary Calculation

Soil		Desorption Solution			
Sample ID	DAT	[mL]	Clothianidin [% of applied]	Clothianidin [µg]	Clothianidin [µg/mL]
Ja57 00 A	0	52	22.11	4.39	0.084
Ja57 00 B	0	54	22.58	4.48	0.083
Ja57 01 A	1	53	19.38	3.85	0.073
Ja57 01 B	1	53	19.22	3.82	0.072
Ja57 03 A	3	56	17.12	3.40	0.061
Ja57 03 B	3	52	15.55	3.09	0.059
Ja57 09 A	9	53	8.05	1.60	0.030
Ja57 09 B	9	53	8.32	1.65	0.031
Ja57 21 A	21	52	4.56	0.91	0.017
Ja57 21 B	21	53	7.22	1.43	0.027
Ja57 28 A	28	53	4.66	0.93	0.017
Ja57 28 B	28	54	4.38	0.87	0.016
Ja57 35 A	35	53	3.99	0.79	0.015
Ja57 35 B	35	53	4.00	0.79	0.015
Ja57 49 A	49	52	3.81	0.76	0.015
Ja57 49 B	49	53	3.64	0.72	0.014
Ja57 63 A	63	53	2.97	0.59	0.011
Ja57 63 B	63	54	3.38	0.67	0.012
Ja57 77 A	77	54	2.69	0.53	0.010
Ja57 77 B	77	54	2.89	0.57	0.011
Ja57 98 A	98	53	2.31	0.46	0.009
Ja57 98 B	98	54	1.69	0.34	0.006
Ja57 12 A	120	51	1.76	0.35	0.007
Ja57 12 B	120	53	2.08	0.41	0.008

Soil		Extract					R_{TDS}	$R_{TDS}oc$
Sample ID	DAT	[mL]	Clothianidin [% of applied]	*correction [% of applied]	Clo hianidin [µg]	Clothianidin [µg/g]	Clothianidin [mL/g]	Clothianidin [mL/g]
Ja57 00 A	0	376	74.23	53.82	10.68	0.107	1.3	115.0
Ja57 00 B	0	380	74.96	55.73	11.06	0.111	1.3	121.2
Ja57 01 A	1	364	65.99	48.80	9.69	0.097	1.3	121.3
Ja57 01 B	1	363	67.38	50.33	9.99	0.100	1.4	126.2
Ja57 03 A	3	379	61.32	47.87	9.50	0.095	1.6	142.4
Ja57 03 B	3	380	61.67	47.32	9.39	0.094	1.6	143.9
Ja57 09 A	9	379	43.02	35.88	7.12	0.071	2.4	214.7
Ja57 09 B	9	379	42.72	35.35	7.02	0.070	2.3	204.8
Ja57 21 A	21	366	31.04	26.83	5.33	0.053	3.1	278.2
Ja57 21 B	21	367	30.97	24.57	4.88	0.049	1.8	164.0
Ja57 28 A	28	382	33.04	28.90	5.74	0.057	3.3	298.7
Ja57 28 B	28	382	31.05	27.32	5.42	0.054	3.4	306.2
Ja57 35 A	35	372	25.76	22.22	4.41	0.044	3.0	268.5
Ja57 35 B	35	378	27.99	24.44	4.85	0.049	3.2	294.3
Ja57 49 A	49	374	25.49	21.98	4.36	0.044	3.0	273.0
Ja57 49 B	49	373	25.80	22.57	4.48	0.045	3.3	298.7
Ja57 63 A	63	372	23.36	20.72	4.11	0.041	3.7	336.0
Ja57 63 B	63	375	24.07	21.20	4.21	0.042	3.4	308.3
Ja57 77 A	77	368	23.58	21.29	4.23	0.042	4.3	388.8
Ja57 77 B	77	371	24.62	22.15	4.40	0.044	4.1	375.8
Ja57 98 A	98	377	20.09	18.04	3.58	0.036	4.1	375.8
Ja57 98 B	98	378	20.30	18.86	3.75	0.037	6.0	548.4
Ja57 12 A	120	380	18.68	16.99	3.37	0.034	4.9	447.0
Ja57 12 B	120	379	20.56	18.72	3.72	0.037	4.8	434.5

* The amount of Clothianidin has to be corrected: amount in the extract - amount in the residual desorption solution (soil pore water)

Appendix 16: Time-Dependent Sorption Evaluation for Soil Hoefchen am Hohenseh plot 4011, Exemplary Calculation (continued)

Determination of distribution coefficient and sorption coefficient:

For each desorption sample, distribution coefficients R_{TDS} were obtained and calculated according to the formula:

$$R_{TDS} = C_{extr} / C_{des} \ [mL/g]$$

where

$$C_{des} = Ai_{des} / V_1 \ [\mu g/mL]$$

and

$$C_{extr} = (A.i_{org} + A.i._{hot} - (C_{des} * V_2)) / W_{soil} \ [\mu g/g] \quad (1)$$

V_0	original volume of the desorption solution employed [mL] (added volume + soil moisture)
V_1	volume of the desorption solution [mL] decanted after centrifugation (supernatant)
V_2	soil pore water, volume of water remaining in soil after desorption (V_0-V_1) [mL]
C_{des}	concentration of test item in aqueous desorption solution / supernatant at equilibrium [µg/mL]
C_{extr}	concentration of test item in sum of organic extracts related to soil dry weight at equilibrium [µg/g]
W_{soil}	dry weight of soil [g]
Ai_{des}	test item in aqueous desorption solution [µg]
Ai_{org}	test item in organic (ambient) extract [µg]
Ai_{hot}	test item in hot organic (microwave) extract [µg]

Equation (1) for calculation of the organically extracted (adsorbed) substance amount, C_{extr} takes into account that after addition of the desorption solution to the (non water-saturated) soil and after shaking and centrifugation, a residual volume of the desorption solution (containing dissolved test item) is still present within the soil. This portion of aqueous solution is named soil pore water V_2. According to OECD TG Guideline 106 [1] the amount of test item contained in the soil pore water is calculated by subtraction of the volume of the desorption solution V_1 (supernatant after centrifugation) from the original volume of solution V_0 employed to soil, multiplied by the measured concentration of test item in the desorption solution C_{des} (equation (1))[1]

Calculation for sample Ja57 00 A:

C_{des} = 4.39 µg / 52 mL = 0.084 µg/mL
C_{extr} = 10.68 µg / 100 g = 0.107 µg/g

[1] This calculation is based on the assumption that the concentration of the test item in the soil pore water remains constant after contact to soil.

Appendix 17: Time-Dependent Sorption Evaluation for Soil Wellesbourne

Soil		Desorption Solution			
Sample ID	DAT	[mL]	Clothianidin [% of applied]	Clothianidin [µg]	Clothianidin [µg/mL]
Ja57 00 C	0	59	30.68	6.09	0.103
Ja57 00 D	0	61	31.65	6.28	0.103
Ja57 01 C	1	61	28.92	5.74	0.094
Ja57 01 D	1	62	29.16	5.79	0.093
Ja57 03 C	3	69	28.04	5.57	0.081
Ja57 03 D	3	61	26.87	5.34	0.087
Ja57 09 C	9	61	19.18	3.81	0.062
Ja57 09 D	9	61	19.70	3.91	0.064
Ja57 21 C	21	60	11.44	2.27	0.038
Ja57 21 D	21	61	16.51	3.28	0.054
Ja57 28 C	28	61	14.65	2.91	0.048
Ja57 28 D	28	61	14.44	2.87	0.047
Ja57 35 C	35	60	13.41	2.66	0.044
Ja57 35 D	35	60	13.93	2.77	0.046
Ja57 49 C	49	61	12.07	2.40	0.039
Ja57 49 D	49	61	12.07	2.40	0.039
Ja57 63 C	63	62	10.51	2.09	0.034
Ja57 63 D	63	64	11.32	2.25	0.035
Ja57 77 C	77	63	10.67	2.12	0.034
Ja57 77 D	77	64	10.55	2.09	0.033
Ja57 98 C	98	60	8.48	1.68	0.028
Ja57 98 D	98	60	8.95	1.78	0.030
Ja57 12 C	120	60	8.05	1.60	0.027
Ja57 12 D	120	59	7.60	1.51	0.026

Soil		Extract					R_{TDS}	R_{TDS}oc
Sample ID	DAT	[mL]	Clothianidin [% of applied]	*correction [% of applied]	Clothianidin [µg]	Clo hianidin [µg/g]	Clothianidin [mL/g]	Clo hianidin [mL/g]
Ja57 00 C	0	380	69.76	48.44	9.62	0.096	0.9	116.4
Ja57 00 D	0	384	67.42	47.18	9.37	0.094	0.9	113.7
Ja57 01 C	1	355	63 07	44.58	8.85	0.089	0.9	117.6
Ja57 01 D	1	371	62 28	44.41	8.82	0.088	0.9	118.0
Ja57 03 C	3	384	59.72	47.12	9.36	0.094	1.2	145.0
Ja57 03 D	3	385	62 56	45.38	9.01	0.090	1.0	128.8
Ja57 09 C	9	390	56 38	44.12	8.76	0.088	1.4	175.4
Ja57 09 D	9	378	56 39	43.79	8.69	0.087	1.4	169.5
Ja57 21 C	21	373	51.60	43.97	8.73	0.087	2.3	288.3
Ja57 21 D	21	376	49.45	38.89	7.72	0.077	1.4	179.6
Ja57 28 C	28	378	50.42	41.06	8.15	0.082	1.7	213.7
Ja57 28 D	28	365	49 89	40.66	8.07	0.081	1.7	214.6
Ja57 35 C	35	374	48.14	39.20	7.78	0.078	1.8	219.2
Ja57 35 D	35	373	47.62	38.34	7.61	0.076	1.7	206.4
Ja57 49 C	49	374	46 26	38.55	7.65	0.077	1.9	243.5
Ja57 49 D	49	394	46.61	38.89	7.72	0.077	2.0	245.7
Ja57 63 C	63	379	41 98	35.54	7.06	0.071	2.1	262.1
Ja57 63 D	63	376	43 86	37.49	7.44	0.074	2.1	264.9
Ja57 77 C	77	371	42.48	36.22	7.19	0.072	2.1	267.2
Ja57 77 D	77	374	42.80	36.86	7 32	0.073	2.2	279.7
Ja57 98 C	98	381	39.52	33.87	6.72	0.067	2.4	299.5
Ja57 98 D	98	382	39.83	33.86	6.72	0.067	2.3	283.9
Ja57 12 C	120	389	38.38	33.02	6 55	0.066	2 5	307.5
Ja57 12 D	120	386	38.35	33.07	6 57	0.066	2.6	320.9

* The amount of Clothianidin has to be corrected: amount in the extract - amount in the residual desorption solution (soil pore water)

Appendix 18: Time-Dependent Sorption Evaluation for Mas du Coq

Soil		Desorption Solution			
Sample ID	DAT	[mL]	Clothianidin [% of applied]	Clo hianidin [µg]	Clothianidin [µg/mL]
Ja57 00 E	0	58	36.14	7.18	0.124
Ja57 00 F	0	59	36.06	7.16	0.121
Ja57 01 E	1	58	33.46	6.64	0.115
Ja57 01 F	1	58	33.49	6.65	0.115
Ja57 03 E	3	58	29.22	5.80	0.100
Ja57 03 F	3	57	29.89	5.94	0.104
Ja57 09 E	9	59	26.73	5.31	0.090
Ja57 09 F	9	60	27.54	5.47	0.091
Ja57 21 E	21	58	22.09	4.39	0.076
Ja57 21 F	21	58	23.11	4.59	0.079
Ja57 28 E	28	58	20.34	4.04	0.070
Ja57 28 F	28	58	21.29	4.23	0.073
Ja57 35 E	35	57	19.36	3.84	0.067
Ja57 35 F	35	58	20.64	4.10	0.071
Ja57 49 E	49	60	18.66	3.71	0.062
Ja57 49 F	49	60	18.60	3.69	0.062
Ja57 63 E	63	56	16.55	3.29	0.059
Ja57 63 F	63	56	16.94	3.36	0.060
Ja57 77 E	77	59	17.22	3.42	0.058
Ja57 77 F	77	59	16.47	3.27	0.055
Ja57 98 E	98	57	13.10	2.60	0.046
Ja57 98 F	98	59	14.23	2.82	0.048
Ja57 12 E	120	56	13.04	2.59	0.046
Ja57 12 F	120	57	13.33	2.65	0.046

Soil		Extract					R_{TDS}	$R_{TDS}oc$
Sample ID	DAT	[mL]	Clothianidin [% of applied]	*correc ion [% of applied]	Clothianidin [µg]	Clo hianidin [µg/g]	Clothianidin [mL/g]	Clothianidin [mL/g]
Ja57 00 E	0	379	62.74	36.57	7.26	0.073	0.6	73.4
Ja57 00 F	0	377	63.45	38.40	7.62	0.076	0.6	78.5
Ja57 01 E	1	364	60.46	36.23	7.19	0.072	0.6	78.5
Ja57 01 F	1	368	61 30	37.05	7.36	0.074	0.6	80.2
Ja57 03 E	3	377	61 01	39.85	7.91	0.079	0.8	98.9
Ja57 03 F	3	377	61 96	39.41	7.82	0.078	0.8	93.9
Ja57 09 E	9	377	60 50	41.93	8.32	0.083	0.9	115.7
Ja57 09 F	9	378	60 84	42.48	8.43	0.084	0.9	115.7
Ja57 21 E	21	366	57 30	41.30	8.20	0.082	1.1	135.5
Ja57 21 F	21	366	58.63	41.90	8.32	0.083	1.1	131.5
Ja57 28 E	28	374	56.74	42.01	8.34	0.083	1.2	149.8
Ja57 28 F	28	375	56 88	41.46	8.23	0.082	1.1	141.2
Ja57 35 E	35	372	56 21	41.60	8.26	0.083	1.2	153.1
Ja57 35 F	35	377	55 37	40.43	8.03	0.080	1.1	142.0
Ja57 49 E	49	375	54.75	42.31	8.40	0.084	1.4	170.0
Ja57 49 F	49	374	53 26	40.86	8.11	0.081	1.3	164.7
Ja57 63 E	63	377	54 22	41.22	8.18	0.082	1.4	174.4
Ja57 63 F	63	379	52.64	39.33	7.81	0.078	1.3	162.6
Ja57 77 E	77	373	53.62	41.65	8.27	0.083	1.4	178.4
Ja57 77 F	77	372	52 88	41.43	8.23	0.082	1.5	185.5
Ja57 98 E	98	382	49.60	39.72	7.89	0.079	1.7	216.1
Ja57 98 F	98	382	49 85	39.96	7.93	0.079	1.7	207.1
Ja57 12 E	120	376	48 22	37.97	7.54	0.075	1.6	203.9
Ja57 12 F	120	377	46 91	36.86	7.32	0.073	1.6	197.0

* The amount of Clothianidin has to be corrected: amount in the extract - amount in the residual desorption solution (soil pore water)

Appendix 19: Time-Dependent Sorption Evaluation for Vilobi d'Onyar

Soil		Desorption Solution			
Sample ID	DAT	[mL]	Clothianidin [% of applied]	Clothianidin [µg]	Clothianidin [µg/mL]
Ja57 00 G	0	61	33.37	6.62	0.109
Ja57 00 H	0	60	33.76	6.70	0.112
Ja57 01 G	1	60	30.68	6.09	0.102
Ja57 01 H	1	60	30.85	6.12	0.102
Ja57 03 G	3	60	28.53	5.67	0.094
Ja57 03 H	3	59	26.77	5.31	0.090
Ja57 09 G	9	60	21.32	4.23	0.071
Ja57 09 H	9	61	22.07	4.38	0.072
Ja57 21 G	21	58	17.84	3.54	0.061
Ja57 21 H	21	59	17.04	3.38	0.057
Ja57 28 G	28	60	15.72	3.12	0.052
Ja57 28 H	28	60	15.86	3.15	0.052
Ja57 35 G	35	59	14.75	2.93	0.050
Ja57 35 H	35	58	15.06	2.99	0.052
Ja57 49 G	49	60	13.50	2.68	0.045
Ja57 49 H	49	61	13.89	2.76	0.045
Ja57 63 G	63	58	11.50	2.28	0.039
Ja57 63 H	63	58	12.38	2.46	0.042
Ja57 77 G	77	62	11.92	2.37	0.038
Ja57 77 H	77	62	12.09	2.40	0.039
Ja57 98 G	98	59	10.33	2.05	0.035
Ja57 98 H	98	60	9.95	1.98	0.033
Ja57 12 G	120	57	8.38	1.66	0.029
Ja57 12 H	120	59	8.87	1.76	0.030

Soil		Extract					R_{TDS}	$R_{TDS}oc$
Sample ID	DAT	[mL]	Clothianidin [% of applied]	*correction [% of applied]	Clothianidin [µg]	Clothianidin [µg/g]	Clothianidin [mL/g]	Clothianidin [mL/g]
Ja57 00 G	0	356	64.89	43.56	8.65	0.086	0.8	79.6
Ja57 00 H	0	367	66.25	43.75	8.69	0.087	0.8	77.7
Ja57 01 G	1	357	63.38	42.92	8.52	0.085	0.8	83.9
Ja57 01 H	1	356	62 00	41.43	8.23	0.082	0.8	80.6
Ja57 03 G	3	359	62 27	43.25	8.59	0.086	0.9	90.9
Ja57 03 H	3	359	61 07	42.46	8.43	0.084	0.9	93.6
Ja57 09 G	9	367	59.66	45.45	9.02	0.090	1.3	127.9
Ja57 09 H	9	370	60 35	46.24	9.18	0.092	1.3	127.8
Ja57 21 G	21	340	53.42	40.51	8.04	0.080	1.3	131.7
Ja57 21 H	21	354	53 92	42.08	8.35	0.084	1.5	145.7
Ja57 28 G	28	375	54.73	44.25	8.79	0.088	1.7	168.9
Ja57 28 H	28	377	54 30	43.73	8.68	0.087	1.7	165.5
Ja57 35 G	35	375	52.41	42.16	8.37	0.084	1.7	168.7
Ja57 35 H	35	376	53 58	42.67	8.47	0.085	1.6	164.4
Ja57 49 G	49	370	50 83	41.83	8.31	0.083	1.9	185.9
Ja57 49 H	49	374	49.71	40.83	8.11	0.081	1.8	179.3
Ja57 63 G	63	388	49 87	41.54	8.25	0.082	2.1	209.5
Ja57 63 H	63	384	49 24	40.28	8.00	0.080	1.9	188.7
Ja57 77 G	77	369	48 32	41.01	8.14	0.081	2.1	213.2
Ja57 77 H	77	371	46.76	39.35	7.81	0.078	2.0	201.8
Ja57 98 G	98	381	43.11	35.93	7.13	0.071	2.1	205.1
Ja57 98 H	98	377	43.64	37.01	7.35	0.073	2.2	223.2
Ja57 12 G	120	379	42.67	36.34	7.22	0.072	2.5	247.1
Ja57 12 H	120	383	43 20	37.03	7.35	0.074	2.5	246.2

* The amount of Clothianidin has to be corrected: amount in the extract - amount in the residual desorption solution (soil pore water)

Appendix 20: Test of Simplified Extraction Method

Material balance of radioactivity of Clothianidin from aerobic soil metabolism (expressed as percent of applied radioactivity) using different extraction methods

Soil: Höfchen 4011

		Replicate No.	DAT								
			TDS method			Microwave, calc. by supernatant			Microwave/NER/CO_2 for mass balance		
			0	58	124	0	58	124	0	58	124
Volatiles											
	$^{14}CO_2$	1	n.a.	9.3	12.8	n.a.	n.a.	n.a.	n.a.	9.3	13.2
		2	n.a.	9.7	13.0	n.a.	n.a.	n.a.	n.a.	9.6	13.3
		Mean		**9.5**	**12.9**					**9.5**	**13.3**
	Volatile organics	1	n.a.	<0.1	<0.1	n.a.	n.a.	n.a.	n.a.	<0.1	<0.1
		2	n.a.	<0.1	0.1	n.a.	n.a.	n.a.	n.a.	<0.1	<0.1
		Mean			**0.1**						
	Total	1	n.a.	9.3	12.8	n.a.	n.a.	n.a.	n.a.	9.3	13.2
		2	n.a.	9.7	13.2	n.a.	n.a.	n.a.	n.a.	9.6	13.3
		Mean		**9.5**	**13.0**					**9.5**	**13.3**
Extractable Radioactivity											
	Calcium chloride solution	1	21.1	8.9	7.3	n.a.	n.a.	n.a.	n.a.	n.a.	n.a.
		2	20.9	8.8	7.5	n.a.	n.a.	n.a.	n.a.	n.a.	n.a.
		Mean	**21.0**	**8.9**	**7.4**						
	Organic extract	1	72.4	52.8	44.8	n.a.	n.a.	n.a.	n.a.	n.a.	n.a.
		2	73.3	53.8	45.9	n.a.	n.a.	n.a.	n.a.	n.a.	n.a.
		Mean	**72.8**	**53.3**	**45.3**						
	Micro wave	1	2.3	9.9	10.4	101.0	77.4	71.3	79.4	60.9	55.4
		2	2.5	9.9	11.1	102.1	77.3	70.1	79.4	60.8	54.5
		Mean	**2.4**	**9.9**	**10.7**	**101.5**	**77.4**	**70.7**	**79.4**	**60.8**	**55.0**
	Total	1	95.8	71.6	62.4	101.0	77.4	71.3	79.4	60.9	55.4
		2	96.7	72.5	64.4	102.1	77.3	70.1	79.4	60.8	54.5
		Mean	**96.2**	**72.1**	**63.4**	**101.5**	**77.4**	**70.7**	**79.4**	**60.8**	**55.0**
Bound Residue		1	2.7	15.6	20.4	n.a.	n.a.	n.a.	18.5	26.6	28.9
		2	3.2	15.8	21.4	n.a.	n.a.	n.a.	18.6	26.7	29.0
		Mean	**2.9**	**15.7**	**20.9**				**18.6**	**26.7**	**28.9**
Material Balance		1	98.5	96.5	95.6	101.0	77.4	71.3	97.9	96.8	97.5
		2	99.8	98.0	99.0	102.1	77.3	70.1	98.0	97.1	96.8
		Mean	**99.2**	**97.2**	**97.3**	**101.5**	**77.4**	**70.7**	**98.0**	**97.0**	**97.2**

n.d. : not detected, n.a. : not analyzed, DAT : day after treatment

Appendix 20: Test of Simplified Extraction Method (continued)

Material balance of radioactivity of Clothianidin from aerobic soil metabolism (expressed as percent of applied radioactivity) using different extraction methods

Soil: Vilobi d´Ónyar

		Rep-licate No.	DAT										
			TDS method				Microwave. calc. by supernatant				Microwave/NER/CO_2 for mass balance		
			0	58	124		0	58	124		0	58	124
Volatiles													
	$^{14}CO_2$	1	n.a.	11.5	17.1		n.a.	n.a.	n.a.		n.a.	11.4	16.9
		2	n.a.	11.4	17.1		n.a.	n.a.	n.a.		n.a.	11.4	17.7
		Mean		**11.5**	**17.1**							**11.4**	**17.3**
	Volatile organics	1	n.a.	<0.1	<0.1		n.a.	n.a.	n.a.		n.a.	<0.1	<0.1
		2	n.a.	<0.1	<0.1		n.a.	n.a.	n.a.		n.a.	<0.1	<0.1
		Mean											
	Total	1	n.a.	11.5	17.1		n.a.	n.a.	n.a.		n.a.	11.4	16.9
		2	n.a.	11.4	17.1		n.a.	n.a.	n.a.		n.a.	11.4	17.7
		Mean		**11.5**	**17.1**							**11.4**	**17.3**
Extractable Radioactivity													
	Calcium chloride solution	1	26.3	12.0	10.7		n.a.	n.a.	n.a.		n.a.	n.a.	n.a.
		2	26.4	11.9	9.5		n.a.	n.a.	n.a.		n.a.	n.a.	n.a.
		Mean	**26.4**	**12.0**	**10.1**								
	Organic extract	1	66.7	49.7	43.7		n.a.	n.a.	n.a.		n.a.	n.a.	n.a.
		2	68.5	50.4	43.5		n.a.	n.a.	n.a.		n.a.	n.a.	n.a.
		Mean	**67.6**	**50.1**	**43.6**								
	Micro wave	1	2.0	7.6	8.4		96.8	74.6	61.1		79.5	59.9	48.6
		2	2.1	7.8	8.6		102.5	74.2	62.1		82.4	59.6	48.8
		Mean	**2.0**	**7.7**	**8.5**		**99.7**	**74.4**	**61.6**		**81.0**	**59.8**	**48.7**
	Total	1	95.0	69.2	62.8		96.8	74.6	61.1		79.5	59.9	48.6
		2	97.0	70.2	61.5		102.5	74.2	62.1		82.4	59.6	48.8
		Mean	**96.0**	**69.7**	**62.2**		**99.7**	**74.4**	**61.6**		**81.0**	**59.8**	**48.7**
Bound Residue		1	2.3	13.2	16.3		n.a.	n.a.	n.a.		16.9	23.1	29.7
		2	2.7	13.2	16.6		n.a.	n.a.	n.a.		15.7	23.0	28.7
		Mean	**2.5**	**13.2**	**16.4**						**16.3**	**23.1**	**29.2**
Material Balance		1	97.3	94.0	96.2		96.8	74.6	61.1		96.4	94.4	95.2
		2	99.7	94.8	95.2		102.5	74.2	62.1		98.1	94.1	95.3
		Mean	**98.5**	**94.4**	**95.7**		**99.7**	**74.4**	**61.6**		**97.3**	**94.2**	**95.2**

n.d. : not detected, n.a. : not analyzed, DAT : day after treatment

Appendix 20: Test of Simplified Extraction Method (continued)

Biotransformation of Clothianidin (expressed as percentage of applied radioactivity), different extraction methods

Soil: Höfchen 4011

Compound	Replicate	DAT				DAT		
		TDS Extraction method				Microwave Extraction method		
	No.	0	58	124		0	58	124
Clothianidin	1	95.8	60.8	45.6		101.0	63.1	52.8
	2	96.7	61.4	48.4		102.1	62.4	52.2
	Mean	**96.2**	**61.1**	**47.0**		**101.5**	**62.7**	**52.5**
TZNG	1	n.d.	2.1	2.8		n.d.	2.7	2.7
	2	n.d.	2.1	3.0		n.d.	2.5	2.6
	Mean		2.1	2.9			2.6	2.6
MNG	1	n.d.	6.5	9.3		n.d.	6.9	9.4
	2	n.d.	6.5	8.6		n.d.	7.5	9.2
	Mean		6.5	9.0			7.2	9.3
TZMU	1	n.d.	1.6	0.2		n.d.	1.8	1.3
	2	n.d.	1.8	0.3		n.d.	1.7	1.0
	Mean		1.7	0.2			1.7	1.2
TMG	1	n.d.	n.d.	n.d.		n.d.	1.2	1.3
	2	n.d.	n.d.	n.d.		n.d.	1.2	1.1
	Mean						1.2	1.2
NTG	1	n.d.	0.5	4.6		n.d.	1.9	3.8
	2	n.d.	0.7	4.1		n.d.	2.0	4.0
	Mean		0.6	4.3			1.9	3.9
TZU	1	n.d.	n.d.	n.d.		n.d.	n.d.	n.d.
	2	n.d.	n.d.	n.d.		n.d.	n.d.	n.d.
	Mean							
TZFA	1	n.d.	n.d.	n.d.		n.d.	n.d.	n.d.
	2	n.d.	n.d.	n.d.		n.d.	n.d.	n.d.
	Mean							
Unidentified radioactivity	1	n.d.	n.d.	n.d.		n.d.	n.d.	n.d.
	2	n.d.	n.d.	n.d.		n.d.	n.d.	n.d.
	Mean							
Total extractable residues	1	95.8	71.6	62.4		101.0	77.4	71.3
	2	96.7	72.5	64.4		102.1	77.3	70.1
	Mean	**96.2**	**72.1**	**63.4**		**101.5**	**77.4**	**70.7**

n.d. : not detected, n.a. : not analyzed, DAT : day after treatment

Appendix 20: Test of Simplified Extraction Method (continued)

Biotransformation of Clothianidin (expressed as percentage of applied radioactivity), different extraction methods

Soil: Vilobi d'Onyar

Compound	Replicate	DAT				DAT		
		TDS Extraction method				Microwave Extraction method		
	No.	0	58	124		0	58	124
Clothianidin	1	95.0	60.4	51.1		96.8	61.5	49.1
	2	97.0	60.7	50.6		102.5	60.8	49.5
	Mean	**96.0**	**60.5**	**50.9**		**99.7**	**61.1**	**49.3**
TZNG	1	n.d.	2.2	2.2		n.d.	2.7	2.9
	2	n.d.	2.9	2.9		n.d.	2.7	2.3
	Mean		2.5	2.5			2.7	2.6
MNG	1	n.d.	5.9	5.5		n.d.	5.2	5.0
	2	n.d.	5.4	4.9		n.d.	5.0	5.0
	Mean		5.7	5.2			5.1	5.0
TZMU	1	n.d.	n.d.	0.2		n.d.	1.3	0.6
	2	n.d.	n.d.	n.d.		n.d.	1.1	0.7
	Mean			0.1			1.2	0.6
TMG	1	n.d.	n.d.	0.2		n.d.	0.5	n.d.
	2	n.d.	n.d.	n.d.		n.d.	0.5	0.8
	Mean			0.1			0.5	0.4
NTG	1	n.d.	0.7	3.6		n.d.	1.7	3.6
	2	n.d.	1.1	3.2		n.d.	2.0	3.9
	Mean		0.9	3.4			1.8	3.7
TZU	1	n.d.	n.d.	n.d.		n.d.	n.d.	n.d.
	2	n.d.	n.d.	n.d.		n.d.	n.d.	n.d.
	Mean							
TZFA	1	n.d.	n.d.	n.d.		n.d.	1.8	n.d.
	2	n.d.	n.d.	n.d.		n.d.	1.8	n.d.
	Mean						1.8	
Unidentified radioactivity	1	n.d.	n.d.	n.d.		n.d.	n.d.	n.d.
	2	n.d.	n.d.	n.d.		n.d.	0.4	n.d.
	Mean						0.2	
Total extractable residues	1	95.0	69.2	62.8		96.8	74.6	61.1
	2	97.0	70.2	61.5		102.5	74.2	62.1
	Mean	**96.0**	**69.7**	**62.2**		**99.7**	**74.4**	**61.6**

n.d. : not detected, n.a. : not analyzed, DAT : day after treatment

Appendix 21: Certificate of Analysis of [Guanidine-^{14}C]Clothianidin

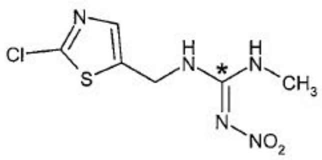
Dr. 5.1.2.e WOO
Bayer CropScience AG
Product Technology
Isotope Chemistry
Aprather Weg 18 a (Building 466)
D-42096 Wuppertal, Germany
5.1.2.e WOO@bayercropscience.com

Bayer CropScience

Radiochemical Data Sheet

Substance : **[guanidine-^{14}C] Clothianidin**

Sample-ID : **KATH 6360**



Total activity	:	44.4 MBq (1.2 mCi)
Specific activity	:	4.34 MBq (117.2 µCi)/mg
Reference synthesis	:	KML 2998-1
Form	:	solid, dried in vacuo
*	:	position of label

Radiochemical purity	:	> 98 %
Method	:	HPLC, radioactivity-detector
Column	:	Nucleodur C18 Gravity®, 5 µm, 125 x 4 mm
Flow rate	:	1.5 ml/min
Eluent	:	A = 0.2 % phosphoric acid, B = acetonitrile
Gradient	:	5 min 0 % B, at 35 min 100 % B, at 40 min 100 % B

Radiochemical purity	:	> 98 %
Method	:	TLC, scan
Plate	:	silica gel Merck 60 F 254®
Eluent	:	acetonitrile

Chemical purity	:	> 98 %
Method	:	HPLC, UV-detector, 210 nm conditions as above

Signature : 5.1.2.e WOO

- 5.1.2.e WOO

Date : 2009-09-10

KATH2009-187.DOC

Appendix 22: GLP Certificate of the Test Facility

Ministerium für Arbeit, Gesundheit und Soziales
des Landes Nordrhein-Westfalen

Fürstenwall 25. 40219 Düsseldorf

Aktenzeichen II A 5 – 31.11.62.05

Gute Laborpraxis/Good Laboratory Practice
GLP-Bescheinigung/Statement of GLP Compliance
(gemäß/according to § 19b Abs. 1 Chemikaliengesetz)

Eine GLP-Inspektion zur Überwachung der Einhaltung der GLP-Grundsätze gemäß Chemikaliengesetz bzw. Richtlinie 88/320/EG wurde durchgeführt in:	Assessment of conformity with GLP according to Chemikaliengesetz and Directive 88/320/EEC at:
☒ Prüfeinrichtung/Test facility	☐ Prüfstandort/Test site

Bayer CropScience Development
Environmental Safety – Metabolism/ADME and Environmental Fate
BCS-D-EnSa-MeA/Efate
Building 6650-6670
D-40789 Monheim

Prüfungen nach Kategorien (gemäß ChemVwV-GLP Nr. 5.3/OECD guidance)	**Areas of Expertise** (according ChemVwV GLP Nr. 5.3/OECD guidance)
Kategorie 1	category 1
Prüfungen zur Bestimmung der physikalisch-chemischen Eigenschaften und Gehaltsbestimmungen	physical-chemical testing
Kategorie 4	category 4
Ökotoxikologische Prüfungen zur Bestimmung der Auswirkungen auf aquatische und terrestrische Organismen	environmental toxicity studies on aquatic and terrestrial organisms
Kategorie 5	category 5
Prüfungen zum Verhalten im Boden, im Wasser und in der Luft; Prüfungen zur Bioakkumulation und zur Metabolisierung	studies on behaviour in water, soil and air; bioaccumulation

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Appendix 22: GLP Certificate of the Test Facility (continued)

Kategorie 6	category 6
Prüfungen zur Bestimmung von Rückständen	residue studies
Kategorie 8	category 8
Analytische Prüfungen an biologischen Materialien	analytical and clinical chemistry testing
Datum der Inspektion	Date of Inspection
07. bis 09. Mai 2007	on 07 until 09 May 2007
Die genannte Prüfeinrichtung befindet sich im nationalen GLP-Überwachungsverfahren und wird regelmäßig auf Einhaltung der GLP-Grundsätze überwacht.	The above mentioned test facility is included in the national GLP Compliance Programme and is inspected on a regular basis.
Auf der Grundlage des Inspektionsberichtes wird hiermit bestätigt, dass in dieser Prüfeinrichtung die oben genannten Prüfungen unter Einhaltung der GLP-Grundsätze durchgeführt werden können.	Based on the inspection report it can be confirmed, that this test facility is able to conduct the aforementioned studies in compliance with the Principles of GLP.

Düsseldorf, den 16. Juni 2009
Im Auftrag

5.1.2.e WOO

Land Nordrhein-Westfalen 13 Ministerium für Arbeit, Gesundheit und Soziales

Dienstsiegel/official-seal

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