European Commission

Combined Draft Renewal Assessment Report prepared according to Regulation (EC) N° 1107/2009
and
Proposal for Harmonised Classification and Labelling (CLH Report) according to Regulation (EC) N° 1272/2008

LENACIL

Volume 3 – B.7 (AS)

Rapporteur Member State: Belgium
Co-Rapporteur Member State: Austria

Version History

When	What
November 2007 – July 2009	Draft Assessment Report (DAR) – prepared by RMS BE in the context of the inclusion of the a.s. in Annex I to Council Directive 91/414/EEC. Updated versions of the initial DAR, as well as addenda to the initial DAR, were issued in the period February 2009 – May 2009 (before and after experts' meetings) and were compiled by EFSA in a final 'addendum' dated July 2009.
December 2012 – March 2013	Addenda to DAR Vol.3, B.8 and **B.7** (Environmental Fate & Behaviour and **Residues**), respectively – prepared by RMS BE in the context of the evaluation of confirmatory information requested by Commission Directive 2010/39/EU.
May 2016	Update of DAR Vol.3, B.6 (Toxicology and metabolism) – prepared by RMS BE in the context of the evaluation of confirmatory data on the relevance of ground water metabolites (following classification of lenacil according to Reg. (EC) No 1272/2008).
May 2019	Draft Renewal Assessment Report (DRAR) – prepared by RMS BE in the context of the application for renewal of approval of the a.s. according to Reg. (EU) No 844/2012. *Note: The DRAR is a stand-alone document containing the evaluations already displayed in the initial DAR (incl. its addenda and updated versions), as well as the new assessments. The revision of the initial DAR has been done in accordance with SANCO/10180/2013 rev.1 (March 2013), with changes to the original text – resulting from assessment of new studies (or reconsideration of old studies or studies that were not yet previously peer-reviewed) – being highlighted by means of yellow shading.*

The RMS is the author of the Assessment Report. The Assessment Report is based on the validation by the RMS, and the verification during the EFSA peer-review process, of the information submitted by the Applicant in the dossier, including the Applicant's assessments provided in the summary dossier. As a consequence, data and information including assessments and conclusions, validated and verified by the RMS experts, may be taken from the applicant's (summary) dossier and included as such or adapted/modified by the RMS in the Assessment Report. For reasons of efficiency, the Assessment Report should include the information validated/verified by the RMS, without detailing which elements have been taken or modified from the Applicant's assessment. As the Applicant's summary dossier is published, the experts, interested parties, and the public may compare both documents for getting details on which elements of the Applicant's dossier have been validated/verified and which ones have been modified by the RMS.

Table of contents

B.7. RESIDUE DATA

B.7.1. STORAGE STABILITY OF RESIDUES

Report author	Hamberger, R.
Report year	2002
Report title	Analytical Report. Generation of Samples for the Determination of Residues of Venzar 80 % WP (containing 80% Lenacil) in Sugar Beets, One Site in Europe, 2002
Report No	20011048/E2-FPSB
Guidelines followed in study	Commission Working Document 7032/VI/95 rev.5.
Major deviations from test guideline	None
Guidance in force at time of submission of supplementary dossier	OECD TG 506 (16 Oct. 2007)
Previous evaluation	DAR (BE, 2007-2009); Additional details reported in DRAR (BE, 2019)
GLP	Yes

Material and Methods:

The storage stability of Lenacil under frozen conditions was determined by fortification of the control samples of sugar beet leaves and roots with Lenacil standard solution at the fortification levels of 4.0 ppm and 0.2 ppm, respectively. The sugar beet samples were homogenised prior to fortification and stored deep-frozen (<–18°C) in glass bottles.

After 254 days storage, the samples of sugar beet (roots and leaves) were analysed for the residues of Lenacil according to a **modified multiresidue method DFG S19** using **HPLC-MS/MS** (Specht *et al.*, 1995). This method had been sufficiently validated in these matrices according to SANCO/3029/99 with a validated LOQ of 0.02 mg/kg: see ***Vol.3 (CA), B.5.1.2.5.1 (studies no. 1-3)***.

Results and discussion:

Mean lenacil residue levels in sugar beet leaves and roots following freezer storage at <-18˚C for 254 days were 90% and 96%, respectively, of the nominal fortified levels. The results are presented in **Table B.7.1-1**.

Table B.7.1-1: Storage stability of Lenacil residues in sugar beet under frozen conditions at <–18°C

Matrix	Nominal fortification level (mg/kg)	Storage period (days)	Lenacil found (mg/kg)[1]	Lenacil found as a % of fortification level
Leaves	4.0	254	3.58 (3.49, 3.66)	90 (87, 92)
Root	0.2	254	0.191 (0.191, 0.190)	96 (96, 95)

[1] Mean of two samples (individual values between brackets)

Conclusion:

The results indicate that residues of lenacil were stable both in sugar beet leaves and roots for at least 254 days (ca. 8.5 months) following storage at <-18˚C.

Assessment and conclusion by RMS:
Acceptability/Reliability: Yes
Outcome and conclusion of the study: Although the fortified samples were not analysed before storage (time zero) and no procedural recoveries (of freshly spiked control samples) were determined at the time point of analysis of the stored samples, the results are sufficiently reassuring (high recoveries compared to nominal fortification level) and it can be concluded that there was no significant reduction in the residue levels of lenacil in sugar beet leaves or roots following frozen storage (8.5 months at <-18°C).

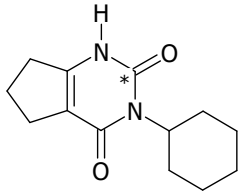
B.7.2. METABOLISM, DISTRIBUTION AND EXPRESSION OF RESIDUES

B.7.2.1. Plants

Report author	Zhang, M. and Glunt, C.D.
Report year	1997
Report title	Metabolism of Lenacil in Sugar Beets
Report No	AMR 3649-95
Guidelines followed in study	Commission Directive 96/68/EC amending Council Directive 91/414/EEC
Major deviations from test guideline	None
Guidance in force at time of submission of supplementary dossier	OECD TG 501 (8 Jan. 2007)
Previous evaluation	Yes, study already evaluated and accepted for the first EU approval review (see DAR – BE, 2007-2009)
GLP	Yes

Material and Methods:

Test substance: ^{14}C-Pyrimidine ring labelled Lenacil (2-^{14}C-DPX-B634)
Structural formula of 2-^{14}C-Lenacil with the asterisk indicating the radiolabeling position:



Specific activity: 8.36 µCi/mg following isotopic dilution with non-radiolabelled Lenacil
Radiochemical purity: > 96 %

Reference standards: Lenacil, IN-G2172 (*Z*-isomer), IN-KD304 (*E*-isomer), IN-KD305 (*E*-isomer), IN-KC939 (*Z/E,* 2:1), IN-KQ961, IN-KC943, IN-KE121, IN-KD302 and IN-KF313.

Preparation of the treatment solution:
The ^{14}C-Lenacil treatment solution was prepared by combining ^{14}C-Lenacil with technical Lenacil and other formulation ingredients to simulate 50 % WP formulation.
A 50-mL treatment solution for the 2 applications was prepared and the final specific activity of the solution was 8.36 µCi/mg. The treatment solution was prepared just before the first application. An aliquot of 20 mL of the treatment solution was used for the first application. The remaining 30 mL treatment solution was stored at –20 °C until the second application. Before each application, an aliquot of the treatment solution was analysed by HPLC and by LSC to confirm the purity and the quantity of the test substance.

Experimental design:
The study was performed under greenhouse conditions.
Sugar beet plants (variety: *HM55 Medium*) were grown in containers (*ca* 40 L capacity) filled with a silt loam soil. Two foliar applications were made at post-emergence stage to the plants, with the first application made at the 4-leaf stage (BBCH 14) and the second application 15 days later at the 6-leaf stage (BBCH 16). The Lenacil test substance was formulated as a 50 % *WP* and applied using a compressed CO_2 sprayer at rates equivalent to 204 and 321 g a.s./ha for the two applications. The total rate applied was 525 g a.s./ha supporting the maximum recommended use rate of 0.5 kg a.s./ha per crop. Untreated plants were grown as controls.
Sugar beet plants (whole) were sampled immediately after the first spray when the treatment solution was dry (0 -day), at 15 -day (immediately after the second treatment solution was dry), and at intervals of 32, 47, 74, 99 and at 130 days (mature stage) after the first treatment. Plants were separated into foliage and roots prior to analysis.

Extraction procedure and analytical procedure:
The total radioactive residues (TRR) in the sugar beet foliage and root samples were quantified by radio combustion analysis and by liquid scintillation counting (LSC) after homogenising in liquid nitrogen.
Samples of foliage and root were extracted using acetonitrile/water (2:1, *v/v*) and analysed by chromatographic comparison in 2 HPLC solvent systems with UV detector against reference standards.
To generate larger amounts of Lenacil metabolites, 7-12 leaf-stage sugar beet leaves were incubated with ^{14}C-Lenacil for 3 to 8 days. LC-MS analysis of the metabolites isolated from excised sugar beet incubation media were confirmed as IN-KC943 and IN-KQ961, respectively.
Metabolite identification was performed mainly on extracts of sugar beet foliage at final harvest (130 days after the first application). The concentrated 130-day foliage extract was further purified for metabolites isolation using semi-preparative HPLC chromatography.
Identification of the glucose conjugates was performed using HPLC analysis of the aglycons following β-glucosidase hydrolysis of the conjugates. After hydrolysis, the control and enzyme-treated samples were analysed by HPLC. The metabolite profile in the 47-day roots extracts and in the 130-day foliage extracts before and after β-glucosidase hydrolysis was analysed by HPLC.
Elucidation of the structure of the metabolites was achieved using mass spectra analysis. Structural confirmation of the metabolites IN-KQ961 and IN-KC943 was based on their LC-MS data in comparison with the synthetic standards. These purified radioactive metabolites together with the synthetic standards were used as references for the metabolite identification in this study.

Results and discussion:

The total radioactive residues (TRR) and the distribution of radioactivity in sugar beet foliage at each sampling interval are given in **Table B.7.2.1-1**. The profile of the extractable radioactivity in sugar beet root is summarised in **Table B.7.2.1-2**.

The TRR in the sugar beet foliage declined steadily from 7.35 mg/kg at 0 day to 0.16 mg/kg in the final harvest at 130 days after the first treatment. In the sugar beet root samples, the level of total residues recovered was low at all sampling intervals, ranging from 0.01 to 0.03 mg/kg from 0 to 99 days after first treatment and <0.01 mg/kg in the mature roots at harvest.

Extractability:

More than 94% of the TRR was extractable from the foliage at each sampling interval. The level of unextractable residues was therefore low at each interval and in the mature foliage this fraction was below 0.01 mg/kg. In the roots, solvent extraction released between 67% and 80% of the TRR at the 47, 74 and 99 day intervals and no residue level above 0.01 mg/kg was recovered as the residual radioactive fraction.

Characterisation/identification of residues in sugar beet foliage:

- Early harvest (0 to 47 days after first treatment):
  The major part of the extracted radioactivity was recovered as the unchanged parent compound (88 – 96 %TRR). Other peaks did not exceed 10 % of TRR in the foliage extracts and no further analysis was performed on these samples.

- Later harvest (74, 99 and 130 days after first treatment):
  Unchanged **parent compound lenacil** was recovered at a level ranging between 28 and 68% of the TRR along with an increasing polar metabolites fraction (see further below). Identified residues in the mature foliage were composed of the 7-hydroxy-lenacil metabolite (IN-KC943) as an unconjugated metabolite at 3.1% of TRR (<0.01 mg/kg) and as glucoside conjugates at a total level of 10.7 % of TRR (<0.02 mg/kg) in the 130-day foliage sample.
  Note: HPLC analysis of the extracts showed a peak with a retention time matching that of metabolite IN-KQ961. However, later results (see further below: "peak 3") indicated that IN-KQ961 showed a similar retention time to that of IN-KC943-glucoside. The peak suspected of corresponding to IN-KQ961 could actually be IN-KC943-glucoside or a mixture of the two. Therefore, the peak was isolated for further β-glucosidase hydrolysis and the resulting peak in the hydrolysate had a retention time matching that of IN-KC943, indicating the existence of IN-KC943 glucose conjugate before hydrolysis with no detectable amount of the metabolite IN-KQ961. The peak was isolated for further hydrolysis but no single metabolite exceeded 10%TRR.

- **Polar metabolites fraction**:
  This fraction (4-8 min. retention time in reversed phase HPLC system) accounted for max. 38% of TRR (0.06 mg eq./kg) and was a mixture of several polar metabolites, some of which could be hydrolysed by β-glucosidase suggesting the existence of glucose conjugates. The polar peak region could be resolved further in another HPLC system, which demonstrated that no single polar metabolite in sugar beet leaves exceeded 10 % TRR or 0.05 mg eq./kg. However, no further structure characterization/identification of these polar metabolites (by retention time matching with reference standards or by Mass Spectrometry) was attempted.

- Glucose conjugates:
  β-glucosidase was used to hydrolyse the 130-day foliage extract for the identification of possible glucose conjugates. This extract contained 3 peaks besides the Lenacil peak:
  (1) Peak 1: This compound was isolated and subjected to enzymatic hydrolysis: see further below.
  (2) Peak 2: This peak was less stable; it disappeared after incubation even in the control sample without *β*-glucosidase. No further effort was made to identify the structure of the metabolite (<10%TRR and <0.05 ppm).
  (3) Peak 3 matched the retention time of IN-KQ961. This compound was isolated and subjected to enzymatic hydrolysis: see here below.

  Both peaks 1 and 3 were isolated by semi-preparative HPLC and subjected to *β*-glucosidase hydrolysis. The hydrolysis showed that both peaks could be hydrolysed by *β*-glucosidase and their degradation compound matched the retention time of IN-KC943. This indicates that **peak 1 and peak 3** contained (mainly) **IN-KC943 glucose conjugates** and no detectable amount of IN-KQ961. The more polar IN-KC943 glucose conjugate (peak 1) might have been a further conjugation on the glucose moiety, but no further structural elucidation was attempted.

HPLC-chromatograms of analysis of 130-day foliage extract (before and after β-glucosidase incubation)

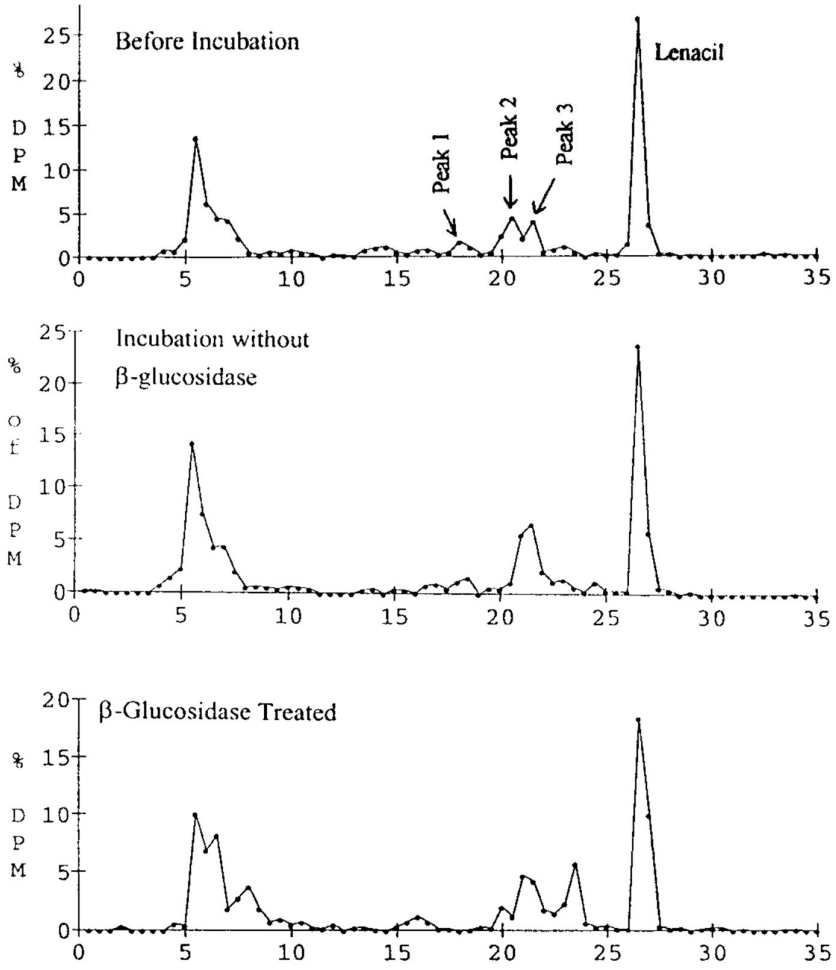


Table B.7.2.1-1: Extractability and investigation of the nature and amounts of residues of Lenacil in sugar beet foliage following 2 foliar spray applications of 2-^{14}C-Pyrimidine labelled Lenacil, respectively at BBCH growth stage 14 and 16, corresponding to application dose rates equivalent to 204 and 321 g a.s./ha, resp.

RAC	Sugar beet foliage						
Harvest day (day after 1st spray)(1)	**0**	**15**	**32**	**47**	**74**	**99**	**130**
Total radioactive residues (TRR) – mg/kg^0							
	7.35	4.71	1.06	1.04	0.69	0.30	0.16
Extractability of TRR – %TRR (mg/kg^0)							
Acetonitrile/water extraction phase	97.4 (7.16)	98.4 (4.63)	99.6 (1.06)	99.5 (1.04)	95.9 (0.66)	95.8 (0.29)	94.4 (0.16)
Elucidation of the radioactive residues – %TRR (mg/kg^0)							
Parent compound (lenacil)	96.0 (7.05)	95.7 (4.51)	88.3 (0.94)	89.9 (0.93)	67.9 (0.47)	52.0 (0.16)	28.4 (0.04)
IN-KC943 (7-hydroxy-Lenacil)	<0.1 (<0.01)	0.3 (0.01)	0.6 (<0.01)	0.3 (<0.01)	<1.0 (<0.01)	1.6 (<0.01)	3.1 (<0.01)
IN-KC943 glucosides(2) *["peak 3"]*	0.5 (0.03)	0.8 (0.04)	4.1 (0.04)	3.6 (0.04)	3.9 (0.03)	5.2 (0.02)	7.7 (0.01)
IN-KC943-glucosyl-conjugate *["peak 1"]*	nd	nd	nd	nd	1.4 (<0.01)	3.6 (0.01)	3.0 (<0.01)
Polar peaks	nd	0.2 (<0.01)	2.1 0.02)	1.5 (0.02)	n.a.	n.a.	n.a.
Glucose conjugates	nd	nd	nd	nd	<0.1 (<0.01)	2.6 (<0.01)	1.6 (<0.01)
Polar metabolites(3)	Nd	0.2 (<0.01)	2.1 (0.02)	1.5 (0.02)	10.5 (0.07)	18.0 (0.05)	37.9 (0.06)
Unknown metabolites	nd	nd	nd	nd	6.1 (0.04)	7.3 (<0.03)	12.7(4) (<0.03)
Total identified metabolites	96.6 (7.09)	96.8 (4.56)	93.0 (0.99)	93.8 (0.98)	74.2 (0.52)	62.4 (0.20)	42.2 (0.07)
Unextracted radioactive residues – %TRR (mg/kg^0)							
	2.6 (0.19)	1.6 (0.08)	0.4 (<0.01)	0.4 (<0.01)	4.1 (0.03)	4.2 (0.01)	5.6 (<0.01)
Accountability: extracted phases + residual radioactive residues (%TRR)							
	100.0	100.0	100.0	99.9	100.0	100.0	100.0

Remarks:
Nd: Not detectable; N.a.: not applicable (e.g. other HPLC system used)
(0) expressed as mg ^{14}C-Lenacil equiv./kg
(1): Harvest at 0 day (immediately after the first spray) and at 15 day (immediately after the second treatment) and at 32, 47, 74 and 99 days after the first treatment and at harvest (130 days after the first treatment).
(2): HPLC analysis showed a peak that matched the retention time of IN-KQ961 (hydroxylated Lenacil), indicating the presence of this metabolite. Later results indicated that IN-KQ961 showed a similar retention time to that of IN-KC943-glucoside and thus, the peak suspected of corresponding to IN-KQ961 could be IN-KC943-glucoside or a mixture of the 2. Therefore, the peak was isolated for further β-glucosidase hydrolysis and the resulting peak in the hydrolysate matched the retention time of IN-KC943, indicating the existence of IN-KC943 glucose conjugate before hydrolysis with no detectable amount of the metabolite IN-KQ961.
(3): This polar fraction was a mixture of several polar metabolites. These peaks were resolved further in another HPLC system and some of the metabolites in this polar area could be hydrolysed by β -glucosidase suggesting the existence of glucose conjugates among these polar metabolites. It was reported that no single polar metabolite in sugar beet leaves exceeded 10 % of the TRR and therefore no structure confirmations were made on these polar Lenacil metabolites.
(4): This fraction was composed of 3 distinct peaks, with a maximum of 7.5 % of TRR (0.012 mg/kg) for *"peak 2"*

The TRR figures were average results from duplicate solvent extraction analyses.

Characterisation/identification of residues in sugar beet root:

The TRR in sugar beet roots were very low (<0.01-0.03 mg/kg). The metabolic profile in root extract was similar to the profile in foliage extract. HPLC analysis of the root showed that no single metabolite in root extracts exceeded 0.01 mg/kg. Therefore, no further characterization/identification of the metabolites was attempted.

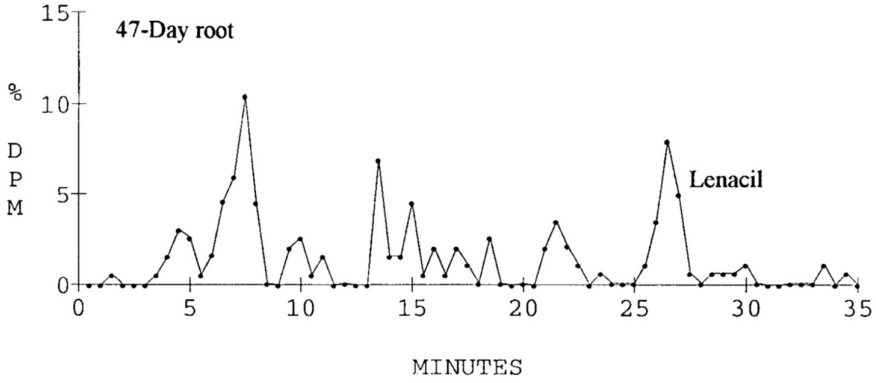


Table B.7.2.1-2: Extractability of the radioactive residues of Lenacil in sugar beet roots following 2 foliar spray applications of 2-^{14}C-Pyrimidine labelled Lenacil, respectively at BBCH growth stage 14 and 16, corresponding to application dose rates equivalent to 204 and 321 g a.s./ha, resp.

RAC	**Sugar beet root**						
Harvest day (day after 1st spray)(1)	**0**	**15**	**32**	**47**	**74**	**99**	**130**
Total radioactive residues (TRR) – mg/kg(0)							
	0.02	0.02	0.02	0.03(2)	0.01(2)	0.03(2)	<0.01
Extractability of TRR – %TRR (mg/kg(0))							
Acetonitrile/water extraction phase	na	na	na	79.2 (0.02)	66.7 (<0.01)	80.0 (0.01)	na
Unextracted radioactive residues – %TRR (mg/kg(0))							
	na	na	na	20.8 (<0.01)	33.3 (<0.01)	20.0 (<0.01)	na
Accountability: extracted phases + residual radioactive residues (%TRR)							
				100.0	100.0	100.0	

Remarks:
Na: not applicable, no solvent extraction analyses were conducted.
(0) expressed as mg ^{14}C-Lenacil equiv./kg
(1): Harvest at 0 day (immediately after the first spray) and at 15 day (immediately after the second treatment) and at 32, 47, 74 and 99 days after the first treatment and at harvest (130 days after the first treatment).
(2): TRR values obtained from solvent extraction respectively for the 47-day, 74-day and 99-day root samples. The other TRR figures resulted directly from radio combustion analysis.

Conclusion:

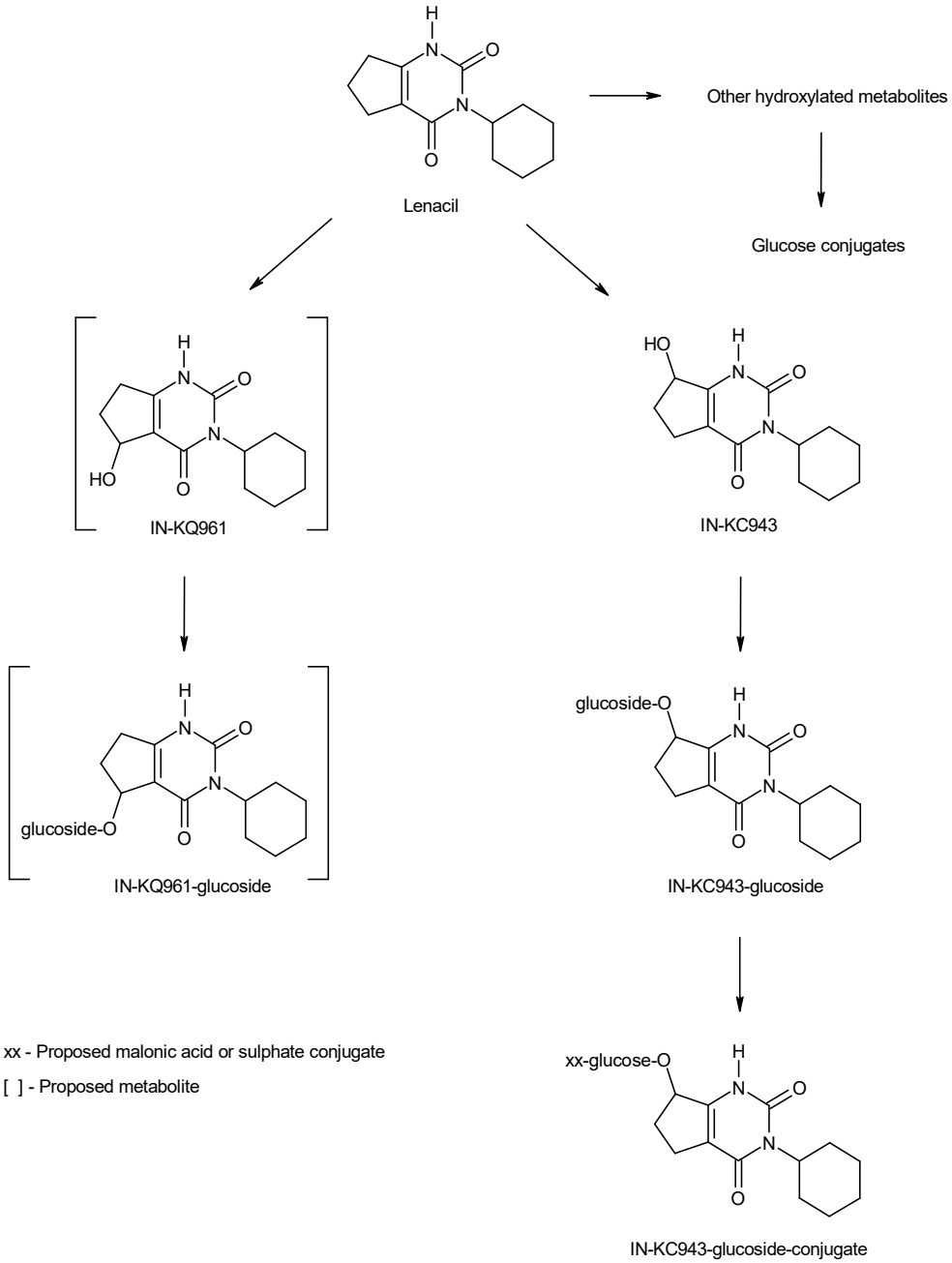
Following 2 foliar applications with lenacil on sugar beet at early growth stage (BBCH 14-16), corresponding to a total application rate of 525 g a.s./ha, the TRR in root samples was max. 0.03 mg/kg (<0.01 mg/kg at mature harvest) and therefore too low for identification of residues. In foliage, parent compound lenacil was the main component of the TRR (88-96% TRR) in the early harvested foliage samples (up to 47 days after first treatment). Other minor compounds were polar metabolites or conjugates. The level of parent lenacil declined to 28%TRR (0.04 mg/kg) in the foliage 130 days after first treatment (i.e. 115 days after the second treatment).
The only identified metabolites were IN-KC943 formed by hydroxylation in position 7 of lenacil (max. 3% of TRR; <0.01 mg/kg) and its glucosides (max. 10.7% of TRR; <0.02 mg/kg). The polar fraction of metabolites, some of which could be hydrolysed by β-glucosidase, accounted for max. 38% of TRR (0.06 mg/kg). As no single polar metabolite exceeded 10% of TRR, no attempts were made to further characterise or identify them.

The following major degradation pathways were observed:

- Hydroxylation of Lenacil on the cyclopentyl ring to generate the metabolite IN-KC943. Although only tentatively identified by chromatographic comparison, the metabolite IN-KQ961 is also possible as another primary hydroxylated product with other more polar compounds.
- Glucose conjugation of those hydroxylated metabolites. 2 types of IN-KC943 conjugates were observed: the polar IN-KC943 glucose conjugate that might have been a further conjugation on the glucose moiety.
  A high ratio of IN-KC943 conjugates/IN-KC943 in the extracts suggested that these hydroxylated products were rapidly transformed to conjugates.

The metabolic pathway of Lenacil in sugar beet leaves is depicted in **Figure B.7.2.1-1**.

Figure B.7.2.1-1: Proposed metabolic pathway of [2-^{14}C]-Lenacil in sugar beet leaves
[Ref.: Zhang & Glunt, 1997]



Assessment and conclusion by RMS:
Acceptability/Reliability: Yes; The study is deemed compliant with OECD guideline 501 (2007). Nevertheless, it is acknowledged that the nature of residues in roots could not be elucidated, due to low total residue levels in root. Therefore, for other uses (on other root crops) that could result in significant residues, additional experiments at exaggerated rates may still be needed to enable metabolite identification.

Outcome and conclusion of the study: see conclusion above

Report author	Chrzanowski, R.L.
Report year	1978
Report title	Metabolism of ^{14}C-lenacil in spinach
Report No	D-2-3
Guidelines followed in study	No information available
Major deviations from test guideline	Not applicable
Guidance in force at time of submission of supplementary dossier	OECD TG 501 (8 Jan. 2007)
Previous evaluation	The study had been previously submitted and mentioned in the context of the assessment of confirmatory data after first EU approval review (see DAR addendum – BE, 2013), but a full assessment had not yet been reported and the studies had not yet been peer reviewed. Full assessment reported in DRAR (BE, 2019) for EU peer review.
GLP	No (pre-GLP)

Material and Methods:

Test substance: [2-^{14}C]- lenacil (radiolabel at 2-position of pyrimidine ring); radiochemical purity: 99%

Application and harvest:
A soil plot, sown with spinach seeds (variety 'Winter Bloomsdale') was treated pre-emergence (immediately after sowing of spinach) in early September 1978, at a rate of approximately 0.6 kg ^{14}C-lenacil/ha (608 g a.s./ha). Mature spinach leaves were harvested 1 to 2 months later (*exact dates not reported*).

Extraction and analytical methodology:
Aliquots of chopped and mixed leaves were analysed for total radioactivity by combustion and liquid scintillation counting (LSC). In addition, spinach leaves were gently rinsed in water for one minute to dislodge surface soil and an aliquot of the rinsing water was analysed for radioactivity (by LSC).

The washed spinach and water rinses were combined in a blender and macerated for 5 minutes with methanol. The mixture was filtered and extracted two more times with methanol/water (2:1, v/v). The methanol-water extracts was evaporated to near dryness (under vacuum at 45°C) and redissolved in water. This aqeous solution was partitioned with hexane (3x) and with ethyl acetate (3x).

Extracts of the same solvent (hexane and ethyl acetate, resp.) were combined for analysis. Aliquots were analysed by LSC and the remainder was evaporated to dryness (under vacuum at 45°C), redissolved in ethyl acetate and analysed by TLC (normal phase: silica gel G-25, ethyl acetate or ethyl acetate/methanol 16:4, v/v)) with autoradiography. ^{14}C-labeled standards of lenacil and several hydroxylated homologs of lenacil *"obtained from rat liver microsome preparation (Belasco, 1976) and soil metabolism studies (Belasco, 1979)"* were used, i.e. IN-G2172, IN-KD304, IN-KD302 and IN-KQ961 (or IN-KC943). Radioactive areas were scraped from the plate and analysed by LSC to quantify radioactivity.

The aqueous extract (after hexane and ethyl acetate partitioning) was evaporated and the residue reconstituted in a buffer solution (pH 5), which was then subjected to enzymatic treatment (mixture of β-glucosidase, cellulase and hesperidinase). After incubation (48 hours at 37°C), water was added and extraction with ethyl acetate was carried out. The ethyl acetate extract was concentrated and analysed by TLC (as above), while the aqueous fraction was divided into two fractions and each analysed for ^{14}C-incorporation into natural products:

- Analysis for polybasic acids and sugars, by means of GC and detection using FID and LSC, according to published method (*Kline et al., 1970*)
- Hydrolysis with 6N HCl under reflux (15 hrs) (pH of hydrolysate <1). The hydrolysate was fractionated – via cation exchange chromatography – into organic acid and base fractions, which were each further analysed for organic acids and amino acids, respectively, using published methods (*Atkins et al., 1971; Fernandez-Flores et al., 1970; Gehrke et al. 1965*)

The initial residual fraction (after methanol/water extraction) was analysis for lignin and cellulose by means of reflux with 10% NaOH and according to procedures described in published literature (*Powell et al., 1925*), respectively.

Results and discussion:

TRR in mature unwashed spinach leaves was 0.22 mg/kg (lenacil eq.), of which about 23% (0.05 mg eq./kg) was associated with surface soil contaminating the leaves (dislodged by rinsing with water).

7% of the TRR was extractable into organic solvent, with an additional 11% TRR becoming soluble after enzyme hydrolysis (of the polar ^{14}C material in the water soluble fraction).

Only a trace of lenacil (<1%TRR; <0.002 mg/kg) was found. All of the remaining organo-soluble ^{14}C-residues co-chromatographed with standards of ring hydroxylated homologs of lenacil, or unidentified oxidation products of lenacil prepared by rat liver microsomal metabolism. In total, those metabolites were claimed to account for about 18%TRR (0.04 ppm). See **Table B.7.2.1-3**.

A large and distributed polar fraction (63%TRR; 0.14 mg eq./kg) was observed, but could not be characterised or identified. The unextractable residue accounted for 19% of the TRR (0.04 mg eq./kg). However, there were no indications of incorporation of radioactivity into biomolecules (sugars, amino acids, polybasic acids, organic acids, lignin and cellulose). Therefore, the soluble polar and insoluble bound residues were presumed to be *"additional conjugates and further oxidation products of ring hydroxylated lenacil homologs"*.

Table B.7.2.1-3: Nature and amount of residues in spinach leaves following pre-emergence treatment with ^{14}C-lenacil (Chrzanowski, 1978)

Compound	%TRR	mg/kg (lenacil equivalent)
Lenacil	<1	<0.002
IN-KQ961 or IN-KC943 [a]	1.1	0.002
IN-G2172 and IN-KD304 [a]	3.6	0.008
	7.1	0.016
Unidentified lenacil oxidation products from liver microsomal study (2 closely eluting components) [b]	5.9	0.013
IN-KD302	<1	<0.002
Water soluble (polar) fraction	63	0.14
Unextracted	19	0.04
Total	**100**	**0.22**

[a] identity/stereochemistry of reference standards not clearly assigned
[b] *"suspected to be cis/trans lenacil isomers hydroxylated in both rings, but unconfirmed"*

Conclusions:

A soil plot, sown with spinach seeds was immediately treated (pre-emergence) with ^{14}C-lenacil at 0.6 kg/ha. Mature spinach leaves were harvested. TRR was 0.22 mg/kg. Only traces of parent lenacil (<1%TRR; <0.002 mg/kg) were present. Hydroxylated lenacil metabolites, conjugates of those and further oxidation products were detected. A large fraction of water soluble polar residues (63%TRR, 0.04 mg eq./kg) could not be further characterised or identified. As there were no indications of incorporation of radioactivity into biomolecules, the author suggested that the polar residues may represent additional conjugates and further oxidation products of hydroxylated lenacil metabolites.

Assessment and conclusion by applicant:
"The spinach study LENA/RES 1, originally submitted as confirmatory data after Annex I inclusion and conducted with test material [2-^{14}C]lenacil, is a non-GLP study without information on the guidelines used. Information (e.g. application details) and results reported are rather limited, and mainly TLC was used for separation and tentative identification of compounds in the extracts. The study does not meet current guidelines, and no reliable conclusion can be derived from this study."

Assessment and conclusion by RMS:
Acceptability/Reliability: Not acceptable as guideline-compliant study; only as supportive information (with limited reliability).
The study was conducted at an experimental station of E.I. DuPont de Nemours & Co. (i.e. the producer at that time) and was not conducted according to GLP principles (pre-GLP). The information and results included in the study report are not sufficiently detailed and/or are incomplete (e.g. no chromatograms were included, no method description was included for the published methods). Furthermore, the identification of the 4 metabolites was only tentative, as it was based on retention time matching with reference standards on TLC (and because the stereochemical configuration was not clearly defined for all the reference standards).

Outcome and conclusion of the study:
Although the reliability of the reported results cannot be fully verified on the basis of the limited information presented in the study report, the main findings seem to indicate that, in the case of pre-emergence soil treatment immediately after sowing, the parent compound lenacil is only a minor/negligible fraction of the total residues (<1%TRR) in mature spinach leaves. After treatment at 600 g/ha, no significant levels of parent lenacil in spinach are to be expected (<0.01 mg/kg). The limited characterization of metabolites indicated the presence of several ring-hydroxylated homologs of lenacil, which are at least partially conjugated.

Report author	Chrzanowski, R.L.
Report year	1978
Report title	Metabolism of ^{14}C-lenacil in strawberries
Report No	LLME-3-78
Guidelines followed in study	No information available
Major deviations from test guideline	Not applicable
Guidance in force at time of submission of supplementary dossier	OECD TG 501 (8 Jan. 2007)
Previous evaluation	The study had been previously submitted and mentioned in the context of the assessment of confirmatory data after first EU approval review (see DAR addendum – BE, 2013), but a full assessment had not yet been reported and the studies had not yet been peer reviewed. Full assessment reported in DRAR (BE, 2019) for EU peer review.
GLP	No (pre-GLP)

Material and Methods:

Test substance: [2-^{14}C]- lenacil (radiolabel at 2-position of pyrimidine ring); radiochemical purity: 99%

Application and harvest:
Soil in which young strawberry plants (variety 'Guardian') were growing, was treated with ^{14}C- lenacil/ha at an application rate of 1.630 kg a.s./ha. The application was made uniformly from a sprayer to the test area. One of the two plots received 2 applications, while the other plot received only 1 application (see **Table B.7.2.1-4** below). *Crop growth stages at application were not reported.* Mature strawberry fruits were harvested about 2 months after the last application (*exact dates not reported*).

Table B.7.2.1-4: Timing/rate of application of ^{14}C-lenacil to 2 plots of strawberry plants (Chrzanowski, 1978)

	September 1977	**April 1978**	**Early June 1978**
Plot 1	(1) 1.630 kg/ha	(2) 1.630 kg/ha	Harvest
Plot 2	-	(1) 1.630 kg/ha	Harvest

Extraction and analytical methodology:
Samples of strawberries were freeze dried and aliquots were analysed for total radioactivity by combustion and liquid scintillation counting (LSC). In addition, strawberries were rinsed in water for one minute to dislodge surface residues and an aliquot of the rinsing water was analysed for radioactivity (by LSC).

The washed strawberries and water rinses were combined in a blender and macerated for 5 minutes with methanol. The mixture was filtered and extracted two more times with methanol/water (2:1, v/v). The methanol-water extracts was evaporated to near dryness (under vacuum at 45°C) and redissolved in water. This aqeous solution was partitioned with hexane (3x) and with ethyl acetate (3x).

Extracts of the same solvent (hexane and ethyl acetate, resp.) were combined for analysis. Aliquots were analysed by LSC and the remainder was evaporated to dryness (under vacuum at 45°C), redissolved in ethyl acetate and analysed by TLC (normal phase: silica gel G-25, ethyl acetate or ethyl acetate/methanol 16:4, v/v)) with radiography. ^{14}C-labeled standards of lenacil and several hydroxylated homologs of lenacil *"from rat liver microsome preparation"* were used *(no defined metabolite structure was provided in the structure; only 2 metabolite 'types', with hydroxyl-substitution on the cylohexyl ring or on the cyclopentapyrimidine ring).* Radioactive areas were scraped from the plate and analysed by LSC to quantify radioactivity.

The aqueous extract (after hexane and ethyl acetate partitioning) was evaporated and the residue reconstituted in a buffer solution (pH 5.5), which was then subjected to enzymatic treatment (mixture of β-glucosidase, cellulase and hesperidinase). After incubation (48 hours at 37°C), extraction with ethyl acetate was carried out. The ethyl acetate extract was concentrated and analysed by TLC (as above), while the aqueous fraction was combined with

the original residual solid fraction and subjected to hydrolysis using 6N HCl under reflux (15 hrs) (pH of hydrolysate <1). The hydrolysate was fractionated – via cation exchange chromatography – into organic acid, neutral and base fractions. The neutral fraction was further analysed for (reducing) sugars by reaction with phenylhydrazine (hydrochloride) and quantification (by combustion-LSC) of the radioactive phenylosazone derivatives formed.

Results and discussion:

TRR in mature strawberry fruits was only 0.06 mg/kg (lenacil eq.) and no difference was found in the TRR level between the strawberries that received a double treatment (autumn + spring) versus a single treatment (spring only). Only 2% of the TRR was associated with surface soil contaminating the fruits (dislodged by rinsing with water).

19% of the TRR was extractable into organic solvent, with an additional 7% TRR becoming soluble after enzyme hydrolysis (of the polar ^{14}C material in the water soluble fraction).

Only a trace of lenacil (0.3%TRR; <0.001 mg/kg) was found. It was stated that the majority of the remaining organo-soluble ^{14}C-residue co-chromatographed with standards of ring hydroxylated homologs of lenacil. Some of them were conjugated, as revealed by enzymatic hydrolysis. Together, these ring hydroxylated lenacil metabolites (free and conjugated) accounted for about 25% of the TRR (0.014 mg eq./kg). However, further confirmatory work was not done due to the very low levels of each compound (≤0.01 ppm). The remaining water soluble polar and bound residues were fractionated after hydrolysis into organic base (amino acids), organic acid, sugar and neutral fractions. See **Table B.7.2.1-5**.

Table B.7.2.1-5: Nature and amount of residues in strawberry fruits following (soil) treatment with ^{14}C-lenacil (Chrzanowski, 1978)

Compound	%TRR	mg/kg (lenacil equivalent)
Lenacil	0.3	<0.001
Ring hydroxylated lenacils (free and conjugated) [a]	25	0.014
Polar organic bases (amino acids)	6.4	0.004
Polar organic acids	5.3	0.003
Sugars	3.1	0.002
Polar neutrals	22	0.012
Non-hydrolysable insoluble fraction	38	0.022
Total	**100**	**0.057**

[a] identity of reference standards not clearly assigned

Conclusions:

Soil in which young strawberry plants were growing, received a single or double treatment with ^{14}C-lenacil at 1.630 kg/ha. TRR in mature strawberries was about 0.06 mg/kg. Only traces of parent lenacil (0.3%TRR; <0.001 mg/kg) were present. Hydroxylated lenacil metabolites, conjugates of those were tentatively detected (25%TRR, 0.014 ppm). A large fraction of water soluble polar residues (37%TRR, 0.021 ppm) was characterized, but could not be further identified. As there were some indications of incorporation of radioactivity into biomolecules (e.g. sugars, amino acids), the author suggested that the polar residues may reflect incorporation of ^{14}C into natural plant constituents.

Assessment and conclusion by applicant:
"The strawberry study LLME-3-78, originally submitted as confirmatory data after Annex I inclusion and conducted with test material [2-^{14}C]lenacil, is a non-GLP study without information on the guidelines used. Information (e.g. application details) and results reported are rather limited, and mainly TLC was used for separation and tentative identification of compounds in the extracts. The study does not meet current guidelines, and no reliable conclusion can be derived from this study."

Assessment and conclusion by RMS:
Acceptability/Reliability: Not acceptable as guideline-compliant study; only as supportive information (with limited reliability).
The study was conducted at an experimental station of E.I. DuPont de Nemours & Co. (i.e. the producer at that time) and was not conducted according to GLP principles (pre-GLP). The information and results included in the study report are not sufficiently detailed and/or are incomplete (e.g. insufficient details on method and timing of application, no chromatograms were included etc.). Furthermore, the assignment of the metabolites as 'hydroxylated lenacils' was very general and certainly only tentative, as it was apparently based on retention time matching with reference standards on TLC (and because the stereochemical configuration was not clearly defined for the reference standards).

Outcome and conclusion of the study:
Although the reliability of the reported results cannot be fully verified on the basis of the limited information presented in the study report, the main findings seem to indicate that, in the case of post-emergence (soil) treatment, the parent compound lenacil is only a minor/negligible fraction of the total residues (<1%TRR) in mature strawberry fruits. After treatment at about 1.6 kg/ha and a PHI of about 2 months, no significant levels of parent lenacil in strawberry fruit are to be expected (<0.01 mg/kg). The limited characterization of metabolites suggested the presence of several ring-hydroxylated homologs of lenacil, which are at least partially conjugated.

B.7.2.2. Poultry

No data were provided.

B.7.2.3. Lactating ruminants

No data were provided.

B.7.2.4. Pigs

No data were provided.

B.7.2.5. Fish

No data were provided.

B.7.3. MAGNITUDE OF RESIDUE TRIALS IN PLANTS

Report author	Tillkes, M.
Report year	1998
Report title	Magnitude of Residue of Lenacil and Triflusulfuron methyl in Sugar Beet grown in France following application of Venzar and DPX-MX843-1 – Season 1995
Report No	F-95-001-RES
Guidelines followed in study	Not reported
Major deviations from test guideline	Not reported
Guidance in force at time of submission of supplementary dossier	OECD TG 509 (7 Sept. 2009); OECD GD on crop field trials (ENV/JM/MONO(2011)50/rev1 – 7 Sept. 2016)
Previous evaluation	DAR (BE, 2007-2009)
GLP	Yes

Material and methods:

In the growing season of 1995, 3 supervised residue field trials on sugar beet were conducted at different locations in Northern France. The test substances used in the field residue trials were as follows:
- VENZAR (WP, Wettable powder) containing 80 % (w/w) Lenacil
- Blend of VENZAR WP (containing 61.5 % (w/w) of Lenacil) with DPX-MX843-1 (a formulation containing 11.5 % of Triflusulfuron methyl, also referred to as SAFARI (WG)).

The experimental design of the residue trials performed in Northern France can be described as follows:
Treatment 1: A pre-emergence application of VENZAR 80 WP at a rate of 0.760-0.806 kg a.s./ha.
Treatment 2: Blend of VEZAR 80 (WP) and SAFARI (WG) referred as DPX-MX843-1 was applied twice with last application at BBCH 14 or BBCH 19 (respectively 4 or 8-10 leaf stage) at a total dose rate of 0.528-0.546 kg a.s./ha. The interval between applications ranged from 7 to 12 days.
Treatment 3: Blend of VENZAR 80 (*WP*) and SAFARI (WG) referred as DPX-MX843-1 was applied 3 or 4 times with the final application at BBCH 14 or BBCH 17/18 (respectively 4 or 7/8 leaf stage) at a total dose rate of 0.314 to 0.407 kg a.s./ha. The interval between applications ranged from 6 to 12 days.

The samples were stored frozen for approximately 26 months prior to extraction and analysis.

The samples of sugar beet (roots and leaves) were analysed for the residues of Lenacil according to **modified DFG S19 multi-residue method** (with modified extraction) using **GC-MS**. An LOQ of 0.01 mg/kg was claimed for this method. However, insufficient validation data were available to consider the method suitable (according to SANCO/3029/99): see ***Vol.3 (CA), B.5.1.2.5.1 (study no. 4)***.

Results and discussion:

No residues at or above the claimed LOQ (0.01 mg/kg) were found, neither in the control samples, nor in the treated samples. Results for the treated samples are summarised in **Table B.7.3.1-1** (see further below).
Detailed residue results per trial are shown in the residue data summary sheets presented in **Appendix B.7.9.2.**

Assessment and conclusion by RMS:
Acceptability/Reliability: No. The frozen storage period of the samples prior to analysis (26 months) is not supported by storage stability data. Furthermore, the analytical method used was insufficiently validated. Therefore, results of these trials cannot be fully relied upon.

Outcome and conclusion of the study: Following 2 foliar spray applications to sugar beets (0.528-0.546 kg a.s./ha) at relatively early crop growth stages (BBCH 14-19) in 3 trials in Northern France, residues of lenacil were reported to be below the LOQ (<0.02 mg/kg) in sugar beet root and tops at mature harvest (113-140 DAT). Nevertheless, results cannot be fully relied upon, due to the lack of method validation and storage stability data (see above).

Report author	Pollmann, B. → incl. analytical final report (Mende P., 2002)
Report year	2002
Report title	Generation of Samples for the Determination of Residues of Venzar 80 % WP (containing 80% Lenacil) in Sugar Beets, five sites in Europe, 2001
Report No	20011048/E1-FPSB
Guidelines followed in study	EU Commission Working Document "Guidelines for the generation of data concerning residues as provided in Annex II, part A, section 6 and Annex III, part A, section 8 of Directive 91/414/EEC, concerning the placing of plant protection products on the market" – 1607/VI/97 rev. 1; BBA (1990); IVA (1992)
Major deviations from test guideline	Not reported
Guidance in force at time of submission of supplementary dossier	OECD TG 509 (7 Sept. 2009); OECD GD on crop field trials (ENV/JM/MONO(2011)50/rev1 – 7 Sept. 2016)
Previous evaluation	DAR (BE, 2007-2009)
GLP	Yes

Material and methods:

In the growing season of 2001, 4 supervised residue field trials on sugar beet were conducted at different locations in Germany.
One spray application of VENZAR 80 WP (80% w/w lenacil) was performed at a dose rate ranging between 0.502 and 0.539 kg a.s./ha and at BBCH growth stage 37 (leaves covering 70 % of ground).
Sugar beet roots and leaves with tops were sampled at maturity, which was about 70-90 days after treatment. In one of the trials (G01N005R), samples of leaves, tops and roots were taken at different growth stages (PHIs) up to harvest, to investigate decline of residue levels.
The samples were stored frozen for 5 months prior to extraction and analysis.

The samples of sugar beet (roots and leaves) were analysed for the residues of Lenacil according to a **modified DFG S19 multi-residue method** using **HPLC-MS/MS** (Specht *et al.*, 1995). This method had been sufficiently validated in these matrices according to SANCO/3029/99 with a validated LOQ of 0.02 mg/kg: see ***Vol.3 (CA), B.5.1.2.5.1 (studies no. 1-3)***.

Results and discussion:

No results above the LOQ were detected in the control samples. Results for the treated samples are summarised in **Table B.7.3.1-1** (see further below).
Detailed residue results per trial are shown in the residue data summary sheets presented in **Appendix B.7.9.2.**
Underlined values were selected for deriving MRL and/or STMR values.

Assessment and conclusion by RMS:
Acceptability/Reliability: yes

Outcome and conclusion of the study: Following a foliar spray application to sugar beets (0.502-0.539 kg a.s./ha; BBCH growth stage 37) in 4 trials in Germany, residues of lenacil were below the LOQ (<0.02 mg/kg) in sugar beet root at mature harvest (70-90 DAT), while in tops at harvest residues up to 0.04 mg/kg were found.

Report author	Pollmann, B. → incl. analytical report (Hamberger R., 2002)
Report year	2002
Report title	Generation of Samples for the Determination of Residues of Venzar 80 % WP (containing 80% Lenacil) in Sugar Beets, one site in Europe, 2002
Report No	20011048/E2-FPSB
Guidelines followed in study	EU Commission Working Document "Guidelines for the generation of data concerning residues as provided in Annex II, part A, section 6 and Annex III, part A, section 8 of Directive 91/414/EEC, concerning the placing of plant protection products on the market" – 1607/VI/97 rev. 1; BBA (1990); IVA (1992)
Major deviations from test guideline	Not reported
Guidance in force at time of submission of supplementary dossier	OECD TG 509 (7 Sept. 2009); OECD GD on crop field trials (ENV/JM/MONO(2011)50/rev1 – 7 Sept. 2016)
Previous evaluation	DAR (BE, 2007-2009)
GLP	Yes

Material and methods:

In the growing season of 2002, 1 supervised residue field trial (P02N001R) on sugar beet was conducted in Portugal.
One spray application of VENZAR 80 WP (80% w/w lenacil) was performed at a dose rate of 0.520 kg a.s./ha and at BBCH growth stage 38 (leaves covering 80 % of ground).
Sugar beet roots and leaves with tops were sampled at maturity, which was 60 days after treatment.
The samples were stored frozen for 1 month prior to extraction and analysis.

The samples of sugar beet (roots and leaves) were analysed for the residues of Lenacil according to a **modified DFG S19 multi-residue method** using **HPLC-MS/MS** (Specht *et al.*, 1995). This method had been sufficiently validated in these matrices according to SANCO/3029/99 with a validated LOQ of 0.02 mg/kg: see ***Vol.3 (CA), B.5.1.2.5.1 (studies no. 1-3)***.

Results and discussion:

No results above the LOQ were detected in the control samples. Results for the treated samples are summarised in **Table B.7.3.1-2** (see further below). Detailed residue results per trial are shown in the residue data summary sheets presented in **Appendix B.7.9.2.** Underlined values were selected for deriving MRL and/or STMR values.

Assessment and conclusion by RMS:
Acceptability/Reliability: yes

Outcome and conclusion of the study: Following a foliar spray application to sugar beets (0.520 kg a.s./ha; BBCH growth stage 38) in 1 trial in Portugal, residues of lenacil were below the LOQ (<0.02 mg/kg) in sugar beet root at mature harvest, while 0.03 mg/kg was found in tops at harvest (60 DAT).

Report author	Anderson, I. & Kakkonen, J.E. → incl. analytical phase report (Witte, A., 2006)
Report year	2006
Report title	Decline of Lenacil Residues in Sugar Beet (Root and Tuber Vegetables) Following a Single Application of Venzar® 80WP (Lenacil) – Southern Europe, Season 2005
Report No	688479 → analytical phase report no: 20051414/01-RSB
Guidelines followed in study	EU Commission Working Document "Guidelines for the generation of data concerning residues as provided in Annex II, part A, section 6 and Annex III, part A, section 8 of Directive 91/414/EEC, concerning the placing of plant protection products on the market" – 1607/VI/97 rev. 2; Appendix B "General Recommendations for the Design, Preparation and Realization of Residue Trials" (7029/VI/95 Rev. 5- 22-Jul-1997)
Major deviations from test guideline	Not reported
Guidance in force at time of submission of supplementary dossier	OECD TG 509 (7 Sept. 2009); OECD GD on crop field trials (ENV/JM/MONO(2011)50/rev1 – 7 Sept. 2016)
Previous evaluation	DAR (BE, 2007-2009)
GLP	Yes

Material and methods:

In the growing season of 2005, 2 supervised residue field trials on sugar beet were conducted at different locations in Spain.
One spray application of VENZAR 80 WP (80% w/w lenacil) was performed at a dose rate ranging from 0.503 to 0.510 kg a.s./ha and at BBCH growth stages 31-32 (beginning of crop cover – leaves covering 10% of ground).
Sugar beet roots and leaves with tops were sampled at maturity, which was 106-120 days after treatment.
In one of the trials, samples of leaves were also taken at several earlier growth stages (shorter PHIs) up to harvest, to investigate decline of residue levels.

The samples were stored frozen for 2.5 months prior to extraction and analysis.

The samples of sugar beet (roots and leaves) were analysed for the residues of Lenacil by means of HPLC-MS/MS, according to a **modified version of the multi-residue method published by Fillion *et al.* (2000)**. This method had been sufficiently validated in these matrices according to SANCO/3029/99 with a validated LOQ of 0.02 mg/kg: see ***Vol.3 (CA), B.5.1.2.5.1 (study no. 5)***.

Results and discussion:

No results above the LOQ were detected in the control samples. Results for the treated samples are summarised in **Table B.7.3.1-2** (see further below). Detailed residue results per trial are shown in the residue data summary sheets presented in **Appendix B.7.9.2**. Underlined values were selected for deriving MRL and/or STMR values.

Assessment and conclusion by RMS:
Acceptability/Reliability: yes

Outcome and conclusion of the study: Following a foliar spray application to sugar beets (0.503-0.510 kg a.s./ha; BBCH growth stages 31-32) in 2 trials in Spain, residues of lenacil were below the LOQ (<0.02 mg/kg) in sugar beet roots and tops at mature harvest (106 – 120 DAT).

Table B.7.3.1-1: Lenacil residues in sugar beet in Northern EU

Location; Year; Trial	Application				Portion analysed	PHI (days)	Residue (mg/kg)	Ref.
	Formulation (type and content of a.s.)	No.; crop stage at final application	kg a.s./ha	kg a.s./hL				
N.France; 1995; Soudron, 51 F-95-001-RES	WP; 800 g/kg	1; pre-emergence	0.806	0.445	roots tops	168 168	< 0.01 < 0.01	(Tillkes M., 1998)
	WP; 800 g/kg*	2; 4 leaves (BBCH GS 14)	0.257 0.271	0.146 0.144	roots tops	129 129	< 0.01 < 0.01	
	WP; 800 g/kg*	4; 7 leaves (BBCH GS 17)	0.084 0.081 0.072 0.077	0.044 0.044 0.044 0.044	roots tops	121 121	< 0.01 < 0.01	
N.France; 1995; Rupéreux, 77 F-95-001-RES	WP; 800 g/kg	1; pre-emergence	0.794	0.358	roots tops	174 174	< 0.01 < 0.01	
	WP; 800 g/kg*	2; 4 leaves (BBCH GS 14)	0.270 0.276	0.116 0.128	roots tops	140 140	< 0.01 < 0.01	
	WP; 800 g/kg*	4; 7/8 leaves (BBCH GS 17/18)	0.078 0.083 0.075 0.085	0.036 0.036 0.036 0.040	roots tops	131 131	< 0.01 < 0.01	
N.France; 1995; Epagny F-95-001-RES	WP; 800 g/kg	1; pre-emergence	0.760	0.400	roots tops	161 161	< 0.01 < 0.01	
	WP; 800 g/kg*	2; 8-10 leaves (BBCH GS 19)	0.271 0.259	0.134 0.134	roots tops	113 113	< 0.01 < 0.01	
	WP; 800 g/kg*	3; 4 leaves (BBCH GS 14)	0.084 0.078 0.245	0.040 0.040 0.134	roots tops	130 130	< 0.01 < 0.01	
Germany; 2001; Niedersachsen G01N003R	WP; 800 g/kg	1: BBCH 37 (covering 70% ground)	0.539	0.166	roots tops	70 70	**< 0.02** **< 0.02**	(Pollmann B., 2002a); (Mende P., 2002)
Germany; 2001; Niedersachsen G01N004R	WP; 800 g/kg	1: BBCH 37 (covering 70% ground)	0.526	0.167	roots tops	70 70	**< 0.02** **0.04**	

Germany; 2001; Würtemberg G01N005R	WP; 800 g/kg	1: BBCH 37 (covering 70% ground)	0.502	0.167	leaves leaves leaves roots tops roots tops	0 15 28 42 42 77 77	3.8 1.4 0.09 < 0.02 0.04 **< 0.02** **< 0.02**	
Germany; 2001; Rheinland-Pfalz G01N006R	WP; 800 g/kg	1: BBCH 37 (covering 70% ground)	0.514	0.167	roots tops	90 90	**< 0.02** **< 0.02**	

Remarks:
-* Applied as a physical blend with DPX-MX843-1
- Residues of Lenacil in untreated roots and tops were less than the LOQ (< 0.01 or < 0.02 mg/kg) at all sites.
- Tops = leaves plus top (crown) of the root.

Table B.7.3.1-2: Lenacil residues in sugar beet in Southern EU

Location; Year; Trial	Application				Portion analysed	PHI (days)	Residue (mg/kg)	Ref.
	Formulation (type and content of a.s.)	No.; crop stage at final application	kg a.s./ha	kg a.s./hL				
Portugal; 2002; Vila Franca P02N001R	WP; 800 g/kg	1: BBCH 38 (leaves covering 80% of ground)	0.520	0.125	roots tops	60 60	**< 0.02** **0.03**	(Pollmann B., 2002b); (Hamberger R., 2002)
Spain; 2005; Burgos 688479	WP; 800 g/kg	1: BBCH 31 (beginning of crop cover)	0.503	0.252	leaves leaves leaves leaves roots tops	0 14 28 42 120 120	19 0.55 0.03 < 0.02 **< 0.02** **< 0.02**	(Anderson I. and Kakkonen J.E., 2006)
Spain; 2005; Araba 688479	WP; 800 g/kg	1: BBCH 31-32 (beginning of crop cover)	0.510	0.251	roots tops	106 106	**< 0.02** **< 0.02**	

Remarks:
-Residues of Lenacil in untreated roots and tops were less than the LOQ (< 0.01 or < 0.02 mg/kg) at all sites.
-Tops = leaves plus top (crown) of the root.

Publicly available([1]) scientific literature:

Report author	Kucharski, M.; Sadowski, J.; Wujek, B.; Trajdos, J.
Report year	2011
Report title	Influence of adjuvants addition on lenacil residues in plant and soil
Source	Polish Journal of Agronomy (2011), Volume 5, pp. 39–42
Guidelines followed in study	Not reported
Previous evaluation	No; considered for the purpose of renewal
GLP	No

Abstract:

<< The aim of this work was to evaluate the influence of an adjuvant addition on lenacil residues in soil and roots of sugar beet. Field experiments were conducted during a three-year-period from 2008 until 2010 on arable fi elds located in southwestern Poland. Chemical weed control in sugar beet was carried by commercial formulation of lenacil. Herbicide was applied alone (recommended and reduced doses) and in mixture with adjuvants (oil, surfactant and multicomponent). Lenacil residue was analysed using HPLC/UV. At lifting time the residues of lenacil in soil amounted to 0.0006–0.0042 mg kg-1. In sugar beet roots samples, the residues of lenacil were lower than in soil and amounted to <0.0005–0.0020 mg kg-1. The addition of adjuvants caused an increase of the active substance residues in soil and roots of sugar beet in comparison with the treatments, where lenacil was used without adjuvant (reduced dose). The increase of the lenacil residues was statistically signifi cant for most of soil and plant samples and amounted average to 45 and 41% respectively. Influence of single adjuvant on lenacil residues in soil and plant was different for each year (experimental season). The residues of lenacil determined in roots of sugar beet did not exceed acceptable value (MLR). >>

Material and methods:

Field experiments were conducted during a three-year period from 2008 until 2010 on arable fields located in southwestern Poland (black soil, pH = 6.1–6.5, organic carbon content 2.04–2.13% and clay content 45–52%). The field trial was set up as a randomized complete block design with four replicates. All farming activities were carried out in accordance with conventional agricultural practice and in line with recommendations from officials. Chemical weed control in sugar beet was carried by commercial formulation of lenacil (Venzar 80 WP, DuPont de Nemours) at the doses 800 and 600 g of active substance per ha (recommended and reduced doses respectively). Herbicide was applied alone (in both doses) and in mixture (reduced dose) with three different adjuvants (A):

1) A1: Atplus 60 EC (paraffin oil) in the rate 1.5 L/ha – oil adjuvant;
2) A2: Break Thru S 240 (polymethylsiloxane copolymer) in the rate 0.3 L/ha – surfactant adjuvant;
3) A3: BackRow (multicomponent adjuvant) – in the rate 0.3 L/ha.

BackRow adjuvant based on a blend of non-ionic surfactants, emulsifiers, sticking agents and specialist oils has been specifically designed to optimize coverage and desorption of pre-emergence herbicides onto the soil surface (producer's information).

Herbicide and its mixtures were applied pre-emergency after sugar beet sowing (in the third decade of April). The effect of herbicide and adjuvant application on residues in plant and soil profile was studied. Samples of soil and roots of sugar beet were taken at the day of lifting (in the first decade of October – 159–166 days after treatment). The samples were taken from the middle of each plot to avoid interference and side effects from the neighboring plots. The soil samples were taken at the top soil layer (0–15 cm of depth). In years 2008–2010 weather conditions, especially rainfalls, from April to July (for 3 months after herbicide treatment), the most deviated from average conditions in this region of Poland (recorded from 1961 to 2000); (Table 1).

Table 1. Average rainfalls recorded in southwest Poland.

Year	Average rainfall [mm]			
	April	May	June	July
2008	55.1	40.7	29.3	44.2
2009	24.7	65.7	180.8	145.1
2010	50.6	136.8	49.4	116.8
(1961–2000)	37.6	61.3	71.4	80.0

[1] http://www.iung.pulawy.pl/PJA/wydane/5/PJA56Kucharski.pdf

Samples taken from experiments were well mixed and stored in polyethylene bags at minus 20°C until sample extraction. The analytical procedure for the determination of lenacil consisted of three elementary processes:

- extraction of analyzed substance from matrix (using Extractor DIONEX ASE 350, extraction solvent – methanol);
- cleaning of extract using SPE (Solid Phase Extraction) column with C18 active solid;
- final determination using HPLC-UV (reversed phase C18 stationary column; mobile phase: acetonitrile/water/methanol 50/40/10 (% v/v/v); detection at UV wavelength 230 nm).

The recovery of the lenacil was determined by fortification of soil samples at concentrations of 0.001, 0.01, 0.1 and 1.0 mg/kg in three replicates. The average recovery for all concentrations was 96% for soil and 92% for plant.
"This method was based on procedure described in Polish Standard (PN-R-04121, 1997)."
"All experimental data were calculated using the statistical program Statgraphics Centurion, version XV."

Results and discussion:

Results obtained from all experiments are shown in Table 2. At lifting time, in soil samples taken from plots, the residues of lenacil amounted to 0.0006–0.0042 mg/kg. In sugar beet roots samples, the residues of lenacil were lower than in soil and amounted to <0.0005–0.0020 mg/kg.
The level of residues was dependent on the dosage of substance, addition of adjuvant and weather condition in individual vegetation seasons.

- The residues level was strongly affected by rainfalls occurring after herbicide application; In 2008, the average rainfalls from April to July amounted to 169.3 mm and were lower than for long-term observations (250.3 mm); (Tab. 1). The two next years were wetter – the average rainfalls amounted to 416.3 and 353.6 mm respectively. An increase of rainfalls influence the leaching herbicide into soil profile (substance moves to deeper soil layer) and residues detected in the top soil layer and plants are lower (Cuevas et al., 2007). This effect was more evident when intensive rainfalls occurred at first weeks after treatment (year 2010).
- The addition of adjuvants caused an increase of the active substance residues in soil and roots of sugar beet in comparison with the treatments, where lenacil was used without adjuvant (reduced dose). The increase of the lenacil residues was statistically significant for most of soil and plant samples and amounted average to 45 and 41% respectively. Influence of single adjuvant on lenacil residues in soil and plant was different for each year (experimental season).

Influence of adjuvants on herbicide residues in soil and plant, degradation rate and leaching depend on the kind of adjuvant (Kucharski and Sadowski, 2009). The effect of organic additives, especially oil substances, on increase of herbicide retention, mobility and immobilization in soil top layer was described by other authors (Koskinen et al., 2006; Todoruk and Langford, 2006; Kaushik and Neera, 2007; Cao et al., 2008). The addition of adjuvants could influence speed of degradation and increase herbicide residues in soil and plant, but usually adjuvants are applied with herbicides in reduced doses (70–80% of recommended) and herbicidal residues determined at harvest time are lower than those obtained from treatments, where full (recommended) doses of herbicide (without adjuvant) were applied (Kucharski, 2003).

Table 2. Residues of lenacil in soil and roots of sugar beet.

Object	Residues# [mg kg^{-1}]					
	2008		2009		2010	
	soil	root	soil	root	soil	root
Lenacil (FD)	0.0042	0.0020	0.0026	0.0011	0.0015	0.0006
Lenacil (RD)	0.0023	0.0009	0.0012	0.0006	0.0006	ND
Lenacil (RD) + A1	0.0034	0.0015	0.0015	0.0009	0.0011	0.0005
Lenacil (RD) + A2	0.0031	0.0012	0.0020	0.0007	0.0009	ND
Lenacil (RD) + A3	0.0033	0.0014	0.0019	0.0008	0.0008	ND
LSD(0.05)	0.00072	0.00028	0.00067	0.00012	0.00021	-

average residues for 4 replications
FD – full (recommended) dose; RD – reduced dose
A1 – oil adjuvant; A2 – surfactant adjuvant, A3 – multicomponent adjuvant
ND – residue did not detect (<0.0005 mg kg^{-1})

Assessment and conclusion by RMS:
Acceptability/Reliability: Not acceptable as guideline-compliant study; only as supportive information (with limited reliability).

Outcome and conclusion of the study:
In this study, the influence of adjuvant addition (oil, surfactant, and multicomponent) on lenacil residues in soil and roots of sugar beet was investigated. The addition of adjuvants caused an apparent increase of the active substance residues in soil and roots of sugar beet in comparison with the treatments where lenacil was used without adjuvant. Although the reported average recovery values for the analytical method used in this study suggest an adequate accuracy of the analytical method, details on the validation data package (per fortification level) were not reported. Anyhow, reported residue levels of lenacil were clearly below 0.01 mg/kg in all root and soil samples. However, it is to be noted that neither the timing of application, nor the number of applications was clearly reported in the publication. Besides the addition of adjuvants, also the weather conditions (especially rainfall) appeared to have an influence on the residue levels in the top soil layer and roots of sugar beet.

B.7.4. FEEDING STUDIES

B.7.4.1. Poultry

No data were provided.

B.7.4.2. Ruminants

No data were provided.

B.7.4.3. Pigs

No data were provided.

B.7.4.4. Fish

No data were provided.

B.7.5. EFFECTS OF PROCESSING

Following data waiver was provided by the applicant (Doc. M-CA, section 6 – rev. 1 Oct. 2016):

<< According to Commission Regulation (EU) No 283/2013 processing studies are not necessary "if no significant (>0.1 mg/kg) or no analytically quantifiable residues occur in the plant product being processed". No quantifiable lenacil residues (<0.02 mg/kg) were found in sugar beet roots (the commodity that is processed) following applications according to the supported GAP and the theoretical maximum daily intake (TMDI) is less than 10% of the ADI (see Point CA 6.9 in this document). Therefore, no studies on the effects of processing are necessary. >>

Assessment and conclusion by RMS:
The data waiver as presented by the applicant is not fully acceptable. The citation of the legal data requirement by the applicant is not correct. As a matter of fact, Reg. (EU) No 283/2013 stipulates *that "Studies on the nature of residues in processing shall be provided where residues in products of plant or animal origin subject to processing may occur at a level of or higher than 0.01 mg/kg".*

Due to the fact that the validated LOQ of the analytical methods used in the supervised residue trials on sugar beet was 0.02 mg/kg, results in sugar beet root were reported as <0.02 mg/kg. However, chromatograms provided in the supervised residue trial study reports indicated that the actual residue levels were considerably lower than 0.02 mg/kg. Furthermore, it is to be noted that several residue trials were performed at later crop growth stage (BBCH 37-38) compared to BBCH 31 (cGAP), which represents a more critical residue situation and nevertheless, residues were < 0.02 mg/kg.

Therefore, the occurrence of residue levels of lenacil at or above 0.01 mg/kg in sugar beet roots is considered unlikely. This is also supported by the metabolism study on sugar beet, which showed that the TRR in roots was <0.01 mg/kg (parent eq.) 130 days after treatment (*vide supra – **B.7.2.1***).

Further RMS's considerations and conclusions: see ***Vol.1, 2.7.6***.

B.7.5.1. Nature of the residue

No data were provided.

B.7.5.2. Distribution of the residue in peel and pulp

No data were provided.

B.7.5.3. Magnitude of residues in processed commodities

No data were provided.

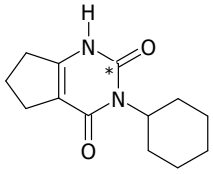
B.7.6. RESIDUES IN SUCCEEDING OR ROTATIONAL CROPS

B.7.6.1. Metabolism in rotational crops

Report author	Hurst, L.
Report year	2013
Report title	[^{14}C]-Lenacil: Uptake and Metabolism in Confined Rotational Crops
Report No	8240837
Guidelines followed in study	OECD Guideline 502: Metabolism in Rotational Crops (January 2007)
Major deviations from test guideline	None
Guidance in force at time of submission of supplementary dossier	OECD Guideline 502: Metabolism in Rotational Crops (January 2007)
Differences between old and current guideline	Not applicable
Previous evaluation	Addendum to DAR (BE, 2013) (confirmatory data after first EU approval review)
GLP	Yes

Materials and methods:

A single treatment of [^{14}C]-lenacil (radiolabel at 2-position of pyrimidine ring; specific radioactivity 0.658 MBq/mg, purity >97%) in a Venzar 80 WP formulation was applied to bare sandy loam soil (Boughton Loam, UK) at a rate equivalent to 485 g a.i./ha.



Following **application**, the top 5 cm of soil was mixed and spring wheat (variety *Tybalt*), spinach[2] (variety *Scenic F1 hybrid*) and turnips (variety *Rapa Bianca*) were sown at intervals of 30 or 68, 182 and 365 days after treatment (DAT) and grown in a controlled greenhouse environment. The selected crops represented small cereal grain, leafy vegetable and root crop groups.

For each plant back interval the following **crop samples** were harvested: immature spinach leaves (BBCH 32-39), mature spinach leaves (BBCH 49), turnip roots/leaves (BBCH 45-49), wheat forage (BBCH 21-30), wheat hay (BBCH 41-49), wheat straw and grain (BBCH 89). Samples were homogenized in presence of dry ice prior to analysis.

Soil samples (20 cm depth) were removed from separate but equivalent treated soil at an interval of 30, 60, 90, 120, 180 and 292 DAT. At 182 DAT soil samples were additionally taken to 5 cm depth.

Total radioactive residues (TRR) were determined in each crop fraction and soil sample on each sampling occasion by liquid scintillation counting (LSC) after combustion. Crop samples were sequentially extracted with acetonitrile, acetonitrile:water (1:1 v/v), water, dilute acid, dilute base and acetone. The acetonitrile and acetonitrile:water (1:1 v/v) extracts were combined, cleaned up using Solid Phase Extraction (SPE) prior to analysis. Soil samples were extracted with acetonitrile and acetonitrile:water (3:1 v/v) and the extracts concentrated for analysis. **Extract analysis** was conducted using HPLC (Zorbax RX-C8, 250 x 4.6 mm, 5µm; gradient elution with mixtures of 0.01 M (aq.) ammonium actetate (pH 5) and acetonitrile) with UV detection (wavelength 254 nm) and fraction collection followed by LSC analysis. Selected extracts were additionally analysed by HPLC-MS (+ESI; high resolution full scan MS) in view of structural elucidation. Reference standards of following known or proposed lenacil degradation products were available for comparison: IN-KE121, IN-KF313, IN-KQ957, IN-KD304, IN-KD302, IN-KC943 and IN-KQ961 (structural formulas: see Table B.7.9-5).

Unextracted residues – if ≥10% TRR or ≥0.05 mg/kg – were subjected to additional treatments, i.e. incubation with sodium acetate buffer (0.1 M, pH 5) and enzyme hydrolysis (using Driselase®, i.e. preparation containing carbohydrate active enzymes), as well as harsh acid reflux (6M HCl), to achieve further extraction.

[2] Originally, lettuce (variety *Iceberg*) was planted at 30 DAT, however significant phytotoxic effects were observed and all plants were removed and spinach was planted as an alternative crop at 68 DAT.

Enzymatic hydrolysis was also performed on selected solvent extracts to investigate the presence of conjugated metabolites.

Results and discussion

Total radioactive residue (TRR) levels in the different crop and soil samples are summarised in **Table B.7.6.1-1**. TRR levels in crop fractions were lower at later plant-back intervals, except for immature spinach. Significant radioactive residues (i.e. TRR ≥ 0.01 mg/kg) remained in all samples at all plant back intervals. TRR was generally higher in animal feed items (up to 6.1 mg/kg; in 30 DAT wheat straw sample) compared to human food items (0.01 – 0.26 mg/kg lenacil equivalent). TRR in turnip leaves was higher than in turnip roots, indicating a significant translocation of residues from roots into upper plant parts.

In soil, no significant decline of the TRR level was observed over time (30 – 292 DAT).

Table B.7.6.1-1 Total Radioactive Residues (TRRs) in crop samples and soil samples

Matrix	*Plant-back interval (days)*	*TRR (ppm) [pyrimidine ring-2-^{14}C]*	*Plant-back interval (days)*	*TRR (ppm) [pyrimidine ring-2-^{14}C]*	*Plant-back interval (days)*	*TRR (ppm) [pyrimidine ring-2-^{14}C]*
Immature spinach	68 DAT	0.072	182 DAT	0.119	365 DAT	0.071
Mature spinach	68 DAT	0.259	182 DAT	0.173	365 DAT	0.074
Turnip leaves (tops)	30 DAT	0.687	182 DAT	0.117	365 DAT	0.077
Turnip roots	30 DAT	0.115	182 DAT	0.018	365 DAT	0.012
Wheat forage	30 DAT	0.694	182 DAT	0.508	365 DAT	0.162
Wheat hay	30 DAT	1.101	182 DAT	0.255	365 DAT	0.142
Wheat straw	30 DAT	6.129	182 DAT	1.622	365 DAT	0.639
Wheat grain	30 DAT	0.143	182 DAT	0.065	365 DAT	0.020
Soil[3]	30 DAT	0.224	60 DAT	0.301	-	-
Soil[3]	90 DAT	0.260	120 DAT	0.181	-	-
Soil[3]	182 DAT	0.218[4]	292 DAT	0.244	-	-

[3] Soil core samples to 20 cm depth
[4] TRR in short core sample (5 cm depth) was 0.327 mg/kg, indicating that a larger proportion of the residues remained in the top 5 cm of the soil.

Extractability of crop residues

Generally, radioactive residues were extracted to a great extent by neutral organic solvent extraction using acetonitrile. The combined fractions from acetonitrile and acetonitrile/water extraction represented extractable residues in the range of 86 – 97% TRR in most **crop** samples. Notable exceptions were however *wheat straw*, where residues were rather released by more polar solvent extraction (acetonitrile/water and water) and *wheat grain*, where much lower extractability was observed (33 – 68% TRR using acetonitrile(/water)).

Dilute acid extraction and dilute base extractions released each ≤5% TRR, with the notable exception of wheat grain from the 182 DAT plant-back, where 21% TRR (0.014 mg/kg lenacil equivalent) was released into dilute alkali.

The distribution of the residues among the fractions generated by the solvent extraction procedure is summarised in **Table B.7.6.1-2, Table B.7.6.1-3** and **Table B.7.6.1-4**. An acceptable accountability was achieved (procedural recoveries relative to TRR from combustion analysis were in range of 79.2 – 125.1%).

Overall, the extraction profile was very similar at each plant-back interval and also consistent between crops (except for wheat straw and wheat grain as mentioned above).

After solvent extractions, only low levels of **unextracted radiolabel** (<10% TRR and <0.05 mg/kg) remained in each crop fraction, except for wheat grain and wheat straw. Incubation with pH 5 buffer and enzymatic hydrolysis of the unextracted residues contained in wheat grain and wheat straw (30 DAT sample) released only an additional 3 – 4 % of the TRR. However, a remarkable further 27% TRR was released from wheat grain following 6N acid reflux. In wheat straw, only a further 1% TRR was released following acid reflux. The remaining unextracted radioactive residues after the whole extraction procedure accounted for approximately 6% TRR (0.3 mg/kg) in wheat straw and 19% TRR (0.03 mg/kg) in wheat grain.
These results confirm that a proportion of the unextracted residue is highly bound and most probably is intrinsically associated with the structure of the plant (i.e. incorporated into natural products, such as lignin as a major component of the straw and starch as a major component of the grain).

Table B.7.6.1-2 Distribution and characterisation of radioactive residues in rotational crop matrices when dosed with ^{14}C-labeled lenacil (30 DAT plant-back; 68 DAT for spinach)

Fraction	*Immature spinach*		*Mature spinach*		*Turnip leaf*		*Turnip root*		*Wheat forage*		*Wheat hay*		*Wheat straw*		*Wheat grain*	
	%TRR	*ppm*	*%TRR*	*ppm*	*%TRR*	*ppm*	*%TRR*	*ppm*	*%TRR*	*ppm*	*%TRR*	*ppm*	*%TRR*	*ppm*	*%TRR*	*ppm*
Extracted residue	**93.7**	**0.067**	**97.0**	**0.251**	**98.8**	**0.679**	**96.7**	**0.111**	**97.8**	**0.679**	**97.9**	**1.078**	**81.1**	**4.969**	**53.7**	**0.077**
Acetonitrile	71.9	0.052	70.3	0.182	82.0	0.564	73.2	0.084	79.6	0.553	75.3	0.829	9.6	0.590	2.6	0.004
Acetonitrile:water (1:1 v/v)	13.7	0.010	22.6	0.058	14.7	0.101	15.6	0.018	15.8	0.110	19.3	0.213	48.1	2.946	40.4	0.058
Water	3.5	0.003	0.8	0.002	1.0	0.007	1.2	0.001	0.9	0.006	1.5	0.016	15.8	0.967	4.3	0.006
0.1M HCl	1.4	0.001	0.3	0.001	0.3	0.002	1.5	0.002	0.3	0.002	0.3	0.004	3.0	0.183	2.2	0.003
0.1M NaOH	3.0	0.002	2.5	0.006	0.7	0.005	4.9	0.006	0.8	0.006	1.1	0.012	3.4	0.206	3.1	0.004
Acetone	0.2	ND	0.5	0.001	0.1	0.001	0.3	ND	0.4	0.003	0.4	0.004	1.2	0.076	1.2	0.002
Unextracted residue	**6.3**	**0.005**	**3.0**	**0.008**	**1.2**	**0.008**	**3.3**	**0.004**	**2.2**	**0.015**	**2.1**	**0.023**	**18.9**	**1.160**	**46.3**	**0.066**

ND: not detected (<0.001 mg/kg); NA: not applicable

Grey shaded cells: extract fractions used for further HPLC analysis (containing >10%TRR and/or >0.01 mg/kg)

Table B.7.6.1-3 Distribution and characterisation of radioactive residues in rotational crop matrices when dosed with ^{14}C-labeled lenacil (182 DAT plant-back)

Fraction	*Immature spinach*		*Mature spinach*		*Turnip leaf*		*Turnip root*		*Wheat forage*		*Wheat hay*		*Wheat straw*		*Wheat grain*	
	%TRR	*ppm*	*%TRR*	*ppm*	*%TRR*	*ppm*	*%TRR*	*ppm*	*%TRR*	*ppm*	*%TRR*	*ppm*	*%TRR*	*ppm*	*%TRR*	*ppm*
Extracted residue	**98.4**	**0.117**	**98.1**	**0.170**	**97.8**	**0.114**	**95.9**	**0.017**	**97.3**	**0.494**	**97.4**	**0.248**	**93.6**	**1.518**	**61.7**	**0.040**
Acetonitrile	83.5	0.099	74.8	0.129	80.5	0.094	69.5	0.013	64.9	0.330	72.7	0.186	8.2	0.132	2.2	0.001
Acetonitrile:water (1:1 v/v)	12.6	0.015	19.4	0.034	12.5	0.015	18.6	0.003	27.2	0.138	20.5	0.052	66.6	1.080	30.4	0.020
Water	0.3	ND	0.2	ND	1.9	0.002	3.3	0.001	3.4	0.017	2.4	0.006	14.4	0.234	2.6	0.002
0.1M HCl	0.3	ND	0.3	0.001	0.7	0.001	1.1	ND	0.5	0.003	0.3	0.001	2.5	0.040	4.4	0.003
0.1M NaOH	1.3	0.001	0.5	0.001	1.0	0.001	0.5	ND	0.8	0.004	1.1	0.003	1.1	0.018	20.9	0.014
Acetone	0.4	ND	2.9	0.005	1.2	0.001	2.8	0.001	0.4	0.002	0.5	0.001	0.8	0.014	1.2	0.001
Unextracted residue	**1.6**	**0.002**	**1.9**	**0.003**	**2.2**	**0.003**	**4.1**	**0.001**	**2.7**	**0.014**	**2.6**	**0.007**	**6.4**	**0.104**	**38.3**	**0.025**

ND: not detected (<0.001 mg/kg); NA: not applicable

Grey shaded cells: extract fractions used for further HPLC analysis (containing >10%TRR and/or >0.01 mg/kg)

Table B.7.6.1-4 Distribution and characterisation of radioactive residues in rotational crop matrices when dosed with ^{14}C-labeled lenacil (365 DAT plant-back)

Fraction	*Immature spinach*		*Mature spinach*		*Turnip leaf*		*Turnip root*		*Wheat forage*		*Wheat hay*		*Wheat straw*		*Wheat grain*	
	%TRR	*ppm*	*%TRR*	*ppm*	*%TRR*	*ppm*	*%TRR*	*ppm*	*%TRR*	*ppm*	*%TRR*	*ppm*	*%TRR*	*ppm*	*%TRR*	*ppm*
Extracted residue	**96.2**	**0.068**	**96.2**	**0.071**	**95.8**	**0.074**	**93.6**	**0.011**	**95.7**	**0.155**	**94.3**	**0.134**	**91.2**	**0.583**	**72.8**	**0.015**
Acetonitrile	71.5	0.051	76.7	0.057	81.3	0.062	69.4	0.008	68.9	0.111	61.7	0.087	14.1	0.090	4.1	0.001
Acetonitrile:water (1:1 v/v)	23.3	0.017	18.8	0.014	12.5	0.010	20.8	0.003	24.5	0.040	28.9	0.041	61.0	0.389	63.5	0.013
Water	0.4	ND	0.2	ND	1.4	0.001	1.2	ND	2.1	0.003	3.2	0.005	14.9	0.095	0.7	ND
0.1M HCl	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA
0.1M NaOH	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA
Acetone	1.0	0.001	0.5	ND	0.6	ND	2.3	ND	0.2	ND	0.5	0.001	1.2	0.008	4.5	0.001
Unextracted residue	**3.8**	**0.003**	**3.8**	**0.003**	**4.2**	**0.003**	**6.4**	**0.001**	**4.3**	**0.007**	**5.7**	**0.008**	**8.8**	**0.056**	**27.2**	**0.005**

ND: not detected (<0.001 mg/kg); NA: not applicable

Grey shaded cells: extract fractions used for further HPLC analysis (containing >10%TRR and/or >0.01 mg/kg)

Characterisation and identification of extractable crop residues

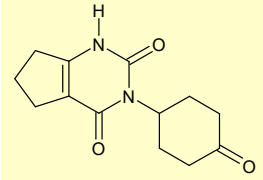
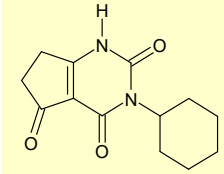
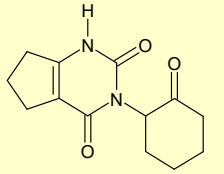
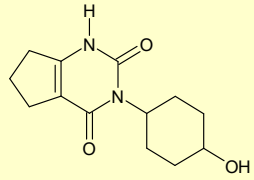
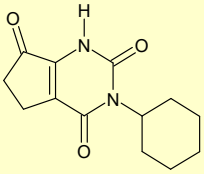
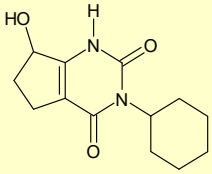
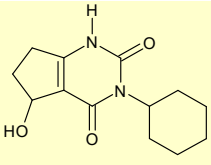
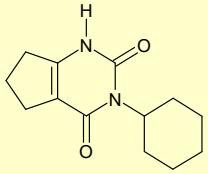
Crop extracts containing significant levels of radioactive residues (i.e. >10%TRR and/or >0.01 mg/kg) (see shaded cells in the **Table B.7.6.1-2**, **Table B.7.6.1-3** and **Table B.7.6.1-4**) were combined and further analysed by HPLC. Results are presented in **Table B.7.6.1-7, Table B.7.6.1-8** and **Table B.7.6.1-9**.

Parent compound lenacil was present at very low levels in immature and mature spinach (up to 1% TRR; 0.003 mg/kg). In turnip leaves and roots, lenacil accounted for 3 to 10% of the TRR (max. 0.011 mg/kg in turnip root), with lower levels observed at 182 and 365 DAT compared to 30 DAT. In the 30 DAT wheat samples, lenacil was present in forage and hay at 31% TRR (0.22 mg/kg) and 25% TRR (0.28 mg/kg), respectively, and in straw at 1% TRR (0.075 mg/kg). The amount of lenacil present in the corresponding 182 DAT and 365 DAT samples was lower with 9 to 15% and 2 to 12% TRR in the forage and hay, respectively, and 0.4 to 0.5% of the TRR in the straw. Lenacil was only detected at a very low level in grain (0.5% TRR) and only in the 30 DAT samples.

In all crop extracts from each plant-back interval, a very complex residue profile was observed with a **large number of unknown metabolites** present. HPLC elution profiles could be roughly divided in a rather polar compound region (retention time < 10 min; CR1, CR10, CR14, CR7, CR12) and a region with retention time mainly between 10 and 20 minutes.

Although the available 7 reference standards of known lenacil metabolites (see **Table B.7.6.1-5**) eluted almost all – very closely – also in that latter region (10-20 minutes), the presence of the known metabolites could not be confirmed by co-chromatography with reference standards, due to the very close elution of peaks and insufficient peak resolution (possibly due to UV absorbing matrix components and to the low radioactivity levels requiring off-line LSC detection following fraction collection).

Table B.7.6.1-5: Structural formulas of reference standards used

IN-KE121	IN-KF313	IN-KQ957	IN-KD304
			
IN-KD302	**IN-KC943**	**IN-KQ961**	**Lenacil**
			

However, based on mass accuracy and peak retention time data obtained by HPLC-MS analysis of isolated metabolite fractions, a **tentative characterization/identification** could be derived for the following unknown metabolite regions: see **Table B.7.6.1-6** below. No LC-MS/MS fragmentation data could be obtained due to the low MS response of all compounds present in each isolate.

Table B.7.6.1-6: Tentative characterisation/identification of selected metabolite regions		
Metabolite region analysed by HPLC-MS	***Tentative characterization/identification***	***Remarks***
CR7	**hydroxylated analogue of lenacil** *(other than IN-KD304, IN-KQ961 or IN-KC943)*	m/z 251 *[lenacil –H +OH]*
CR12	**hydroxylated analogue of lenacil** *(other than IN-KD304, IN-KQ961 or IN-KC943)*	m/z 251 *[lenacil –H +OH]*
CR11	**IN-KD304** *(other peaks present could not be characterized)*	m/z 251 *[lenacil –H +OH]*
CR16	**IN-KQ961** *(main peak)*	MS signal at m/z 233 *[lenacil -2H]* – suggesting an oxidized analogue of lenacil – was observed for isolate CR16 as well as for KQ961 reference standard, while molecular mass of KQ961 is actually 251 u. It is therefore assumed by the RMS that KQ961 is readily fragmented/dehydrated (*[-H_2O]*) during MS ionization.
CR1, CR2, CR5	*no characterization possible*	No test compound related MS signals were detected.

Furthermore, enzymatic hydrolysis of the 30 DAT turnip leaf extract – which contained a considerable amount of polar radioactivity (CR01 at 27.7% TRR; 0.192 mg/kg) – produced a significant change in the HPLC profile of the extractable residue, with retention times matching those of reference standards of different hydroxylated or oxidised lenacil analogues (IN-KD304, IN-KE121, IN-KQ957, IN-KQ961 and IN-KC943). IN-KQ961 was the only known metabolite comprising > 10% TRR (12% TRR; 0.083 mg/kg)[5]. Unconjugated forms of the metabolites IN-KE121, IN-KQ957 and IN-KC943 may form part of the metabolite regions CR2 to CR6, CR8, CR9 and CR13, which chromatographically are similar to hydroxylated metabolite standards, but are not fully identified as such. Based on these results, it can be concluded that the polar material has a **multi-component** nature and that polar radioactivity CR01, and potentially other unknown compounds (incl. less polar radioactivity regions), correspond to **(glucose)[6] conjugated forms of known hydroxylated lenacil metabolites**.

Definitive identification of all metabolite regions appeared to be a difficult analytical challenge, mainly due to co-extracted material, low sensitivity of compound structures to ionisation for MS and the very close elution by HPLC. The latter suggests very close structural similarity of the different metabolites and thus supports the proposed metabolic pathway of different positional hydroxylations on either the cyclohexyl or cyclopentyl ring, or both rings (see further: **Figure B.7.6.1-1**).

[5] However, RMS notes that this is more or less the same as the level of CR16 – tentatively identified as KQ961 – observed in 30 DAT turnip leaf extract before enzyme treatment (i.e. 11.6% TRR; 0.080 mg/kg).
[6] The use of carbohydrate active enzymes in the experiment suggests that glucose is at least one of the conjugating moieties.

In **spinach** extracts, polar radioactivity (CR1 and CR14) accounted for 21 to 40% of the TRR (with slightly higher %TRR levels towards later plant-back). The remaining extractable residue was multi-component comprising up to 14 radioactive regions, only one of which (CR7 – characterised as hydroxylated lenacil) accounted for >10% of the TRR (up to 16 % TRR) in the mature and immature crop at each plant back interval, with a max. level of 0.038 mg/kg.

In **turnip** extracts, polar radioactivity (CR1, CR10 and CR14) accounted for 28 to 30% of the TRR in the 30 and 365 DAT leaf samples (lower amount of 6% TRR was observed in the 182 DAT leaves) and 11 to 37% of the TRR in the 30, 182 and 365 DAT root samples (proportion of polar compounds higher with later plant-back). The remaining extractable residue was multi-component comprising up to 14 radioactive regions, of which CR2, CR4, CR5, CR11 (identified as KD304) and CR16 (identified as KQ961) accounted for ≥10% of the TRR in leaves. In roots, CR2 and CR6 accounted for >10% of the TRR; however only at 30 DAT above 0.01 mg/kg (max. 0.028 mg/kg (24% TRR) of CR2 and 0.021 mg/kg (18%TRR) of CR6).

Also in extracts of **wheat** fractions a large number of metabolites was observed at each plant back interval. In the forage, CR5, as well as the polar region CR1 at 365 DAT, accounted for >10% TRR, whilst in hay CR2, CR5 and CR7 (hydroxylated lenacil) and the polar region CR1 accounted for >10% TRR. In straw the metabolite regions CR2, CR5, CR6, CR7, CR12 and the polar region CR1 accounted for >10% TRR. In grain only the polar region CR1 accounted for >10% TRR (max. of 0.025 mg/kg at 30 DAT). The different extraction profile observed in the wheat grain might result from a greater translocation of the radioactivity taken up and consequently more extensively transformed/conjugated radioactive residues.

Table B.7.6.1-7: Amount (%TRR and mg/kg), characterisation and identification of radioactive residues in crop extracts from the 30/68 DAT plant-back interval

Sample	DAT	Lenacil	CR1	CR10	CR14	CR7	CR12	CR11	CR16	CR2	CR3	CR4	CR5	CR6	CR8	CR9	CR13
Proposed identification		**Lenacil**	**Polar Metabolites ((glucose) conjugates)**			**Hydroxylated lenacil**		**KD304**	**KQ961**	**$CH_3CN/CH_3CN/H_2O$ soluble and chromatographically similar to other hydroxylated (and oxidised) lenacil metabolites e.g. KE121, KQ957, KF313,KC943**							
Immature spinach	%TRR	ND	**16.1**	ND	5.2	10.2	6.3	ND	5.4	5.6	6.0	ND	7.0	6.7	1.9	3.3	7.6
	mg/kg	ND	**0.011**	ND	0.004	0.007	0.004	ND	0.004	0.004	0.004	ND	0.005	0.005	0.001	0.002	0.005
Mature spinach	%TRR	1.2	**23.8**	ND	1.9	**14.9**	**8.6**	1.0	2.9	**4.8**	3.1	2.7	**6.1**	2.3	4.1	4.0	**7.7**
	mg/kg	0.003	**0.061**	ND	0.005	**0.038**	**0.022**	0.003	0.007	**0.012**	0.008	0.007	**0.016**	0.006	0.010	0.010	**0.020**
Turnip leaves	%TRR	6.5	**27.7**	ND	ND	3.1	ND	12.7	11.6	9.9	ND	9.5	3.0	ND	0.9	5.5	ND
	mg/kg	0.045	**0.192**	ND	ND	0.021	ND	0.088	0.080	0.069	ND	0.066	0.021	ND	0.006	0.038	ND
Turnip roots	%TRR	**9.6**	1.9	6.3	3.3	1.0	1.2	5.7	ND	**24.1**	ND	ND	4.2	**18.1**	ND	ND	ND
	mg/kg	**0.011**	0.002	0.007	0.004	0.001	0.001	0.007	ND	**0.028**	ND	ND	0.005	**0.021**	ND	ND	ND
Wheat forage	%TRR	31.3	6.9	ND	ND	2.5	ND	ND	ND	7.9	8.6	3.8	24.1	ND	ND	ND	3.0
	mg/kg	0.217	0.048	ND	ND	0.017	ND	ND	ND	0.055	0.060	0.026	0.168	ND	ND	ND	0.021
Wheat hay	%TRR	25.2	6.7	ND	ND	ND	2.7	ND	ND	12.9	ND	5.1	17.9	9.7	3.5	ND	ND
	mg/kg	0.283	0.075	ND	ND	ND	0.030	ND	ND	0.145	ND	0.057	0.200	0.109	0.039	ND	ND
Wheat straw	%TRR	1.4	**11.9**	ND	ND	**5.1**	**3.0**	ND	ND	**11.6**	ND	**2.5**	**12.7**	**10.5**	**4.0**	**3.2**	**3.6**
	mg/kg	0.075	**0.656**	ND	ND	**0.282**	**0.166**	ND	ND	**0.641**	ND	**0.140**	**0.702**	**0.580**	**0.223**	**0.176**	**0.198**
Wheat grain	%TRR	0.5	**17.5**	ND	2.8	0.7	ND	1.7	2.7	ND	ND	ND	0.5	1.3	2.4	0.7	1.0
	mg/kg	0.001	**0.025**	ND	0.004	0.001	ND	0.002	0.004	ND	ND	ND	0.001	0.002	0.003	0.001	0.001

ND = Not detected (<0.001 mg/kg).
Spinach crop planted 68 DAT, turnip and wheat crops plants 30 DAT
Minor components and background radioactivity were also present in each sample and collectively this accounted for between 4% and 13% of the residue in the crop samples.
Shaded metabolites indicate where (tentative) identification/characterisation was possible using HPLC chromatography and mass spectral data.
Metabolites in **bold** are those that exceed 0.01 mg/kg/0.1 mg/kg in plant commodities relevant for human/livestock, resp. These metabolites are characterised but currently are not fully identified.

Table B.7.6.1-8: Amount (%TRR and mg/kg), characterisation and identification of radioactive residues in crop extracts from the 182 DAT plant-back interval

Sample	DAT	Lenacil	CR1	CR10	CR14	CR7	CR12	CR11	CR16	CR2	CR3	CR4	CR5	CR6	CR8	CR9	CR13	CR15
Proposed identification		**Lenacil**	**Polar Metabolites ((glucose) conjugates)**			**Hydroxylated lenacil**		**KD304**	**KQ961**	**$CH_3CN/CH_3CN/H_2O$ soluble and chromatographically similar to other hydroxylated (and oxidised) lenacil metabolites e.g. KE121, KQ957, KF313, KC943**								
Immature spinach	%TRR	ND	**22.9**	ND	ND	**15.0**	3.6	2.8	5.2	4.5	3.8	6.9	7.3	3.8	2.2	1.9	**9.3**	ND
	mg/kg	ND	**0.026**	ND	ND	**0.017**	0.004	0.003	0.006	0.005	0.004	0.008	0.008	0.004	0.003	0.002	**0.011**	ND
Mature spinach	%TRR	ND	**35.3**	ND	ND	**14.0**	**6.6**	1.1	3.0	3.6	3.7	**6.7**	5.0	1.4	ND	ND	**9.1**	1.8
	mg/kg	ND	**0.060**	ND	ND	**0.024**	**0.011**	0.002	0.005	0.006	0.006	**0.011**	0.009	0.002	ND	ND	**0.015**	0.003
Turnip leaves	%TRR	3.1	4.5	ND	1.2	7.5	4.3	ND	3.8	7.9	ND	9.7	12.2	6.1	0.6	9.0	4.4	5.4
	mg/kg	0.004	0.005	ND	0.001	0.009	0.005	ND	0.004	0.009	ND	0.012	0.014	0.007[a]	0.001	0.011	0.005	0.006
Turnip roots	%TRR	3.0	19.9	ND	9.4	5.7	8.1	ND	7.0	3.6	1.1	4.7	1.9	3.1	ND	2.6	6.2	3.9
	mg/kg	0.001	0.004	ND	0.002	0.001	0.001	ND	0.001	0.001	ND	0.001	ND	0.001	ND	ND	0.001	0.001
Wheat forage	%TRR	15.1	4.4	ND	ND	9.9	3.0	ND	3.2	8.0	5.3	9.4	14.0	2.3	2.9	3.0	3.5	1.5
	mg/kg	0.078	0.022	ND	ND	0.051	0.015	ND	0.017	0.041	0.027	0.048	0.072	0.012	0.015	0.015	0.018	0.007
Wheat hay	%TRR	11.7	3.7	ND	0.9	12.6	1.2	ND	4.3	8.5	4.0	9.9	13.3	2.1	3.1	2.4	3.0	1.9
	mg/kg	0.030	0.010	ND	0.002	0.032	0.003	ND	0.011	0.022	0.010	0.025	0.034	0.005	0.008	0.006	0.008	0.005
Wheat straw	%TRR	0.4	**17.0**	ND	0.6	4.0	**11.8**	6.1	3.6	5.6	3.0	3.9	**12.8**	4.6	0.6	3.0	2.2	ND
	mg/kg	0.007	**0.275**	ND	0.010	0.065	**0.191**	0.098	0.059	0.090	0.049	0.063	**0.208**	0.074	0.009	0.049	0.035	ND
Wheat grain	%TRR	ND	3.1	7.1	1.8	2.4	3.9	1.5	1.0	ND	0.5	1.0	0.9	0.4	0.4	0.5	0.8	ND
	mg/kg	ND	0.002	0.005	0.001	0.002	0.003	0.001	0.001	ND	ND	0.001	0.001	ND	ND	ND	0.001	ND

ND = Not detected (<0.001 mg/kg).

[a] = CR3 and CR6 co-elute, the result is given for both compounds.

Minor components and background radioactivity were also present in each sample and collectively this accounted for between 3% and 13% of the residue in the crop samples.

Shaded metabolites indicate where (tentative) identification/characterisation was possible using HPLC chromatography and mass spectral data.

Metabolites in **bold** are those that exceed 0.01 mg/kg/0.1 mg/kg in plant commodities relevant for human/livestock, resp. These metabolites are characterised but currently are not fully identified.

Table B.7.6.1-9: Amount (%TRR and mg/kg), characterisation and identification of radioactive residues in crop extracts from the 365 DAT plant-back interval

Sample	DAT	Lenacil	CR1	CR14	CR7	CR12	CR11	CR16	CR2	CR3	CR4	CR5	CR6	CR8	CR9	CR13
Proposed identification		Lenacil	Polar Metabolites ((glucose) conjugates)		Hydroxylated lenacil		KD304	KQ961	$CH_3CN/CH_3CN/H_2O$ soluble and chromatographically similar to other hydroxylated (and oxidised) lenacil metabolites e.g. KE121, KQ957,KF313,KC943							
Immature spinach	%TRR	0.7	**28.6**	1.6	**16.0**	6.3	1.9	4.9	1.2	3.2	ND	5.4	2.6	0.6	9.9	5.6
	mg/kg	0.001	**0.021**	0.001	**0.011**	0.004	0.001	0.004	0.001	0.002	ND	0.004	0.002	ND	0.007	0.004
Mature spinach	%TRR	ND	**25.9**	**14.2**	11.3	9.0	1.4	2.8	2.4	ND	5.9	ND	2.9	1.3	5.1	4.4
	mg/kg	ND	**0.019**	**0.011**	0.008	0.007	0.001	0.002	0.002	ND	0.004	ND	0.002	0.001	0.004	0.003
Turnip leaves	%TRR	2.9	28.0	1.8	5.3	4.4	ND	2.6	6.7	4.4	11.1	ND	ND	0.3	3.7	7.4
	mg/kg	0.002	0.021	0.001	0.004	0.003	ND	0.002	0.005	0.003	0.008	ND	ND	ND	0.003	0.006
Turnip roots	%TRR	4.0	28.5	8.1	5.8	5.6	4.2	3.6	5.6	1.3	5.7	ND	0.6	ND	5.4	3.2
	mg/kg	ND	0.003	0.001	0.001	0.001	0.001	ND	0.001	ND	0.001	ND	ND	ND	0.001	ND
Wheat forage	%TRR	9.2	15.1	3.9	4.9	2.5	ND	ND	7.6	7.7	3.6	12.6	ND	8.1	0.8	5.7
	mg/kg	0.015	0.025	0.006	0.008	0.004	ND	ND	0.012	0.013	0.006	0.021	ND	0.013	0.001	0.009
Wheat hay	%TRR	2.0	23.4	1.3	5.2	4.1	5.5	4.3	ND	ND	3.2	17.1	5.6	3.5	3.7	2.4
	mg/kg	0.003	0.033	0.002	0.007	0.006	0.008	0.006	ND	ND	0.005	0.024	0.008	0.005	0.005	0.003
Wheat straw	%TRR	0.5	7.8	2.0	**19.5**	4.9	ND	2.8	4.7	ND	ND	15.1	5.8	2.1	1.0	6.2
	mg/kg	0.003	0.050	0.013	**0.124**	0.031	ND	0.018	0.030	ND	ND	0.096	0.037	0.013	0.006	0.040
Wheat grain	%TRR	ND	19.1	13.3	2.9	3.2	1.2	1.4	1.5	ND	1.0	1.1	1.8	0.2	1.1	0.8
	mg/kg	ND	0.004	0.003	0.001	0.001	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND

ND = Not detected.

Minor components and background radioactivity were also present in each sample and collectively this accounted for between 6% and 18% of the residue in the crop samples. Shaded metabolites indicate where (tentative) identification/characterisation was possible using HPLC chromatography and mass spectral data.

Metabolites in **bold** are those that exceed 0.01 mg/kg/0.1 mg/kg in plant commodities relevant for human/livestock. These metabolites are characterised but currently are not fully identified.

Soil analysis

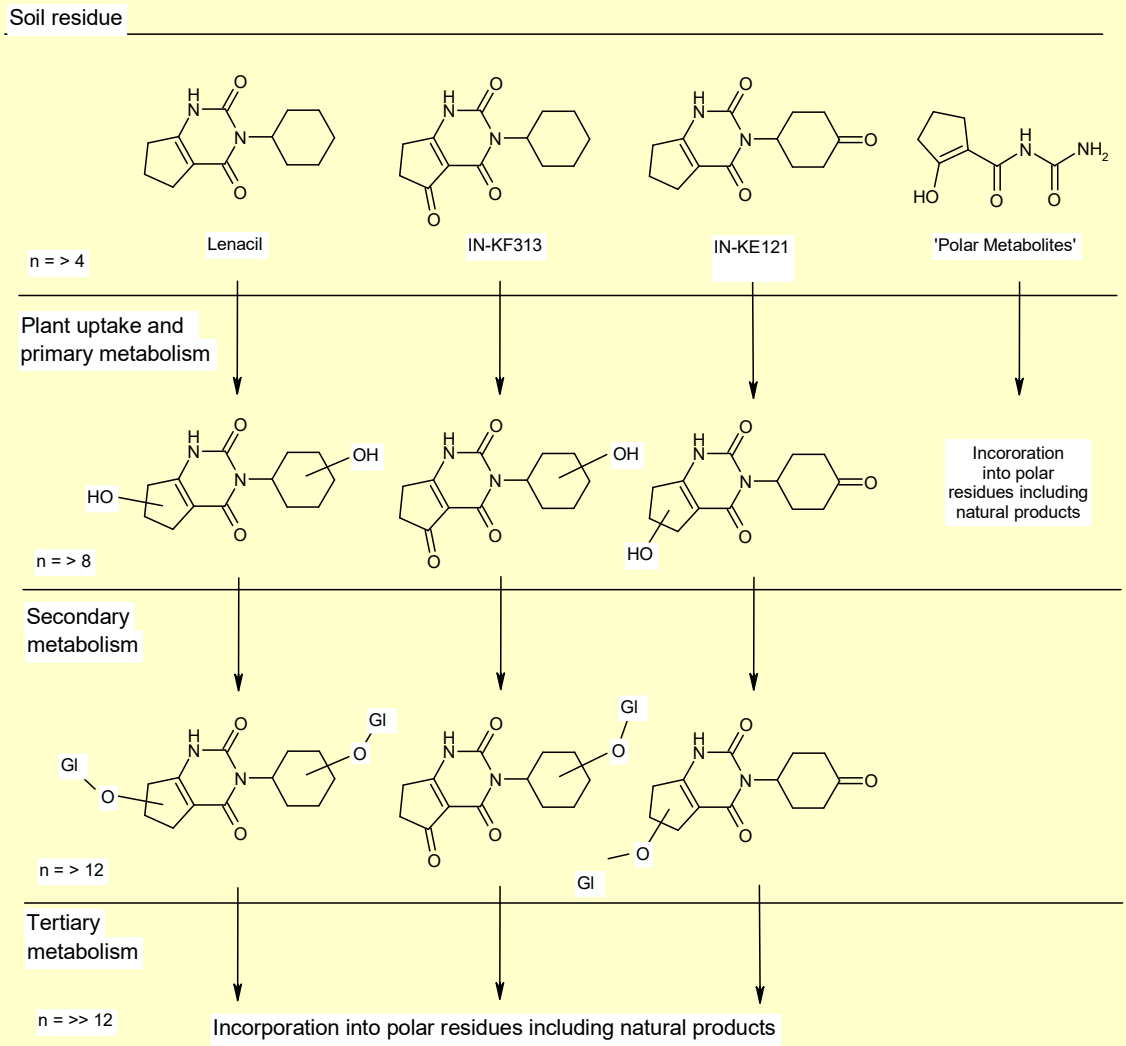
The extractability of residues in **soil** decreased from 72% TRR (at 30 DAT) to 41 % TRR (at 182 DAT), indicating an increased binding of residues to the soil matrix.

Levels of **lenacil** in soil declined from 57% TRR (0.021 mg/kg) at 30 DAT to 20% TRR (0.008 mg/kg) at 182 DAT.

Metabolite **IN-KE121** was tentatively identified (by co-elution with reference standard) and represented 9.6% of TRR at 30 DAT and 3.8% of TRR at 182 DAT. In the 182 DAT soil extract, **IN-KF313** and **IN-KQ957** were tentatively identified (3.4 and 3.9% TRR, respectively). Besides, HPLC chromatograms also showed a complex profile with a polar radioactivity region, as well as a range of many minor degradation products, for which no further attempt of identification was made.

The applicant proposed following **pathway** for uptake of lenacil residues and metabolism in rotational crops: see **Figure B.7.6.1-1.**

Figure B.7.6.1-1: Proposed uptake of lenacil residues and metabolic pathway of lenacil in rotational crops



Phytotoxicity

Lettuce sown at 30 DAT showed significant signs of phytotoxicity, i.e. growth and development was affected (root growth stunted; leaves development hindered). Therefore, lettuce was replaced (at 68 DAT) by spinach (as alternative leafy crop).

The other crops (spinach, turnip and wheat) appeared to have sufficient resilience to the herbicidal action of lenacil residues. Minimal effects were observed (changes in colour or morphology), but by the time of harvest, those crops showed either no morphological changes or showed a slight advanced growth (compared to control crops).

Conclusions:

In this study, an application was to bare (sandy loam) soil at a rate equivalent to 485 g lenacil/ha.

Overall the study shows that lenacil related soil residues are taken up by succeeding crops (cereal, leafy vegetable and root crop) and that there is significant translocation of residues from roots into upper plant parts. However, the level of the resulting residue is dependent on the level of available residue in the soil: TRR levels in almost all crop fractions declined over time, which is consistent with increase of soil bound residues (not available for uptake by plants) over time as demonstrated by soil extraction/analysis.

The amount of parent lenacil detected in rotational crops was lower with increased soil ageing (i.e. with longer plant-back intervals), which is consistent with the fact that lenacil is readily metabolised in most soils. In all crop fractions representative of commodities relevant for human consumption, lenacil residues were below 0.02 mg/kg (for all plant-back intervals), with a maximum of 0.011 mg/kg in turnip roots (sown 30 DAT).

The degradation of lenacil in soil, combined with metabolism in the plant, produces a complex metabolite profile in rotational crops, including a considerable amount of polar residues. There was no significant difference in metabolite profile observed between different crop types, except maybe for wheat, where a more extensive transformation/conjugation of residues was noted and where there were indications of tight binding to and/or incorporation in natural plant constituents (in grain and straw). However, the extractable residue always consists of multiple regions, which appear as compounds that are more polar in nature than lenacil.

Despite attempts made using e.g. LC-MS and enzymatic hydrolysis, definitive identification of all metabolite regions was not possible. However, characterisation and tentative identification of metabolite regions indicate that lenacil and its hydroxylated (and oxidized) metabolites are taken up from the soil and further transformation in the plant occurs via further hydroxylation in a range of positions and via (glucose) conjugation.

Phytotoxic effects were observed in lettuce sown at 30 DAT, while phytotoxic effects were not reported in wheat or turnip sown at the same interval and were not reported in spinach sown 68 DAT.

Assessment and conclusion by RMS:

Acceptability/Reliability: yes

RMS considers that reasonable and significant attempts have been made to characterise and identify the metabolites. Attempts to identify all metabolites were not successful, but the achieved characterisation of the metabolites enabled the elucidation of the overall metabolic pathway. Due to the complex and unresolved chromatographic profile and the variety of positions for hydroxylation (with as consequence probably several metabolites having the same molecular mass), unequivocal identification of the metabolites seems very difficult to achieve.

Outcome and conclusion of the study: see ‘conclusions’ here above.

B.7.6.2. Magnitude of residues in rotational crops

No limited field tests or field residue trials in rotational crops have been conducted; the need for such studies was waived by the applicant, who referred to the results of the confined rotational crop study (*vide supra* – ***B.7.6.1***) (ref.: Doc. M-CA, section 6 – rev. 1 Oct. 2016):

<< The assessment of the potential occurrence of residues in rotational crops in the uptake and metabolism study in rotational crops presented under CA 6.6.1 showed that residues were below the LOQ in all crop fractions representative for commodities relevant for human consumption for all plant-back intervals. Accordingly; no study for limited field tests was required or conducted. >>

Assessment and conclusion by RMS:

To judge the potential **magnitude of residues (and relevance of metabolites)** in rotational crops, the RMS has made a distinction between representative commodities relevant for human consumption[7] and representative commodities relevant for livestock consumption[8], taking into account the different potential for human exposure to the metabolites. Residue levels exceeding 0.01 mg/kg or 0.1 mg/kg (for human food and livestock feed, respectively) are indicated in bold in **Tables B.7.6.1-7, B.7.6.1-8 and B.7.6.1-9** (*vide supra*).

- In crop parts relevant for human consumption at the **30/68 DAT** plant-back interval, residues exceeding 0.01 mg/kg occur in spinach, turnip roots and wheat grain. These residues have been identified as conjugated lenacil hydroxyl metabolites (CR1, max 0.061 mg/kg) and free lenacil hydroxyl metabolites (CR7, 0.038 mg/kg and CR12, 0.022 mg/kg). Unidentified (but characterised) metabolites are present at low levels and are represented by the metabolite regions, CR2 (max 0.028 mg/kg), CR5 (0.016 mg/kg), CR6 (0.021 mg/kg) and CR13 (0.020 mg/kg).
  Residues of parent **lenacil** were not detected or detected at trace levels <0.01 mg/kg in spinach and wheat grain, but were found at higher levels in turnip root (0.011 mg/kg).

- In crop parts relevant for livestock consumption at the **30/68 DAT** plant back interval, residues exceeding 0.1 mg/kg occur only in wheat straw and turnip leaves. These residues have been identified as conjugated lenacil hydroxyl metabolites (CR1, max 0.656 mg/kg) and free lenacil hydroxyl metabolites (CR7, 0.282 mg/kg and CR12, 0.166 mg/kg). Residues remaining unidentified (but characterised) are represented by metabolite regions CR2 (0.641 mg/kg), CR4 (0.140 mg/kg), CR5 (0.702 mg/kg), CR6 (0.580 mg/kg), CR8 (0.223 mg/kg), CR9 (0.176 mg/kg) and CR13 (0.198 mg/kg).
  Residues of parent **lenacil** were not detected or detected at trace levels <0.01 mg/kg in spinach and wheat grain, but were found at higher levels in turnip roots (0.011 mg/kg), turnip leaves (0.045 mg/kg), wheat straw (0.075 mg/kg), wheat forage (0.217 mg/kg) and wheat hay (0.283 mg/kg).

- In crop parts relevant for human consumption at the **182 DAT** plant-back interval, residues exceeding 0.01 mg/kg occur only in spinach with max. of 0.06 mg/kg for CR1, which was shown to be multi-component in nature (a.o. conjugated lenacil hydroxyl metabolites). Other metabolites (CR4, CR7, CR12, CR13) were present at very low levels (0.011 – 0.024 mg/kg) and were identified as free lenacil hydroxylated analogues (CR7, CR12) or tentatively characterised as free or conjugated hydroxylated/oxidised lenacil analogues (CR4, CR13).
  Residues of parent **lenacil** were either not detected (spinach, wheat grain) or detected at trace levels <0.01 mg/kg (turnip root).

- In crop parts relevant for livestock consumption at the **182 DAT** plant-back interval, residues exceeding 0.1 mg/kg occur only in wheat straw. These residues have been identified as conjugated lenacil hydroxyl metabolites (CR1, 0.275 mg/kg) and a free lenacil hydroxyl metabolite (CR12, 0.191 mg/kg). Residues remaining unidentified (but characterised) are represented by metabolite region CR5 (0.208 mg/kg).
  Residues of parent **lenacil** were not detected (spinach, wheat grain) or detected at trace levels <0.01 mg/kg (turnip root/leaves, wheat straw), but were found at higher levels in wheat forage (0.078 mg/kg) and wheat hay (0.030 mg/kg).

[7] spinach, turnip roots and wheat grain
[8] spinach (representative of other green forage crops), turnip (tops and roots, representative of root vegetables) and wheat (straw and grain, representative of all cereals)

- In crop parts relevant for <u>human consumption</u> at the **365 DAT** plant-back interval, residues exceeding 0.01 mg/kg occur only in spinach with max. 0.021 mg/kg for CR1, which was shown to be multi-component in nature (a.o. conjugated lenacil hydroxyl metabolites).
  Residues of parent **lenacil** were not detected or detected at trace levels <0.01 mg/kg.

- In crop parts relevant for <u>livestock consumption</u> at the **365 DAT** plant-back interval, residues exceeding 0.1 mg/kg occur only in wheat straw. These residues have been characterised as a lenacil hydroxyl metabolite (CR7, 0.124 mg/kg).
  Residues of parent **lenacil** were not detected or detected at trace levels <0.01 mg/kg, except in wheat forage (0.015 mg/kg).

B.7.7. OTHER STUDIES

B.7.7.1. Effect on the residue level in pollen and bee products

No experimental data were provided, but the applicant provided the following waiver (Doc. M-CA, section 6 – rev. 1 Oct. 2016):

<< Sugar beets and fodder beets are treated with Lenacil 500 g/L SC during an early life stage (BBCH 10-31), and sugar and fodder beet development does not contain a growth stage which includes flowering. Therefore, residues in pollen and bee products are not expected and a study investigating the effect on residue levels in pollen and bee products is not considered necessary. >>

<u>Assessment and conclusion by RMS</u>:
The RMS considers the applicant's arguments correct and valid. Further RMS's considerations and conclusions: see ***Vol.1, 2.7.8.1***.

B.7.8. REFERENCES RELIED ON

B.7.8.1. Scientific peer-reviewed open literature

Report author	Criollo, R
Report year	2016
Report title	Lenacil – Literature data (Document M-CA, Section 9)
Report No	DuPont-43896 EU
Guidelines followed in study	Submission of scientific peer-reviewed open literature under Regulation (EC) No 1107/2009, EFSA Journal 2011; 9(2):2092
Major deviations from test guideline	None
Guidance in force at time of submission of supplementary dossier	Submission of scientific peer-reviewed open literature under Regulation (EC) No 1107/2009, EFSA Journal 2011; 9(2):2092
Previous evaluation	No; submitted for the purpose of renewal
GLP	Not applicable

In accordance with art. 8(5) of Reg. (EC) No 1107/2009, the applicant performed a comprehensive search of scientific peer-reviewed open literature. The report describes the general search and evaluation process as well as details on search profiles, search histories and summary tables of the results.

For the sections concerning residues/consumer safety, the main search was conducted on 11 February 2016. This search covered articles published since 2005, which is in line with the legally requested period of the last 10 years prior to dossier submission.

The initial search was a single concept search capturing all data points using search terms and synonyms for the **active substance lenacil** (*i.a.* CAS no, chemical name, commercial names, company codes). As a large number of search results returned from the single concept search, making assessment for relevance impractical, a separate, focussed search was conducted for grouped data points (search per discipline). This search was a refinement of the initial single concept search. The search filter for residues comprised key words such as reflected in the search statements shown here below.

L56 239 S L41 AND (RESIDUE? OR MULTIRESIDUE? OR STORAGE STABILITY OR METABOLIC OR METABOLISM OR DEGRADATION OR BREAKDOWN OR PLANT# OR CROP? OR FEED OR ANIMAL# OR LIVESTOCK# OR HEN OR CATTLE OR RUMINANT# OR GOAT? OR COW# OR PIG? OR FISH OR MILK OR HONEY)
L57 126 S L41 AND (PROCESS? OR HYDROLY? OR ROTATION? OR SUCCEED? OR RISK OR ASSESSMENT OR RISK ASSESSMENT OR CONSUME? OR EXPOSURE OR CROSS CONTAMINATION OR BIOMONITORING OR MONITORING OR ENVIRONMENTAL CONTAMINA?)

The search was conducted using several scientific literature databases (AGRICOLA, BIOSIS, CABA, HCAPLUS, CNSB, DDFU, EMBASE, ESBIOBASE, FSTA, IPA, MEDLINE, NTIS, PASCAL, PQSCITECH, SCISEARCH, TOXCENTER, CAS Registry, HSDB). Patent literature was excluded from the search.

The relevance criteria chosen for the selection of peer reviewed scientific open literature (in relation to the data requirements for residues/consumer safety) are listed here below:

1. The dose levels or application rates reflect the proposed GAP.
2. The test system, target crop, or species are prescribed by Regulation (EC) No 1107/2009 or the relevance is explained if not standard.
3. Well identified test material, including its purity and impurity profile, is described.
4. Study design and/or execution are consistent with relevant study guidelines.
5. The endpoint is relevant to an EU data point as prescribed by Regulations (EU) No 283/2013 and 284/2013
6. The application method(s) complies with Good Agriculture Practice (GAP)
7. Appropriate in-life/processing conditions are used and/or are well described

Combining the searches for the different disciplines resulted in 311 unique summary records, which were subjected to further screening on relevance and selection.

As a first step, a rapid assessment based on summary records (title/abstracts) was performed by expert reviewers. Based on this rapid assessment, the records were assigned to one of the following categories:

- Metabolism and Residue (43 records)
- Toxicology (14 records)
- Environmental Fate (59 records)
- Ecotoxicology (16 records)
- Obviously irrelevant (179 records)

Summary records were assigned to the category 'Obviously irrelevant' if they were clearly related to one of the following topics:

- Efficacy
- Resistance of targets
- Analytical method development, calibration
- New ways of synthesis
- Studies on a molecular level, which cannot be related to environmental risk assessment
- Non-EU monitoring studies, non-EU field studies
- Publications in non-EU language without English abstract
- Abstract refers to a conference contribution and does not contain data, full text not available
- Not relevant due to missing information: Studies with target organisms

The 'Obviously irrelevant' records were not considered further.

For the other four categories, non-relevant studies were excluded by applying the relevance criteria previously mentioned (*vide supra*). When the summary records did not contain sufficient information to assess relevance, full text documents were reviewed in detail for relevance according to the previously defined criteria.

From the total of 311 summary records that were reviewed, 305 were not relevant, resulting in 6 studies that have been selected for inclusion in the dossier. All of the 43 records assigned to the category 'Residues' were assigned as irrelevant, based on the rapid assessment for relevance. The justifications provided by the applicant for non-relevance of each of those individual records were reviewed by the RMS and were considered acceptable, with the exception of following 3 publications, for which RMS considered a review of the full-text necessary to decide on their (non-)relevance. The publications from 2009 and 2014 were in Polish language only (abstract available in English). The publication from 2011 was brought up by the co-RMS.

Author(s)	Year	Title	Source	Applicant's justification for non-relevance	RMS's judgement on relevance (after review of full-text document)
Kucharski, M.; Sadowski, J.; Wujek, B.	2009	Influence of herbicide application system on lenacil residues in soil and roots of sugar beet.	Progress in Plant Protection (2009), Volume 49, Number 4, pp. 1868-1874.	The paper is not relevant as it does not contain an endpoint which is relevant for the risk assessment. The study compares the influence of different application methods on the concentrations of various herbicides in soil and sugar beet roots at harvest. Within this scope soil concentrations of lenacil were measured at harvest of sugar beets. However, detailed information on the soil and climate is missing and it is not possible to derive a reliable degradation endpoint based on the data.	RMS agrees with applicant that the publication is **not relevant** in the framework of renewal of the a.s.lenacil. Lenacil (formulated as Venzar 80 WP) was applied in combination with other herbicides, according to 3 different application schemes (for lenacil: A: 1x 960 g/ha pre-emergence; B: 3x 720 g/ha post-emergence; C: 4x 320g/ha from start of weed emergence on and with interval of 7-10 days). None of these application schemes are in accordance with the GAP of the representative uses for lenacil supported for renewal. Further details on experimental conditions, locations etc. have not been reported. Reported residue level values for lenacil in sugar beet root (and in soil) were <0.01 mg/kg for all samples and all application schemes. However, no experimental proof of analytical method validation was reported in the publication.

Author(s)	Year	Title	Source	Applicant's justification for non-relevance	RMS's judgement on relevance (after review of full-text document)
Kucharski, M.; Sadowski, J.	2014	Herbicide residues in sugar beet roots.	Progress in Plant Protection (2014), Volume 54, Number 1, pp. 5-8	Analytical method development, calibration. Not subject to relevant for residues analyzed and/or residues occurrence from lenacil application on crops and side effects on health	RMS is of the opinion that the publication is **not relevant** in the framework of renewal of the a.s.lenacil. Besides other herbicides, also lenacil residues were determined in sugar beet roots, in the framework of monitoring tests over 3 growing seasons (2010-2012) on 53 sugar beet plantations located in South-Western Poland. Details on application schemes and conditions are not provided in the publication; it is just mentioned that farmers indicated they had followed the label recommendations. For the analysis of lenacil in sugar beet root, HPLC-UV was used and an LOQ of 0.0005 mg/kg was mentioned in the publication. 2 Lenacil was found in 2 samples (out of the 27 samples that were analysed for lenacil) and detected concentration range was reported as 0.0015-0.0026 mg/kg.

Author(s)	Year	Title	Source	Applicant's justification for non-relevance	RMS's judgement on relevance (after review of full-text document)
Kucharski, M.; Sadowski, J.; Wujek, B.; Trajdos, J.	2011	Influence of adjuvants addition on lenacil residues in plant and soil	Polish Journal of Agronomy (2011), Volume 5, pp. 39–42	No justification provided by applicant (publication brought up by co-RMS AT).	In this study the influence of adjuvant addition (oil, surfactant, and multicomponent) on lenacil residues in soil and roots of sugar beet was investigated. The addition of adjuvants caused an increase of the active substance residues in soil and roots of sugar beet in comparison with the treatments, where lenacil was used without adjuvant. The increase of the lenacil residues was statistically significant for most of soil and plant samples and amounted average to 45 and 41% respectively. Influence of single adjuvant on lenacil residues in soil and plant was different for each year (experimental season). However, the residues of lenacil determined in roots of sugar beet were very low (<0.0005–0.0020 mg/kg) and did not exceed existing MRL values of lenacil. Anyway, the significant increase of lenacil residues after use of adjuvants in combination with a lenacil product is potentially important information. Therefore, the published results from this study have been **summarized in the DRAR Vol.3, B.7.3**.

As in the literature search conducted in 2016 (see above), the single concept search and subsequent focussed search per discipline was focussed on the active substance alone, the RMS requested the applicant (during the preparation of the DRAR) to perform a literature search for the relevant **metabolites** as well. An additional search was conducted on the 14th of August 2018 and results were provided to the RMS before finalisation of the DRAR, in the format of an additional literature review report:

Report author	Anonymous
Report year	2018 (3 Sept. 2018)
Report title	Literature Review Report – Scientific peer-reviewed *[sic]* of open literature for the approval of pesticide active substance lenacil (focus on its metabolites) as under Article 8(5) of Regulation (EC) No 1107/2009
Report No	106052-CA9-1
Guidelines followed in study	Submission of scientific peer-reviewed open literature under Regulation (EC) No 1107/2009, EFSA Journal 2011; 9(2):2092
Major deviations from test guideline	None
Guidance in force at time of submission of supplementary dossier	Submission of scientific peer-reviewed open literature under Regulation (EC) No 1107/2009, EFSA Journal 2011; 9(2):2092
Previous evaluation	No; submitted for the purpose of renewal
GLP	Not applicable

The additional search for the metabolites was conducted on the 14th of August 2018 and covered the same time window as the literature search for the active substance, i.e. 2005-2016. Several scientific literature databases were used (AGRICOLA, BIOSIS, CABA, CAPLUS, CNSB, EMBASE, ESBIOBASE, FSTA, MEDLINE, PQSCITECH, SCISEARCH, TOXCENTER). The approach used for the search was the single concept search, employing key words (related to the chemical names), CAS no (where available) and company codes for the metabolites IN-KE121, IN-KF313, IN-KC943, IN-KQ961, IN-KD304 and IN-KQ957.
A total of 2 summary records were retrieved by this search. One was the EFSA conclusion on confirmatory data of lenacil (EFSA, 2013), which has obviously been considered in the DRAR (though not specifically relevant for residues; rather environmental fate and behaviour). A second article related to toxicology, but was considered irrelevant, as it concerned a totally different compound with the same code as one of the metabolites of lenacil (IN-KE121).

Conclusion:
Overall, the RMS considers the literature search and review as performed by the applicant to be acceptable and in line with the recommendations made in EFSA Journal 2011; 9(2):2092 (EFSA, 2011). The search process has been sufficiently documented in all details with search profiles, search histories and summary tables according to the guidance of EFSA (EFSA, 2011).

B.7.8.2. Other references cited (not submitted by the applicant)

Belgium, 2007. Draft Assessment Report (DAR) on the active substance lenacil prepared by the rapporteur Member State, Belgium, in the framework of Directive 91/414/EEC, November 2007.

Belgium, 2009. Final Addendum to the DAR on lenacil, compiled by EFSA, July 2009.

Belgium, 2012. Addendum to the Draft Assessment Report section B8, confirmatory data, December 2012.

Belgium, 2013. Addendum to the Draft Assessment Report section B7, confirmatory data, March 2013.

EFSA, 2013. Conclusion on the peer review of the pesticide risk assessment of confirmatory data submitted for the active substance lenacil. EFSA Journal 2013,11(9):3354, 19 pp. doi:10.2903/j.efsa.2013.3354

B.7.8.3. Studies submitted (or referred to) by the applicant

Datapoint/ dossier ref. (cf. Reg. (EU) 283/2013) *[former EU dossier ref.]*	Author(s)	Year	Title Company Report No. Source (where different from company) GLP or GEP status Published or not	Vertebrate study Y/N	Data protection claimed Y/N	Justification if data protection is claimed	Owner	Previous evaluation
CA, 6.1 *[IIA, 6.0/01]*	Hamberger, R.	2002	Analytical report. Generation of samples for the determination of residues of Venzar (80% WP (containing 08% lenacil) in sugar beets, one site in Europe, 2002. IFU Umweltanalytik GmbH, Report No.: 20011048/E2-FPSB GLP: Yes, Unpublished	N	N	-	FMC*	DAR (2007-2009)
CA, 6.2.1 *[IIA, 6.1/01]*	Zhang, M.; Glunt, C.D.	2002	Metabolism of lenacil in sugar beets. DuPont Doc. No.: 3649-95 GLP: Yes, Unpublished	N	N	-	FMC*	DAR (2007-2009)
CA, 6.2.1/01 *(supportive/ supplementary only)*	Chrzanowski, R.L.	1978	Metabolism of ^{14}C-Lenacil in Spinach. DuPont Biochemicals Department, Wilmington, Delaware, USA, Report No.: D-2-3 GLP : No Unpublished	N	N	-	FMC*	Submitted for the purpose of renewal; DRAR (2019)
CA, 6.2.1/02 *(supportive/ supplementary only)*	Chrzanowski, R.L.	1978	Metabolism of ^{14}C-Lenacil in Strawberries. DuPont Biochemicals Department, Wilmington, Delaware, USA, Report No.: LLME-3-78 GLP : No Unpublished	N	N	-	FMC*	Submitted for the purpose of renewal; DRAR (2019)
CA, 6.3.1 *[IIA, 6.3/02]*	Pollmann, B	2002	Generation of samples for the determination if residues of Venzar	N	N	-	FMC*	DAR (2007-2009)

Datapoint/ dossier ref. (cf. Reg. (EU) 283/2013) *[former EU dossier ref.]*	Author(s)	Year	Title Company Report No. Source (where different from company) GLP or GEP status Published or not	Vertebrate study Y/N	Data protection claimed Y/N	Justification if data protection is claimed	Owner	Previous evaluation
			(80% WP (containing 08% lenacil) in sugar beets, five site in Europe, 2001. GAB Biotechnologie GmbH IFU Umweltanalytik GmbH Report No.: 20011048/E1-FPSB GLP: Yes, Unpublished → incl. analytical report (Mende, P., 2002)					
CA, 6.3.1 *[IIA, 6.3/03]*	Pollmann, B	2002	Generation of samples for the determination if residues of Venzar (80% WP (containing 08% lenacil) in sugar beets, one site in Europe, 2002. GAB Biotechnologie GmbH IFU Umweltanalytik GmbH, Report No.: 20011048/E2-FPSB GLP: Yes, Unpublished → incl. analytical report (Hamberger, R., 2002)	N	N	-	FMC*	DAR (2007-2009)
CA, 6.3.1 *[IIA, 6.3/04]*	Anderson, I., Kakkonen, J.E.	2002	Decline of lenacil residues in sugar beet (root and tuber vegetables) following a single application of Venzar® 80WP (lenacil) – Southern Europe, Season 2005. Report No.: CRL 688479 GLP: Yes, Unpublished → incl. analytical phase report (Witte, A., 2006)	N	N	-	FMC*	DAR (2007-2009)
CA 6.3	Kucharski, M., Sakowski, J.,	2011	Influence of adjuvants addition on lenacil residues in plant and soil	N	N	Not applicable	public	DRAR (2019)

Datapoint/ dossier ref. (cf. Reg. (EU) 283/2013) *[former EU dossier ref.]*	Author(s)	Year	Title Company Report No. Source (where different from company) GLP or GEP status Published or not	Vertebrate study Y/N	Data protection claimed Y/N	Justification if data protection is claimed	Owner	Previous evaluation
(supportive/ supplementary only)	Wujek, B., Trajdos, J.		Polish Journal of Agronomy (2011), Volume 5, pp. 39–42 Non-GLP, published					
CA, 6.6.1/01	Hurst, L., Fletcher, T.	2013	[14C]-Lenacil: Uptake and metabolism in confined rotational crops. Smithers Viscient (ESG) Ltd., Report No.: 8240837 GLP, Unpublished	N	Y	**Applicant:** *"The study is necessary for the regulatory decision, conducted according to GLP and has not previously been protected. Although the study was previously submitted in the framework of post-approval confirmatory data, data protection is now claimed by the applicant because no EU data protection under 91/414/EEC has been granted for this study. Accordingly, data protection should be granted at MS level either with post Annex I product reregistration (30 months) or with a new product registration (10 years)."* **RMS:** Previously, this study had been submitted and considered at EU level, to address the confirmatory data request following the original EU review under 91/414/EEC. Thus, the study is to be regarded rather as a part of the original approval dossier and cannot be	FMC*	DAR addendum (2013)

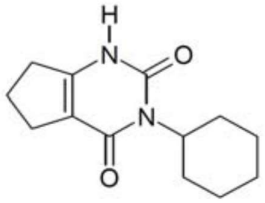
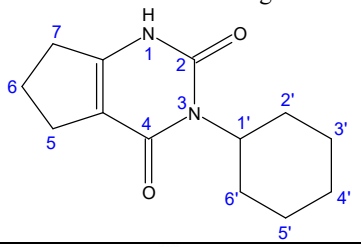
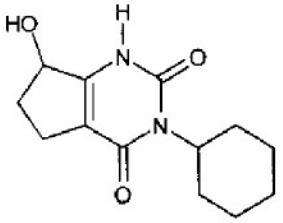
Datapoint/ dossier ref. (cf. Reg. (EU) 283/2013) *[former EU dossier ref.]*	Author(s)	Year	Title Company Report No. Source (where different from company) GLP or GEP status Published or not	Vertebrate study Y/N	Data protection claimed Y/N	Justification if data protection is claimed	Owner	Previous evaluation
						considered as 'necessary' in the sense of art. 7(1)(d) of Reg. 844/2012. Furthermore, the study has also been submitted at national/zonal level in some MSs (e.g. BE), in the framework of the national authorisation procedure (re-registration of PPPs containing lenacil). Therefore, the applicant's claim that the study "*has not previously been protected*" deserves careful consideration at MS level; A study cannot be granted data protection 2 times (cf. Reg. (EC) No 1107/2009 art. 59(2)(b)).		

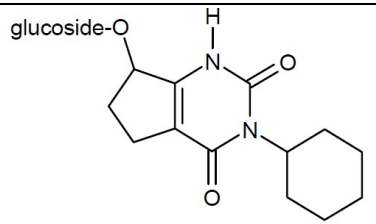
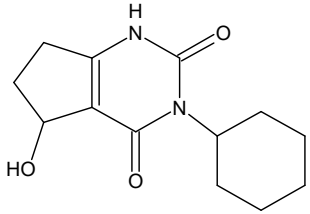
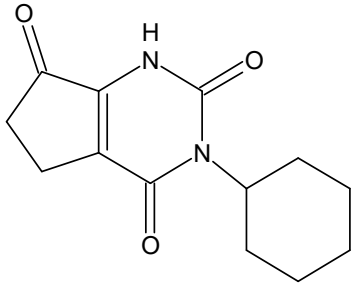
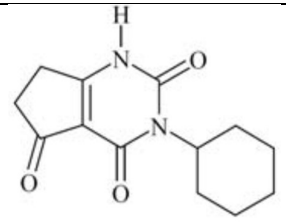
* FMC: FMC International Switzerland Sàrl

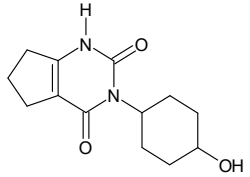
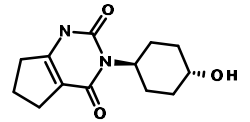
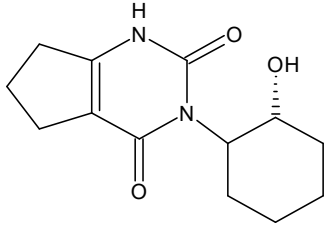
B.7.9. APPENDICES

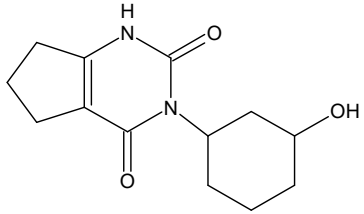
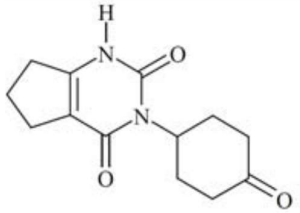
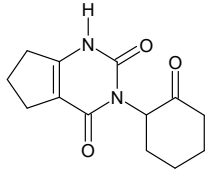
B.7.9.1. List of metabolites and their occurrence in plants and livestock animals

The table below includes metabolites mentioned in this volume (Vol.3, B.7), as metabolite (tentatively) identified in one or more studies and/or as reference standard. The table also includes the parent compound, as a reference. For a full overview table with all metabolites mentioned across all sections, including those occurring in the rat and/or the environment: see DRAR ***Vol.1, 3.4.2***.

Code Number (Synonyms)	Description	Occurrence in plant and/or livestock animals	Structural formula
Lenacil DPX-B0634	**Chemical name (IUPAC):** 3-cyclohexyl-1,5,6,7-tetrahydrocyclopentapyrimidine-2,4(3H)-dione **Common name:** Lenacil **CAS number**: 2164-08-1 **Molecular weight:** 234.3 g/mol	Plant (sugar beet)	 IUPAC-numbering: 
IN-KC943	**Chemical name (IUPAC):** 3-cyclohexyl-6,7-dihydro-7-hydroxy-1-H-cyclopentapyrimidine-2,4(3H,5H)-dione **CAS number**: Not available **Molecular weight:** 251.3 g/mol	Plant (sugar beet foliage 3.1%TRR/<0.01ppm; *spinach 1.1%TRR***)	

Code Number (Synonyms)	Description	Occurrence in plant and/or livestock animals	Structural formula
IN-KC943-glucoside	**CAS number**: Not available **Molecular weight:** 414.46 g/mol	Plant (sugar beet foliage 11%TRR/<0.02ppm)	
IN-KQ961	**Chemical name (IUPAC):** 3-Cyclohexyl-6,7-dihydro-5-hydroxy-1H-cyclopentapyrimidine-2,4-(3H,5H)-dione **CAS number**: Not available **Molecular weight:** 251.3 g/mol	*Plant (spinach 1.1%TRR**)* Rotational crops (spinach, turnip, wheat)	
IN-KD302	**Chemical name (IUPAC):** 3-Cyclohexyl-5,6-dihydro-1H-cyclopentapyrimidine-2,4,7-(3H)-trione	*Plant (spinach <1%TRR**)*	
IN-KF313	**Chemical name (IUPAC):** 3-Cyclohexyl-6,7-dihydro-1H-cyclopentapyrimidine-2,4,5-(3H)-trione **CAS number**: Not available **Molecular weight:** 248.28 g/mol	Soil, water and sediment	

Code Number (Synonyms)	Description	Occurrence in plant and/or livestock animals	Structural formula
IN-KD304 *Z*-isomer (KD304 - *Z*-isomer; IN-G2172) *E*-isomer (KD304 - *E*-isomer)	**Chemical name (IUPAC):** *Cis*-6,7-dihydro-3-(4-hydroxycyclohexyl)-1H-cyclopentapyrimidine-2,4-(1H,3H)-dione *Trans*-6,7-dihydro-3-(4-hydroxycyclohexyl)-1H-cyclopentapyrimidine-2,4-(3H,5H)-dione	*Plant (spinach 11%TRR**);* Rotational crops (spinach, turnip, wheat)	 *Z*-isomer  *E*-isomer
IN-KD305 (*E*-isomer)	**Chemical name (IUPAC):** *Trans*-1,5,6,7-Tetrahydro-3-(2-hydroxycyclohexyl)-2H-cyclopentapyrimidine-2,4-(3H)-dione	/	

Code Number (Synonyms)	Description	Occurrence in plant and/or livestock animals	Structural formula
IN-KC939 (Z/E 2:1)	**Chemical name (IUPAC):** 6,7-Dihydro-3-(3-hydroxycyclohexyl)-1H-cyclopentapyrimidine-2,4-(3H,5H)-dione	/	
IN-KE121	**Chemical name (IUPAC):** 6,7-Dihydro-3-(4-oxocyclohexyl)-1H-cyclopentapyrimidine-2,4-(3H,5H)-dione **CAS number**: Not available **Molecular weight:** 248.28 g/mol	Soil, water and sediment	
IN-KQ957	**Chemical name (IUPAC):** 6,7-Dihydro-3-(2-oxocyclohexyl)-1H-cyclopentapyrimidine-2,4-(3H,5H)-dione	Soil (tentative identification in soil of confined rotational crop study)**	

** tentative (identity and occurrence based on co-chromatography with standards; not confirmed by other means)

B.7.9.2. Tier I summaries of the residue trials in support of the representative uses

FIELD TRIALS, CROP RESIDUE SUMMARY

Active substance (common name)	:	Lenacil	Commercial Product (name)	:	Venzar
Crop/crop group	:	Sugar beet / root and tuber vegetables	Producer of commercial product	:	Schirm GmbH
Responsible body for reporting(name, address)	:	Schirm GmbH Geschwister-Scholl-Str. 127, 39218 Schönebeck, Germany	Indoor/Glasshouse/Outdoor	:	Outdoor
Country	:	France	Other active substance in the		
Content of active substance (g/kg or g/L)	:	Lenacil 800 g/kg	formulation (common name and content)	:	None; but some treatments applied as a blend with DPX-MX843-1
Formulation (eg WP)	:	WP	Residues calculated as	:	Lenacil

1	2	3	4	5			6	7	8	9	10	11
Report no. Location (region)	Commodity/ variety	Date of: 1. Sowing or planting 2. Flowering 3. Harvest	Method of treatment	Application rate per treatment			Dates of treatment(s) or no. of treatment(s) and last date	Growth stage at last treatment or date	Portion analysed	Residues (mg/kg)	PHI (days)	Remarks
				kg a.s./hL	Water (L/ha)	kg a.s./ha						
F-95-001-RES Soudron, 51, N.France (North EU)	Sugar beet / Anik	1) 06 Apr 95 2) - 3) 29 Sep 95	Broadcast spray	0.445	181.5	0.806	14 Apr 95	Pre-em	Roots Tops	< 0.01 < 0.01	168 168	LOQ 0.01 mg//kg
			Broadcast spray	0.146 0.144	176 188	0.257* 0.271*	11 May 95 23 May 95	4 leaves (BBCH 14)	Roots Tops	< 0.01 < 0.01	129 129	Analytical method: DFG Method S19 (GLC/MSD) Analysis date: Nov 97
			Broadcast spray	0.044 0.044 0.044 0.044	192 185 165 176	0.084* 0.081* 0.072* 0.077*	5 May 95 11 May 95 23 May 95 I Jun 95	7 leaves (BBCH 17)	Roots Tops	< 0.01 < 0.01	121 121	
			-	-	-	0	-	-	Roots Tops	< 0.01 < 0.01	-	Frozen storage period : 26 months.

* Applied as Venzar blended with DPX-MX843-1 (a formulation containing triflusulfuron methyl).

Active substance (common name)	:	Lenacil	Commercial Product (name)	:	Venzar
Crop/crop group	:	Sugar beet / root and tuber vegetables	Producer of commercial product	:	Schirm GmbH
Responsible body for reporting(name, address)	:	Schirm GmbH Geschwister-Scholl-Str. 127, 39218 Schönebeck, Germany	Indoor/Glasshouse/Outdoor	:	Outdoor
Country	:	France	Other active substance in the		
Content of active substance (g/kg or g/L)	:	Lenacil 800 g/kg	formulation (common name and content)	:	None; but some treatments applied as a blend with DPX-MX843-1
Formulation (eg WP)	:	WP	Residues calculated as	:	Lenacil

1	2	3	4	5			6	7	8	9	10	11
Report no. Location (region)	Commodity/variety	Date of: 1. Sowing or planting 2. Flowering 3. Harvest	Method of treatment	Application rate per treatment			Dates of treatment(s) or no. of treatment(s) and last date	Growth stage at last treatment or date	Portion analysed	Residues (mg/kg)	PHI (days)	Remarks
				kg a.s./hL	Water (L/ha)	kg a.s./ha						
F-95-001-RES Rupéreux, 77, N.France (North EU)	Sugar beet / Platine	1) 13 Apr 95 2) - 3) 11 Oct 95	Broadcast spray	0.358	222	0.794	20 Apr 95	Pre-em	Roots Tops	< 0.01 < 0.01	174 174	LOQ 0.01 mg//kg Analytical method: DFG Method S19 (GLC/MSD) Analysis date: Nov 97 Frozen storage period : 25 months.
			Broadcast spray	0.116 0.128	233 215	0.270* 0.276*	15 May 95 24 May 95	4 leaves (BBCH 14)	Roots Tops	< 0.01 < 0.01	140 140	
			Broadcast spray	0.036 0.036 0.036 0.040	216 230 210 213	0.078* 0.083* 0.075* 0.085*	5 May 95 15 May 95 24 May 95 2 Jun 95	7/8 leaves (BBCH 17,18)	Roots Tops	< 0.01 < 0.01	131 131	
			-	-	-	0	-	-	Roots Tops	< 0.01 < 0.01	-	

* Applied as Venzar blended with DPX-MX843-1 (a formulation containing triflusulfuron methyl).

Active substance (common name)	:	Lenacil	Commercial Product (name)	:	Venzar
Crop/crop group	:	Sugar beet / root and tuber vegetables	Producer of commercial product	:	Schirm GmbH
Responsible body for reporting(name, address)	:	Schirm GmbH Geschwister-Scholl-Str. 127, 39218 Schönebeck, Germany	Indoor/Glasshouse/Outdoor	:	Outdoor
Country	:	France	Other active substance in the		
Content of active substance (g/kg or g/L)	:	Lenacil 800 g/kg	formulation (common name and content)	:	None; but some treatments applied as a blend with DPX-MX843-1
Formulation (eg WP)	:	WP	Residues calculated as	:	Lenacil

1	2	3	4	5			6	7	8	9	10	11
Report no. Location (region)	Commodity/variety	Date of: 1. Sowing or planting 2. Flowering 3. Harvest	Method of treatment	Application rate per treatment			Dates of treatment(s) or no. of treatment(s) and last date	Growth stage at last treatment or date	Portion analysed	Residues (mg/kg)	PHI (days)	Remarks
				kg a.s./hL	Water (L/ha)	kg a.s./ha						
F-95-001-RES Epagny, 02290, N.France (North EU)	Sugar beet / Orion	1) 14 Apr 95 2) - 3) 29 Sep 95	Broadcast spray	0.400	190	0.760	21 Apr 95	Pre-em	Roots Tops	< 0.01 < 0.01	161 161	LOQ 0.01 mg//kg
			Broadcast spray	0.134 0.134	203 193	0.271* 0.259*	1 Jun 95 8 Jun 95	8-10 leaves (BBCH 19)	Roots Tops	< 0.01 < 0.01	113 113	Analytical method: DFG Method S19 (GLC/MSD)
			Broadcast spray	0.040 0.040 0.134	210 193 183	0.084* 0.078* 0.245*	4 May 95 12 May 95 22 May 95	4 leaves (BBCH 14)	Roots Tops	< 0.01 < 0.01	130 130	Analysis date: Nov 97
			-	-	-	0	-	-	Roots Tops	< 0.01 < 0.01	-	Frozen storage period : 26 months.

* Applied as Venzar blended with DPX-MX843-1 (a formulation containing triflusulfuron methyl).

Active substance (common name)	:	Lenacil	Commercial Product (name)	:	Venzar
Crop/crop group	:	Sugar beet / root and tuber vegetables	Producer of commercial product	:	Schirm GmbH
Responsible body for reporting(name, address)	:	Schirm GmbH Geschwister-Scholl-Str. 127, 39218 Schönebeck, Germany	Indoor/Glasshouse/Outdoor	:	Outdoor
Country	:	Germany	Other active substance in the		
Content of active substance (g/kg or g/L)	:	Lenacil 800 g/kg	formulation (common name and content)	:	None
Formulation (eg WP)	:	WP	Residues calculated as	:	Lenacil

1	2	3	4	5			6	7	8	9	10	11
Report no. Location (region)	Commodity/variety	Date of: 1. Sowing or planting 2. Flowering 3. Harvest	Method of treatment	Application rate per treatment			Dates of treatment(s) or no. of treatment(s) and last date	Growth stage at last treatment or date	Portion analysed	Residues (mg/kg)	PHI (days)	Remarks
				kg a.s./hL	Water (L/ha)	kg a.s./ha						
20011048/E1-FPSB 21739 Dollern (Niedersachsen)	Sugar beet / Wieblu	1) 14 Apr 01 2) - 3) 11 Sep 01	Broadcast spray	0.166	324	0.539	3 Jul 01	BBCH 37 (leaves covering 70 % of ground)	Roots Tops	**< 0.02** **< 0.02**	70 70	LOQ 0.02 mg//kg
G01N003R, Germany (North EU)			-	-	-	0	-	-	Roots Tops	< 0.02 < 0.02	-	Analytical method: (HPLC/MS-MS) Analysis date: 8 Feb 02 Frozen storage period : 5 months.

Active substance (common name)	: Lenacil	Commercial Product (name)	: Venzar
Crop/crop group	: Sugar beet / root and tuber vegetables	Producer of commercial product	: Schirm GmbH
Responsible body for reporting(name, address)	: Schirm GmbH Geschwister-Scholl-Str. 127, 39218 Schönebeck, Germany	Indoor/Glasshouse/Outdoor	: Outdoor
Country	: Germany	Other active substance in the	
Content of active substance (g/kg or g/L)	: Lenacil 800 g/kg	formulation (common name and content)	: None
Formulation (eg WP)	: WP	Residues calculated as	: Lenacil

1	2	3	4	5			6	7	8	9	10	11
Report no. Location (region)	Commodity/variety	Date of: 1. Sowing or planting 2. Flowering 3. Harvest	Method of treatment	Application rate per treatment			Dates of treatment(s) or no. of treatment(s) and last date	Growth stage at last treatment or date	Portion analysed	Residues (mg/kg)	PHI (days)	Remarks
				kg a.s./hL	Water (L/ha)	kg a.s./ha						
20011048/E1-FPSB 21726 Willah (Niedersachsen) G01N004R, Germany (North EU)	Sugar beet / Kassandra	1) 12 Apr 01 2) - 3) 12 Sep 01	Broadcast spray	0.167	315	0.526	4 Jul 01	BBCH 37	Roots Tops	**< 0.02** **0.04**	70 70	LOQ 0.02 mg//kg
			-	-	-	0	-	-	Roots Tops	< 0.02 < 0.02	-	Analytical method: (HPLC/MS-MS) Analysis date: 8 Feb 02 Frozen storage period : 5 months.

Active substance (common name)	:	Lenacil	Commercial Product (name)	:	Venzar
Crop/crop group	:	Sugar beet / root and tuber vegetables	Producer of commercial product	:	Schirm GmbH
Responsible body for reporting(name, address)	:	Schirm GmbH Geschwister-Scholl-Str. 127, 39218 Schönebeck, Germany	Indoor/Glasshouse/Outdoor	:	Outdoor
Country	:	Germany	Other active substance in the		
Content of active substance (g/kg or g/L)	:	Lenacil 800 g/kg	formulation (common name and content)	:	None
Formulation (eg WP)	:	WP	Residues calculated as	:	Lenacil

1	2	3	4	5			6	7	8	9	10	11
Report no. Location (region)	Commodity/variety	Date of: 1. Sowing or planting 2. Flowering 3. Harvest	Method of treatment	Application rate per treatment			Dates of treatment(s) or no. of treatment(s) and last date	Growth stage at last treatment or date	Portion analysed	Residues (mg/kg)	PHI (days)	Remarks
				kg a.s./hL	Water (L/ha)	kg a.s./ha						
20011048/E1-FPSB 75248 Dürrn (Baden-Würtemberg) G01N005R, Germany	Sugar beet / Impuls	1) 27 Apr 01 2) - 3) 19 Sep 01.	Broadcast spray	0.167	301	0.502	4 Jul 01	BBCH 37	Leaves Leaves Leaves Roots Tops Roots Tops	3.8 1.4 0.09 < 0.02 0.04 **< 0.02** **< 0.02**	0 15 28 42 42 77 77	LOQ 0.02 mg//kg Analytical method: (HPLC/MS-MS) Analysis date: 8 Feb 02 Frozen storage period : 5 months.
(North EU)			-	-	-	0	-	-	Leaves Leaves Leaves Roots Tops Roots Tops	< 0.02 < 0.02 < 0.02 < 0.02 < 0.02 < 0.02 < 0.02	-	

Active substance (common name)	: Lenacil	Commercial Product (name)	: Venzar
Crop/crop group	: Sugar beet / root and tuber vegetables	Producer of commercial product	: Schirm GmbH
Responsible body for reporting(name, address)	: Schirm GmbH Geschwister-Scholl-Str. 127, 39218 Schönebeck, Germany	Indoor/Glasshouse/Outdoor	: Outdoor
Country	: Germany	Other active substance in the	
Content of active substance (g/kg or g/L)	: Lenacil 800 g/kg	formulation (common name and content)	: None
Formulation (eg WP)	: WP	Residues calculated as	: Lenacil

1	2	3	4	5			6	7	8	9	10	11
Report no. Location (region)	Commodity/variety	Date of: 1. Sowing or planting 2. Flowering 3. Harvest	Method of treatment	Application rate per treatment			Dates of treatment(s) or no. of treatment(s) and last date	Growth stage at last treatment or date	Portion analysed	Residues (mg/kg)	PHI (days)	Remarks
				kg a.s./hL	Water (L/ha)	kg a.s./ha						
20011048/E1-FPSB 76744 Wörth (Rheinland-Pfalz) G01N006R, Germany (North EU)	Sugar beet / Cyntia	1) 3 Apr 01 2) - 3) 18 Sep 01	Broadcast spray	0.167	308	0.514	20 Jun 01	BBCH 37	Roots Tops	**< 0.02** **< 0.02**	90 90	LOQ 0.02 mg//kg
			-	-	-	0	-	-	Roots Tops	< 0.02 < 0.02	-	Analytical method: (HPLC/MS-MS) Analysis date: 8 Feb 02 Frozen storage period : 5 months.

Active substance (common name)	:	Lenacil	Commercial Product (name)	:	Venzar
Crop/crop group	:	Sugar beet / root and tuber vegetables	Producer of commercial product	:	Schirm GmbH
Responsible body for reporting(name, address)	:	Schirm GmbH Geschwister-Scholl-Str. 127, 39218 Schönebeck, Germany	Indoor/Glasshouse/Outdoor	:	Outdoor
Country	:	Portugal	Other active substance in the		
Content of active substance (g/kg or g/L)	:	Lenacil 800 g/kg	formulation (common name and content)	:	None
Formulation (eg WP)	:	WP	Residues calculated as	:	Lenacil

1	2	3	4	5			6	7	8	9	10	11
Report no. Location (region)	Commodity/variety	Date of: 1. Sowing or planting 2. Flowering 3. Harvest	Method of treatment	Application rate per treatment			Dates of treatment(s) or no. of treatment(s) and last date	Growth stage at last treatment or date	Portion analysed	Residues (mg/kg)	PHI (days)	Remarks
				kg a.s./hL	Water (L/ha)	kg a.s./ha						
20011048/E2-FPSB Vila Franca Xira, P02N001R, Portugal (South EU)	Sugar beet / Hipolita	1) 3 Apr 02 2) - 3) 19 Aug 02	Broadcast spray	0.125	416	0.520	20 Jun 02	BBCH 38	Roots Tops	**< 0.02** **0.03**	60 60	LOQ 0.02 mg//kg
			-	-	-	0	-	-	Roots Tops	< 0.02 < 0.02	-	Analytical method: (HPLC/MS-MS) Analysis date: 18 Sep 02 Frozen storage period : 1 months.

Active substance (common name)	: Lenacil	Commercial Product (name)	: Venzar
Crop/crop group	: Sugar beet / root and tuber vegetables	Producer of commercial product	: Schirm GmbH
Responsible body for reporting(name, address)	: Schirm GmbH Geschwister-Scholl-Str. 127, 39218 Schönebeck, Germany	Indoor/Glasshouse/Outdoor	: Outdoor
Country	: Spain	Other active substance in the	
Content of active substance (g/kg or g/L)	: Lenacil 800 g/kg	formulation (common name and content)	: None
Formulation (eg WP)	: WP	Residues calculated as	: Lenacil

1	2	3	4	5			6	7	8	9	10	11
Report no. Location (region)	Commodity/variety	Date of: 1. Sowing or planting 2. Flowering 3. Harvest	Method of treatment	Application rate per treatment			Dates of treatment(s) or no. of treatment(s) and last date	Growth stage at last treatment or date	Portion analysed	Residues (mg/kg)	PHI (days)	Remarks
				kg a.s./hL	Water (L/ha)	kg a.s./ha						
688479 Test 1, Suzana (Burgos), Spain (South EU)	Sugar beet / Space	1) 16 Mar 05 2) - 3) 16 Sep 05	Broadcast spray	0.252	200	0.503	19 May 05	BBCH 31	Leaves Leaves Leaves Leaves Roots Tops	19 0.55 0.03 < 0.02 **< 0.02** **< 0.02**	0 14 28 42 120 120	LOQ 0.02 mg//kg Analytical method: 20051414/01-RSG (HPLC/MS-MS) Analysis date: Dec 05 Frozen storage period : 2,5 months.
			-	-	-	0	-	-	Leaves Leaves Leaves Leaves Roots Tops	< 0.02 < 0.02 < 0.02 < 0.02 < 0.02 < 0.02	0 14 28 42 120 120	

Active substance (common name)	: Lenacil	Commercial Product (name)	: Venzar
Crop/crop group	: Sugar beet / root and tuber vegetables	Producer of commercial product	: Schirm GmbH
Responsible body for reporting(name, address)	: Schirm GmbH Geschwister-Scholl-Str. 127, 39218 Schönebeck, Germany	Indoor/Glasshouse/Outdoor	: Outdoor
Country	: Spain	Other active substance in the	
Content of active substance (g/kg or g/L)	: Lenacil 800 g/kg	formulation (common name and content)	: None
Formulation (eg WP)	: WP	Residues calculated as	: Lenacil

1	2	3	4	5			6	7	8	9	10	11
Report no. Location (region)	Commodity/variety	Date of: 1. Sowing or planting 2. Flowering 3. Harvest	Method of treatment	Application rate per treatment			Dates of treatment(s) or no. of treatment(s) and last date	Growth stage at last treatment or date	Portion analysed	Residues (mg/kg)	PHI (days)	Remarks
				kg a.s./hL	Water (L/ha)	kg a.s./ha						
688479 Test 2, Erentxun (Araba), Spain (South EU)	Sugar beet / Idoia	1) 29 Mar 05 2) - 3) 16 Sep 05	Broadcast spray	0.251	203	0.510	2 Jun 05	BBCH 31-32	Roots Tops	**< 0.02** **< 0.02**	106 106	LOQ 0.02 mg//kg
			-	-	-	0	-	-	Roots Tops	< 0.02 < 0.02	106 106	Analytical method: 20051414/01-RSG Analysis date: Dec 05 Frozen storage period : 2,5 months.