Analytical method for tetraniliprole (BCS-CL73507) and its transformation products, BCS-CQ63359, BCS-CR60014, BCS-CR74541, BCS-CU81055, BCS-CT30673, BCS-CU81056, BCS-CY28900, BCS-CY28897, and BCS-CY28906 in water

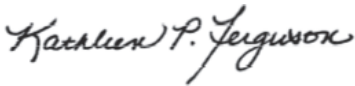|	
Reports:	ECM: EPA MRID No.: 50216523. Williams, J. 2016. In House Laboratory Validation of Analytical Method for the Determination of Tetraniliprole (BCS-CL73507) and its Metabolites: BCS-CQ63359, BCS-CU81055, BCS-CR74541, BCSCR60014, BCS-CU81056, BCS-CT30673, BCS-CY28900, BCS-CY28897 and BCSCY28906 in Water by LC/MS/MS. Report prepared, sponsored, and submitted by Bayer CropScience, Research Triangle Park, North Carolina; 167 pages. Bayer Study/Report No./PSI No.: RAFVP022. Final report issued October 28, 2016.
	ILV: EPA MRID No.: 50170147. Perez, S., M. Marshall. 2016. Independent Laboratory Validation (ILV) of Bayer Method FV-004-W16-01 for the Determination of Residues of Tetraniliprole (BCS-CL73507) and its Metabolites BCS-CQ63359, BCS-CU81055, BCS-CR74541, BCS-CR60014, BCS-CU81056, BCS-CT30673, BCS-CY28900, BCS-CY28897 and BCS-CY28906 in Water using LC-MS/MS. Report prepared by ADPEN Laboratories, Inc., Jacksonville, Florida, and sponsored and submitted by Bayer CropScience, Research Triangle Park, North Carolina; 415 pages. Study ID: RAFVP018. ADPEN Study No.: 16I0104. Final report issued October 28, 2016; amended reported completed November 16, 2016.
Document No.:	MRIDs 50216523 & 50170147
Guideline:	850.6100
Statements:	ECM: The study was conducted in compliance with USEPA FIFRA (40 CFR 160) Good Laboratory Practice (GLP) standards (p. 3 of MRID 50216523). Signed and dated Data Confidentiality, GLP, and Quality Assurance statements were provided (pp. 2-4). The statement of authenticity was not included.
	ILV: The study was conducted in compliance with USEPA FIFRA (40 CFR 160) GLP standards (p. 3 of MRID 50170147). Signed and dated Data Confidentiality, GLP and Quality Assurance statements were provided (pp. 2-4). The statement of authenticity was not included. The reasons for amendments was provided (p. 3).
Classification:	This analytical method is classified as Acceptable. ILV performance data for BCS-CY28897 in the tap water matrix was not satisfactory at 10×LOQ. It could not be determined if the ILV was provided with the most difficult matrices with which to validate the method. ILV recoveries results of BCS-CR60014 and BCS-CT30673 were corrected for residues found in the controls.
PC Code:	090097
EFED Final Reviewer:	Ideliz Negrón-Encarnación, Chemist — Signature: _(signature)_ Date: 7/23/18 — Digitally signed by IDELIZ NEGRON-ENCARNACION Date: 2018.07.23 11:06:24 -04'00'
CDM/CSS-	Lisa Muto, — Signature: _(signature)_

Dynamac JV Reviewers:	Environmental Scientist	Date:	12/13/17
	Kathleen Ferguson, Ph.D., Environmental Scientist	Signature:	*Kathleen P. Ferguson*
		Date:	12/13/17
Secondary Reviewer:	Maria Papiez	Signature:	*Maria Papiez*
PMRA, Health Canada	Chemistry Evaluation Section	Date:	07/19/18

This Data Evaluation Record may have been altered by the Environmental Fate and Effects Division subsequent to signing by CDM/CSS-Dynamac JV personnel.

Executive Summary

The analytical method, Bayer Method FV-004-W16-01, is designed for the quantitative determination of tetraniliprole (BCS-CL73507) and its transformation products BCS-CQ63359, BCS-CR60014, BCS-CR74541, BCS-CU81055, BCS-CT30673, BCS-CU81056, BCS-CY28900, BCS-CY28897, and BCS-CY28906 in water at the stated LOQ of 0.10 µg/L using HPLC/MS/MS. The LOQ is less than the lowest toxicological level of concern in water for all ten analytes. The ILV validated the method using tap water with the first trial for all analytes with insignificant modifications to the analytical instrumentation; however, it could not be determined if the ILV was provided with the most difficult matrix with which to validate the method. The waters of the ECM and ILV were not characterized; the tap water of the ILV appeared to be the same as that of the ECM. All ILV data regarding repeatability, accuracy, precision, linearity, and specificity were satisfactory for all analytes, except that performance data for BCS-CY28897 in the tap water matrix did not meet OCSPP guidelines requirements for precision and accuracy at 10×LOQ [mean 122% (Q & C)]. ILV recoveries results of BCS-CR60014 and BCS-CT30673 were corrected for residues found in the controls. Based on the quantitation ion analysis, all ECM data regarding repeatability, accuracy, precision, linearity, and specificity were satisfactory for all analytes. In the ECM confirmation ion analyses, performance data for BCS-CT30673 and BCS-CU81056 were unsatisfactory for one or both matrices; however, a confirmatory method is not usually required when LC/MS and GC/MS is the primary method.

Table 1. Analytical Method Summary

Analyte(s) by Pesticide	MRID		EPA Review	Matrix	Method Date (dd/mm/yyyy)	Registrant	Analysis	Limit of Quantitation (LOQ)
	Environmental Chemistry Method	Independent Laboratory Validation						
Tetraniliprole (BCS-CL73507)	50216523[1]	50170147[2]		Water	28/10/2016 (In House Validation) 07/09/2016 (Method)[3]	Bayer CropScience	LC/MS/MS	0.10 µg/L
BCS-CQ63359								
BCS-CR60014								
BCS-CR74541								
BCS-CU81055								
BCS-CT30673								
BCS-CU81056								
BCS-CY28900								
BCS-CY28897								
BCS-CY28906								

1 In the ECM, surface water was obtained from study MELNN028 and tap water was obtained from Bayer CropScience, Building 108, Lab 1823; however, the water matrices were not characterized, and the sources were not reported (p. 12 of MRID 50216523).

2 In the ILV, the drinking (tap) water was obtained from Bayer CropScience, Lab 1823, and not characterized (p. 21 of MRID 50170147).

3 Bayer Method No.: FV-004-W16-01 (Appendix 5, pp. 127-167 of MRID 50216523).

I. Principle of the Method

Water samples (4.60 ± 0.05 mL) were placed in 7-mL scintillation vials and fortified, if necessary (p. 12; Appendix 5, p. 139 of MRID 50216523). Internal standard (0.010 mL of 0.50 µg/mL) was added, and the sample was diluted to *ca*. 5.0 mL with 1.25% acetic acid in acetonitrile. After vortex-mixing, an aliquot of the sample was analyzed by LC/MS/MS.

Samples were analyzed for tetraniliprole, BCS-CQ63359, BCS-CR60014, BCS-CR74541, BCS-CU81055, BCS-CT30673, BCS-CU81056, BCS-CY28900, and BCS-CY28897 using a Shimadzu LC 20AD HPLC coupled with an ABSciex 6500LC-MS/MS (Positive polarity; pp. 12-15; Appendix 5, pp. 139-143 of MRID 50216523). The following LC conditions were used: Phenomenex Luna C18(2)-HST (50 mm x 2.0 mm, 2.5 µm particle size; column temperature 40°C), mobile phase of (A) 0.1% aqueous formic acid and (B) 0.1% formic acid in acetonitrile [mobile gradient phase of percent A:B (v:v) at 0.00-0.10 min. 90:10, 4.00-5.00 min. 5:95, 5.01-5.10 min. 90:10], injection volume of 25 µL, and Multiple Reaction Monitoring (MRM) with Electrospray ionization (ESI; 400°C) in positive mode.

Samples were analyzed for BCS-CY28906 using a Shimadzu LC 20AD HPLC coupled with an ABSciex 5500LC-MS/MS (Negative polarity; pp. 12-15; Appendix 5, pp. 139-143 of MRID 50216523). The following LC conditions were used: Phenomenex Luna C18(2)-HST (50 mm x 2.0 mm 2.5 µm particle size; column temperature 40°C), mobile phase of (A) 0.1% aqueous formic acid and (B) 0.1% formic acid in acetonitrile [mobile gradient phase of percent A:B (v:v) at 0.00-0.50 min. 85:15, 1.50-2.00 min. 50:50, 3.50 min. 40:60, 4.00-5.00 min. 5:95, 5.01-5.10 min. 85:15], injection volume of 25 µL, and Multiple Reaction Monitoring (MRM) with Electrospray ionization (ESI; 500°C) in negative mode.

Retention times	Two separate analyses: 1. Electrospray ionization – negative polarity BCS-CY28906 approximately 1.6 min BCS-CU81055 IS approx. 2.0 min (serves as IS for BCS-CY28906) 2. Electrospray ionization – positive polarity BCS-CR60014 approx. 2.2 min BCS-CU81055 approx. 2.3 min BCS-CY28897 approx. 2.4 min BCS-CR74541 approx. 2.4 min BCS-CU81056 approx. 2.6 min BCS-CY28900 approx. 2.6 min BCS-CL73507 approx. 2.7 min BCS-CT30673 approx. 2.8 min BCS-CQ63359 approx. 3.2 min

Two MRM transitions were monitored, one for quantitation and a second for confirmatory purposes:

Analyte	Q1 Mass [m/z]	Q3 Mass [m/z]	
BCS-CL73507	545.1	356.0	quantitation
	545.1	376.0	confirmation
BCS-CQ63359	527.0	389.0	quantitation
	527.0	374.1	confirmation
BCS-CR60014	563.0	356.0	quantitation
	563.0	394.0	confirmation
BCS-CR74541	564.0	356.0	quantitation
	564.0	395.0	confirmation
BCS-CU81055	550.0	395.1	quantitation
	555.0	356.0	confirmation
BCS-CT30673	545.8	408.1	quantitation
	545.8	267.0	confirmation
BCS-CU81056	532.0	394.1	quantitation
	532.0	366.0	confirmation
BCS-CY28900	509.0	371.0	quantitation
	509.0	342.0	confirmation
BCS-CY28897	509.0	371.0	quantitation
	509.0	481.1	confirmation
BCS-CY28906	261.0	136.0	quantitation
	261.0	108.0	confirmation

The ILV performed the ECM methods for each analyte as written, except that different analytical instrumentation was used (pp. 22-25; Appendix 1, p. 295 of MRID 50170147). For tetraniliprole, BCS-CQ63359, BCS-CR60014, BCS-CR74541, BCS-CU81055, BCS-CT30673, BCS-CU81056, BCS-CY28900, and BCS-CY28897, the LC/MS/MS analysis was performed using an Agilent 1290 UPLC coupled to an AB Sciex Triple Quad API 6500 LC/MS/MS system. The chromatographic parameters were the same as those of the ECM, except injection volume was 30 µL. For BCS-CY28906, the LC/MS/MS analysis was performed using an Agilent 1290 UPLC coupled to an AB

Sciex Triple Quad API 5500 LC/MS/MS system. The chromatographic parameters were the same as those of the ECM, except injection volume was 40 µL.

Retention times	Two separate analyses: 1. Electrospray ionization – negative polarity BCS-CY28906 approximately 2.37 min 2. Electrospray ionization – positive polarity BCS-CR60014 approx. 2.13 min BCS-CU81055 approx. 2.23 min BCS-CY28897 approx. 2.29 min BCS-CR74541 approx. 2.32 min BCS-CU81056 approx. 2.47 min BCS-CY28900 approx. 2.51 min BCS-CL73507 approx. 2.55 min BCS-CT30673 approx. 2.60 min BCS-CQ63359 approx. 2.95 min

Retention times estimated from Figures 11a/b-20a/b, pp. 73-212 of MRID 50170147.

Two MRM transitions were monitored, one for quantitation and a second for confirmatory purposes, for each analyte. These were the same as those of the ECM (m/z $\pm$ 0.1, except for BCS-CY28906 which was m/z $\pm$ 1.0):

Analyte	Q1 Mass [m/z]	Q3 Mass [m/z]	
BCS-CL73507	545.1	356.0	quantitation
	545.1	376.0	confirmation
BCS-CQ63359	527.0	389.0	quantitation
	527.0	374.0	confirmation
BCS-CR60014	563.0	356.0	quantitation
	563.0	394.0	confirmation
BCS-CR74541	564.0	356.0	quantitation
	564.0	395.0	confirmation
BCS-CU81055	550.0	395.0	quantitation
	555.0	356.0	confirmation
BCS-CT30673	545.8	408.0	quantitation
	545.8	267.1	confirmation
BCS-CU81056	532.0	394.0	quantitation
	532.0	366.0	confirmation
BCS-CY28900	509.0	371.1	quantitation
	509.0	342.1	confirmation
BCS-CY28897	509.0	371.1	quantitation
	509.0	481.1	confirmation
BCS-CY28906	261.0	136.9	quantitation
	261.0	108.8	confirmation

In the ECM and ILV, the Limit of Quantification (LOQ) was 0.1 µg/L for tetraniliprole (BCS-CL73507) and its transformation products, BCS-CQ63359, BCS-CR60014, BCS-CR74541, BCS-CU81055, BCS-CT30673, BCS-CU81056, BCS-CY28900, BCS-CY28897, and BCS-CY28906

(pp. 9, 20-21; Table 3, p. 26 of MRID 50216523; pp. 15, 28; Appendix 2, p. 302 of MRID 50170147). The method Limit of Detection (LOD) was not reported in the ECM and ILV; calculated LODs ranged 0.011-0.028 µg/L in the ECM for all analytes. LODs were not calculated in the ILV.

II. Recovery Findings

ECM (MRID 50216523): For the quantitation ion transition analysis, mean recoveries and relative standard deviations (RSDs) were within guidelines (mean 70-120%; RSD ≤20%) for analysis of tetraniliprole (BCS-CL73507), BCS-CQ63359, BCS-CR60014, BCS-CR74541, BCS-CU81055, BCS-CT30673, BCS-CU81056, BCS-CY28900, BCS-CY28897, and BCS-CY28906 at fortification levels of 0.10 µg/L (LOQ) and 1.00 µg/L (10×LOQ) in two water matrices (Tables 1-2, pp. 24-25 of MRID 50216523; DER Attachment 2). For the confirmation ion transition analysis, mean recoveries and RSDs were within guidelines for analysis of tetraniliprole, BCS-CQ63359, BCS-CR60014, BCS-CR74541, BCS-CU81055, BCS-CT30673, BCS-CU81056, BCS-CY28900, BCS-CY28897, and BCS-CY28906 at fortification levels of 0.10 µg/L (LOQ) and 1.00 µg/L (10×LOQ) in two water matrices, except for the LOQ analyses in surface water of BCS-CU81056 (RSD 42%) and BCS-CT30673 (RSD 22%) and the LOQ analysis in tap water of BCS-CU81056 (RSD 26%). The unacceptable results of the confirmation ion analysis did not affect the validity of the method since a confirmatory method is not usually required when LC/MS and GC/MS is the primary method. Recovery results of the quantitation ion were comparable to those of the confirmation ion for all analytes/matrices/fortifications, except for the LOQ analyses of BCS-CT30673 (surface water) and BCS-CU81056 (both water matrices). Relative Standard Deviations (RSDs) were reviewer-calculated based on data (means and standard deviations) provided in the study report since these values were not provided by the study authors. The surface water was obtained from study MELNN028 and tap water was obtained from Bayer CropScience, Building 108, Lab 1823; however, the water matrices were not characterized, and the sources were not reported (p. 12).

ILV (MRID 50170147): Mean recoveries and RSDs were within guidelines for analysis of tetraniliprole, BCS-CQ63359, BCS-CR60014, BCS-CR74541, BCS-CU81055, BCS-CT30673, BCS-CU81056, BCS-CY28900, BCS-CY28897, and BCS-CY28906 at fortification levels of 0.10 µg/L (LOQ) and 1.00 µg/L (10×LOQ) in one water matrix, except for the 10×LOQ analysis of BCS-CY28897 [mean 122% (Q & C; Tables 2a/b-11a/b, pp. 32-51]. Recovery results of the quantitation ion were comparable to those of the confirmation ion. Recovery results of BCS-CR60014 and BCS-CT30673 were corrected for residues found in the controls (Tables 2a/b-11a/b, pp. 32-51; Appendix 5, pp. 392-393). The drinking (tap) water was obtained from Bayer CropScience, Lab 1823, and not characterized (p. 21). The method was validated with the first trial for all analytes with insignificant modifications to the analytical instrumentation and parameters (pp. 15, 22-26).

Table 2. Initial Validation Method Recoveries for Tetraniliprole (BCS-CL73507), BCS-CQ63359, BCS-CR60014, BCS-CR74541, BCS-CU81055, BCS-CT30673, BCS-CU81056, BCS-CY28900, BCS-CY28897, and BCS-CY28906 in Water[1,2]

Analyte	Fortification Level (µg/L)	Number of Tests	Recovery Range (%)	Mean Recovery (%)	Standard Deviation (%)	Relative Standard Deviation (%)[3]
			Surface Water			
			Quantitation ion			
Tetraniliprole (BCS-CL73507)	0.100 (LOQ)	7	76-89	83	4.3	5
	1.00	5	87-101	93	5.6	6
BCS-CQ63359	0.100 (LOQ)	7	86-106	93	6.4	7
	1.00	5	98-116	108	6.8	6
BCS-CU81055	0.100 (LOQ)	7	73-92	83	6.3	8
	1.00	5	84-99	91	6.4	7
BCS-CR74541	0.100 (LOQ)	7	76-105	91	10.4	11
	1.00	5	84-98	93	5.1	5
BCS-CR60014	0.100 (LOQ)	7	79-97	87	5.6	6
	1.00	5	87-96	91	3.9	4
BCS-CU81056	0.100 (LOQ)	7	90-106	100	6.3	6
	1.00	5	102-118	108	5.9	5
BCS-CT30673	0.100 (LOQ)	7	77-108	87	10.5	12
	1.00	5	98-106	102	2.9	3
BCS-CY28900	0.100 (LOQ)	7	64-83	76	7.1	9
	1.00	5	84-94	88	4.1	5
BCS-CY28897	0.100 (LOQ)	7	59-84	73	8.0	11
	1.00	5	78-100	89	8.2	9
BCS-CY28906	0.100 (LOQ)	7	77-96	86	6.6	8
	1.00	5	84-96	89	4.3	5
			Confirmation ion			
Tetraniliprole (BCS-CL73507)	0.100 (LOQ)	7	73-85	80	5.3	7
	1.00	5	89-96	92	2.7	3
BCS-CQ63359	0.100 (LOQ)	7	72-111	91	15.0	16
	1.00	5	94-116	107	8.5	8
BCS-CU81055	0.100 (LOQ)	7	68-95	79	9.6	12
	1.00	5	91-102	95	4.8	5
BCS-CR74541	0.100 (LOQ)	7	63-87	81	8.2	10
	1.00	5	81-102	93	8.7	9
BCS-CR60014	0.100 (LOQ)	7	74-91	81	6.3	8
	1.00	5	84-93	89	3.7	4
BCS-CU81056	0.100 (LOQ)	7	40-167	93	39.2	**42**
	1.00	5	104-114	108	3.8	4
BCS-CT30673	0.100 (LOQ)	7	66-124	92	19.8	**22**
	1.00	5	94-110	103	7.0	7
BCS-CY28900	0.100 (LOQ)	7	67-78	74	6.1	8
	1.00	5	84-92	88	3.5	4
BCS-CY28897	0.100 (LOQ)	7	80-101	92	7.6	8
	1.00	5	85-110	100	10.0	10
BCS-CY28906	0.100 (LOQ)	7	78-101	90	8.7	10
	1.00	5	84-101	91	6.2	7

Analyte	Fortification Level (µg/L)	Number of Tests	Recovery Range (%)	Mean Recovery (%)	Standard Deviation (%)	Relative Standard Deviation (%)[3]
			Drinking (Tap) Water			
			Quantitation ion			
Tetraniliprole (BCS-CL73507)	0.100 (LOQ)	7	77-94	87	6.3	7
	1.00	5	87-92	90	1.9	2
BCS-CQ63359	0.100 (LOQ)	7	82-100	92	8.0	9
	1.00	5	84-95	89	4.7	5
BCS-CU81055	0.100 (LOQ)	7	67-97	82	10.2	12
	1.00	5	87-95	90	3.5	4
BCS-CR74541	0.100 (LOQ)	7	80-96	86	6.6	8
	1.00	5	85-93	89	2.7	3
BCS-CR60014	0.100 (LOQ)	7	86-95	90	2.9	3
	1.00	5	90-92	91	0.5	1
BCS-CU81056	0.100 (LOQ)	7	71-103	87	12.4	14
	1.00	5	89-99	92	4.0	4
BCS-CT30673	0.100 (LOQ)	7	70-91	83	7.0	8
	1.00	5	84-92	88	3.2	4
BCS-CY28900	0.100 (LOQ)	7	72-90	82	6.3	8
	1.00	5	83-88	86	2.3	3
BCS-CY28897	0.100 (LOQ)	7	86-95	90	3.8	4
	1.00	5	85-92	89	2.9	3
BCS-CY28906	0.100 (LOQ)	7	76-91	84	5.2	6
	1.00	5	87-93	90	2.7	3
			Confirmation ion			
Tetraniliprole (BCS-CL73507)	0.100 (LOQ)	7	85-104	92	6.9	8
	1.00	5	87-95	91	2.8	3
BCS-CQ63359	0.100 (LOQ)	7	76-106	95	11.0	12
	1.00	5	87-97	91	3.8	4
BCS-CU81055	0.100 (LOQ)	7	85-104	91	8.1	9
	1.00	5	87-94	90	2.8	3
BCS-CR74541	0.100 (LOQ)	7	81-103	89	8.1	9
	1.00	5	87-95	90	2.9	3
BCS-CR60014	0.100 (LOQ)	7	83-98	89	5.7	6
	1.00	5	88-93	91	1.8	2
BCS-CU81056	0.100 (LOQ)	7	61-125	84	22.2	**26**
	1.00	5	82-98	88	7.6	9
BCS-CT30673	0.100 (LOQ)	7	74-99	89	10.1	11
	1.00	5	84-89	88	2.3	3
BCS-CY28900	0.100 (LOQ)	7	68-101	86	12.6	15
	1.00	5	80-92	86	5.7	7
BCS-CY28897	0.100 (LOQ)	7	79-91	86	4.3	5
	1.00	5	83-90	87	2.9	3
BCS-CY28906	0.100 (LOQ)	7	79-98	87	6.6	8
	1.00	5	84-93	89	3.1	3

Data (uncorrected recovery results; pp. 16-17) were obtained from Tables 1-2, pp. 24-25 of MRID 50216523 and DER Attachment 2.

1 The surface water was obtained from study MELNN028 and tap water was obtained from Bayer CropScience, Building 108, Lab 1823; however, the water matrices were not characterized, and the sources were not reported (p.

12).

2 Two ion pair transition were monitored for each analyte (see above).

3 Relative Standard Deviations (RSDs) were reviewer-calculated based on data (means and standard deviations) provided in the study report since these values were not provided by the study authors. Rules of significant figures were followed.

Table 3. Independent Validation Method Recoveries for Tetraniliprole (BCS-CL73507), BCS-CQ63359, BCS-CR60014, BCS-CR74541, BCS-CU81055, BCS-CT30673 BCS-CU81056, BCS-CY28900, BCS-CY28897, and BCS-CY28906 in Water[1,2]

Analyte	Fortification Level (µg/L)	Number of Tests	Recovery Range (%)	Mean Recovery (%)	Standard Deviation (%)	Relative Standard Deviation (%)
			Drinking (Tap) Water			
			Quantitation ion			
Tetraniliprole (BCS-CL73507)	0.100 (LOQ)	5	94-100	96	2.7	2.9
	1.00	5	94-109	102	5.6	5.5
BCS-CQ63359	0.100 (LOQ)	5	86-101	95	5.8	6.1
	1.00	5	88-101	92	5.7	6.2
BCS-CU81055	0.100 (LOQ)	5	91-101	97	3.8	3.9
	1.00	5	97-105	102	3.7	3.7
BCS-CR74541	0.100 (LOQ)	5	87-103	96	5.9	6.1
	1.00	5	96-106	100	3.7	3.7
BCS-CR60014	0.100 (LOQ)	5	99-106	103	2.5	2.4
	1.00	5	93-105	99	5.1	5.2
BCS-CU81056	0.100 (LOQ)	5	95-106	100	5.0	5.0
	1.00	5	97-104	99	2.8	2.8
BCS-CT30673	0.100 (LOQ)	5	89-105	96	6.0	6.3
	1.00	5	91-108	96	6.6	6.9
BCS-CY28900	0.100 (LOQ)	5	83-115	103	12.7	12.4
	1.00	5	112-126	117	6.3	5.4
BCS-CY28897	0.100 (LOQ)	5	92-116	105	10.3	9.7
	1.00	5	115-132	**122**	6.2	5.1
BCS-CY28906	0.100 (LOQ)	5	93-97	95	1.9	2.0
	1.00	5	93-106	100	6.0	6.0
			Confirmation ion			
Tetraniliprole (BCS-CL73507)	0.100 (LOQ)	5	96-107	102	4.3	4.2
	1.00	5	93-104	98	3.9	4.0
BCS-CQ63359	0.100 (LOQ)	5	92-109	101	6.7	6.7
	1.00	5	97-109	102	4.6	4.5
BCS-CU81055	0.100 (LOQ)	5	90-110	97	7.9	8.2
	1.00	5	99-125	109	9.7	8.8
BCS-CR74541	0.100 (LOQ)	5	91-110	98	7.1	7.2
	1.00	5	96-105	101	3.5	3.5
BCS-CR60014	0.100 (LOQ)	5	92-109	101	6.6	6.5
	1.00	5	91-108	101	7.4	7.4
BCS-CU81056	0.100 (LOQ)	5	66-112	87	16.8	19.3
	1.00	5	79-108	96	12.4	13.0
BCS-CT30673	0.100 (LOQ)	5	75-117	91	15.8	17.4

Analyte	Fortification Level (µg/L)	Number of Tests	Recovery Range (%)	Mean Recovery (%)	Standard Deviation (%)	Relative Standard Deviation (%)
	1.00	5	90-116	102	11.9	11.6
BCS-CY28900	0.100 (LOQ)	5	78-114	96	16.2	16.8
	1.00	5	116-122	118	2.3	2.0
BCS-CY28897	0.100 (LOQ)	5	94-111	103	7.4	7.3
	1.00	5	115-129	**122**	5.2	4.3
BCS-CY28906	0.100 (LOQ)	5	93-97	95	1.7	1.8
	1.00	5	90-105	97	6.6	6.9

Data (uncorrected recovery results; Appendix 5, pp. 392-393, except for BCS-CR60014 and BCS-CT30673) were obtained from Tables 2a/b-11a/b, pp. 32-51 of MRID 50170147.
1 The drinking (tap) water was obtained from Bayer CropScience, Lab 1823, and not characterized (p. 21).
2 Two ion pair transition were monitored for each analyte (see above).

III. Method Characteristics

In the ECM and ILV, the LOQ was 0.1 µg/L for tetraniliprole (BCS-CL73507) and its transformation products, BCS-CQ63359, BCS-CR60014, BCS-CR74541, BCS-CU81055, BCS-CT30673, BCS-CU81056, BCS-CY28900, BCS-CY28897, and BCS-CY28906 (pp. 9, 20-21; Table 3, p. 26 of MRID 50216523; pp. 15, 28; Appendix 2, p. 302 of MRID 50170147). In the ECM, the LOQ was defined as the lowest fortification level experimentally providing a mean recovery between 70 and 120% with a relative standard deviation of ≤ 20%, provided that the blank values were below 30% at this level. The LOQ was reported in the ILV from the ECM. No calculations or comparisons to background levels were reported to support the method LOQ. The method LOD was not reported in the ECM and ILV. The LOD was calculated in the ECM using the following equation:

MDL (calculated) = $(S \times t_{0.99})$,

Where MDL is the method detection limit, $t_{0.99}$ equaled 2.650 (the one-tailed t-statistic at the 99% confidence level for n-1 replicates) and S equaled the standard deviation of n samples fortified at the LOQ. Calculated LODs ranged 0.011-0.028 µg/L in the ECM for all analytes. LODs were not calculated in the ILV.

Table 4a. Method Characteristics

Analyte		Tetraniliprole (BCS-CL73507)	BCS-CQ63359	BCS-CR60014	BCS-CR74541	BCS-CU81055
Limit of Quantitation (LOQ)		0.10 µg/L				
Limit of Detection (LOD)	ECM	Not reported (method) 0.011-0.023 µg/L (calculated)				
	ILV	Not reported				
Linearity (calibration curve r² and concentration range)	ECM[1]	$r^2 = 0.9996$ (Q) $r^2 = 0.9990$ (C)	$r^2 = 0.9994$ (Q) $r^2 = 0.9986$ (C)	$r^2 = 0.9998$ (Q & C)	$r^2 = 0.9990$ (Q) $r^2 = 0.9994$ (C)	$r^2 = 0.9994$ (Q) $r^2 = 0.9996$ (C)
	ILV	$r^2 = 0.9992$ (Q) $r^2 = 0.9996$ (C)	$r^2 = 0.9988$ (Q) $r^2 = 0.9994$ (C)	$r^2 = 0.9998$ (Q) $r^2 = 0.9996$ (C)	$r^2 = 0.9998$ (Q & C)	$r^2 = 0.9996$ (Q) $r^2 = 0.9992$ (C)
	Range:	0.025-10 ng/mL				
Repeatable	ECM[2,3] Q/C	Yes at the LOQ and 10×LOQ in surface and tap water matrices.				
	ILV[5,6] Q/C	Yes at the LOQ and 10×LOQ in tap water matrix.[7]				
Reproducible		Yes at the LOQ and 10×LOQ.				
Specific	ECM	Yes. Matrix interferences were <14% of the LOQ (Q) and <16% of the LOQ (C; based on calculated concentration). Minor baseline noise interfered with peak attenuation and integration in many chromatograms; baseline noise interference was more notable in the C ion chromatogram.				
	ILV	Yes. No matrix interferences were observed.				
		Some minor baseline noise interfered with peak attenuation and integration in the C ion chromatogram.				

Data were obtained from pp. 9, 20-21; Table 3, p. 26 (LOD/LOQ); Tables 1-2, pp. 24-25 (recovery results); Appendix 1, Figures 1-20, pp. 31-50 (calibration curves); Appendix 2, pp. 70-117 (chromatograms) of MRID 50216523; pp. 15, 28; Appendix 2, p. 302 (LOQ/LOD); Tables 2a/b-11a/b, pp. 32-51 (recovery results); Figures 21a/b-60a/b, pp. 213-292 (chromatograms); Appendix 6, pp. 395-414 (calibration curves) of MRID 50170147; DER Attachment 2. Q = Quantitation ion transition; C = Confirmation ion transition.

1 Correlation coefficients (r²) were reviewer-calculated based on r values (1/x weighted linear regression analysis) reported in the study report; solvent standards were used (Appendix 1, Figures 1-20, pp. 31-50 of MRID 50216523; DER Attachment 2).

2 In the ECM, surface water was obtained from study MELNN028 and tap water was obtained from Bayer CropScience, Building 108, Lab 1823; however, the water matrices were not characterized, and the sources were not reported (p. 12 of MRID 50216523).

3 Relative Standard Deviations (RSDs) were reviewer-calculated based on data (means and standard deviations) provided in the study report since these values were not provided by the study authors. Rules of significant figures were followed.

4 A confirmatory method is not usually required when LC/MS and GC/MS is the primary method.

5 In the ILV, the drinking (tap) water was obtained from Bayer CropScience, Lab 1823, and not characterized (p. 21 of MRID 50170147).

6 The ILV validated the method with the first trial for all analytes with insignificant modifications to the analytical instrumentation and parameters (pp. 15, 22-26 of MRID 50170147).

7 Recoveries of BCS-CR60014 were corrected for minor residues (1% of the LOQ) found in the control samples (Table 6a, p. 40; Appendix 5, pp. 392-393 of MRID 50170147).

Table 4b. Method Characteristics

Analyte			BCS-CU81056	BCS-CT30673	BCS-CY28900	BCS-CY28897	BCS-CY28906
Limit of Quantitation (LOQ)			0.10 µg/L				
Limit of Detection (LOD)	ECM		Not reported (method) 0.016-0.028 µg/L (calculated)				
	ILV		Not reported				
Linearity (calibration curve r² and concentration range)	ECM[1]		r² = 0.9988 (Q) r² = 0.9972 (C)	r² = 0.9994 (Q & C)	r² = 0.9974 (Q) r² = 0.9970 (C)	r² = 0.9994 (Q & C)	r² = 0.9958 (Q) r² = 0.9966 (C)
	ILV		r² = 0.9990 (Q) r² = 0.9994 (C)	r² = 0.9994 (Q) r² = 0.9990 (C)	r² = 0.9990 (Q) r² = 0.9986 (C)	r² = 0.9994 (Q) r² = 0.9992 (C)	r² = 0.9998 (Q & C)
	Range:		0.025-10 ng/mL				
Repeatable		Q	Yes at the LOQ and 10×LOQ in surface and tap water matrices.				
	ECM[2,3]	C[4]	Yes at 10×LOQ, but No at the LOQ in surface (RSD 42%) and tap (RSD 26%) water matrices.	Yes at the LOQ and 10×LOQ in tap water matrix. Yes at 10×LOQ, but No at the LOQ (RSD 22%) in surface water matrix.	Yes at the LOQ and 10×LOQ in surface and tap water matrices.		
	ILV[5,6]	Q/C	Yes at the LOQ and 10×LOQ (based on Q ion results).	Yes at the LOQ and 10×LOQ in tap water matrix.		Yes at the LOQ, but No at 10×LOQ (means 122%) in tap water matrix.	Yes at the LOQ and 10×LOQ in tap water matrix.
Reproducible			Yes at the LOQ and 10×LOQ (based on Q ion results).	Yes at the LOQ and 10×LOQ.[7]		Yes at the LOQ, but No at 10×LOQ.	Yes at the LOQ and 10×LOQ
Specific	ECM		Yes. Matrix interferences were <14% of the LOQ (Q) and <16% of the LOQ (C; based on calculated concentration). Minor baseline noise interfered with peak attenuation and integration in many chromatograms; baseline noise interference was more notable in the C ion chromatogram.				
	ILV		Yes. No matrix interferences were observed. Some minor baseline noise interfered with peak attenuation and integration in the C ion chromatogram.				

Data were obtained from pp. 9, 20–21; Table 3, p. 26 (LOD/LOQ); Tables 1-2, pp. 24-25 (recovery results); Appendix 1, Figures 1-20, pp. 31-50 (calibration curves); Appendix 2, pp. 70-117 (chromatograms) of MRID 50216523; pp. 15, 28; Appendix 2, p. 302 (LOQ/LOD); Tables 2a/b-11a/b, pp. 32-51 (ILV recovery results); Figures 21a/b-60a/b, pp. 213–292 (ILV chromatograms); Appendix 6, pp. 395–414 (ILV calibration curves) of MRID 50170147; DER Attachment 2. Q = Quantitation ion transition; C = Confirmation ion transition.

1 Correlation coefficients (r²) were reviewer-calculated based on r values (1/x weighted linear regression analysis) reported in the study report; solvent standards were used (Appendix 1, Figures 1-20, pp. 31-50 of MRID 50216523; DER Attachment 2).

2 In the ECM, surface water was obtained from study MELNN028 and tap water was obtained from Bayer CropScience, Building 108, Lab 1823; however, the water matrices were not characterized, and the sources were not reported (p. 12 of MRID 50216523).

3 Relative Standard Deviations (RSDs) were reviewer-calculated based on data (means and standard deviations) provided in the study report since these values were not provided by the study authors. Rules of significant figures were followed.

4 A confirmatory method is not usually required when LC/MS and GC/MS is the primary method.

5 In the ILV, the drinking (tap) water was obtained from Bayer CropScience, Lab 1823, and not characterized (p. 21 of MRID 50170147).

6 The ILV validated the method with the first trial for all analytes with insignificant modifications to the analytical instrumentation and parameters (pp. 15, 22-26 of MRID 50170147).

7 Recoveries of BCS-CT30673 were corrected from minor residues (<5% of the LOQ) found in the control samples (Tables 8a-8b, pp. 44-45; Appendix 5, pp. 392-393 of MRID 50170147).

IV. Method Deficiencies and Reviewer's Comments

1.   The Analytical Method Bayer Method No. FV-004-W16-01 was provided in Appendix 5 of
        MRID 50216523 and Appendix 2 of MRID 50170147:

        Williams, J. 2016. An Analytical Method for the Determination of Residues of
        Tetraniliprole (BCS-CL73507) and its Metabolites BCS-CQ63359, BCS-CU81055, BCS-
        CR74541, BCSCR60014, BCS-CU81056, BCS-CT30673, BCS-CY28900, BCS-CY28897
        and BCSCY28906 in Water Using LC/MS/MS. Report prepared, sponsored, and submitted
        by Bayer CropScience, Research Triangle Park, North Carolina; 40 pages. Bayer Method
        No.: FV-004-W16-01. Final report issued September 7, 2016.

        MRID 50216526 was provided later in the review process and discussed below.

        Williams, J. 2016. An Analytical Method for the Determination of Residues of
        Tetraniliprole (BCS-CL73507) and its Metabolites BCS-CQ63359, BCS-CU81055, BCS-
        CR74541, BCS-CR60014, BCS-CU81056, BCS-CT30673, BCS-CY28900, BCS-CY28897
        and BCS-CY28906 in Water Using LC/MS/MS. Report prepared, sponsored, and submitted
        by Bayer CropScience, Research Triangle Park, North Carolina; 40 pages. Bayer Method
        No.: FV-004-W16-02. Final report issued November 14, 2016.

        This study (MRID 50216526) was a method only study, as well, which was an updated
        version of the Bayer Method which was provided in in Appendix 5 of MRID 50216523 and
        Appendix 2 of MRID 50170147 (Appendix 5, p. 40 of MRID 50216526). The only changes
        to the analytical parameters which the reviewer noticed was that the ion source gas 1 and 2
        was changed from 70 to 30 for the positive MS analysis; therefore, the reviewer did not
        change the DER citations (Appendix 2, p. 312 of MRID 50170147; Appendix 5, p. 143 of
        MRID 50216523; p. 16 of MRID 50216526).

2.   In the ILV analysis, performance data for BCS-CY28897 in the tap water matrix did not
        meet OCSPP guidelines requirements for precision and accuracy at 10×LOQ [mean 122%
        (Q & C); Tables 2a/b-11a/b, pp. 32-51 of MRID 50170147]. OCSPP Guideline 850.6100
        criteria for precision and accuracy states that means for replicates at each spiking level are
        between 70% and 120%.

3.   It could not be determined if the ILV was provided with the most difficult matrix with which
        to validate the method because the waters of the ECM and ILV were not characterized. The
        tap water of the ILV appeared to be the same as the tap water from the ECM.

4.   In the ILV, recoveries results of BCS-CR60014 and BCS-CT30673 were corrected for
        residues found in the controls (<5% of the LOQ, based on quantified residues; Tables 2a/b-
        11a/b, pp. 32-51; Appendix 5, pp. 392-393 of MRID 50170147).

5.   The communications between the ILV Study Director and method developer were reported
        (p. 28; Appendix 7, p. 415 of MRID 50170147). The ILV reported that communication
        involved the notification of the successful trial.

6.   In the ECM confirmation ion analyses, performance data for BCS-CT30673 (RSD 22%) and
        BCS-CU81056 (RSD 42%) in the surface water matrix did not meet OCSPP guidelines

requirements for precision and accuracy at the LOQ (Tables 21-22, pp. 45-46 of MRID 50216523). ECM confirmation ion performance data for BCS-CT30673 (RSD 26%) also did not meet OCSPP guidelines requirements for precision and accuracy at the LOQ in the tap water matrix.

OCSPP Guideline 850.6100 criteria for precision and accuracy states that means for replicates at each spiking level are between 70% and 120% and RSDs are ≤ 20%; however, a confirmatory method is not usually required when LC/MS and GC/MS is the primary method. Therefore, the unacceptable results of the confirmation ion analysis did not affect the validity of the method.

7.   The determination of the LOQ in the ECM and ILV were not based on scientifically acceptable procedures as defined in 40 CFR Part 136 (pp. 9, 20-21; Table 3, p. 26 of MRID 50216523; pp. 15, 28; Appendix 2, p. 302 of MRID 50170147). In the ECM, the LOQ was defined as the lowest fortification level experimentally providing a mean recovery between 70 and 120% with a relative standard deviation of ≤ 20%, provided that the blank values were below 30% at this level. The LOQ was reported in the ILV from the ECM. No calculations or comparisons to background levels were reported to support the method LOQ. The method LOD was not reported in the ECM and ILV. The LOD was calculated in the ECM using the following equation MDL (calculated) = $(S \times t_{0.99})$, where MDL is the method detection limit, $t_{0.99}$ equaled 2.650 (the one-tailed t-statistic at the 99% confidence level for n-1 replicates) and S equaled the standard deviation of n samples fortified at the LOQ. LODs were not calculated in the ILV.

8.   In the ECM, the storage stability of the final sample extracts and calibration solutions were determined to be up to 7 days at 1-5°C with less than 15% degradation (pp. 21-22; Tables 4-7, pp. 27-30 of MRID 50126523).

9.   In the ILV, the total time required to complete one set of 13 samples was reported as one hour to complete sample extraction (p. 28 of MRID 50170147). LC/MS/MS analyses were run overnight.


V. References

U.S. Environmental Protection Agency. 2012. Ecological Effects Test Guidelines, OCSPP 850.6100, Environmental Chemistry Methods and Associated Independent Laboratory Validation. Office of Chemical Safety and Pollution Prevention, Washington, DC. EPA 712-C-001.

40 CFR Part 136. Appendix B. Definition and Procedure for the Determination of the Method Detection Limit-Revision 1.11, pp. 317-319.

DER ATTACHMENT 1.

BCS-CL73507:
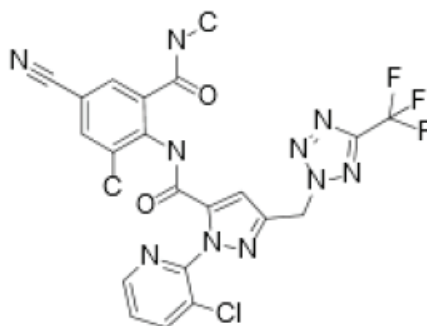
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CAS Number	1229654-66-3
Molecular Formula:	$C_{22}H_{16}ClF_3N_{10}O_2$
Molecular Weight:	544.88 g/mol
Standard No.:	K-2056
Purity:	97.9%
Expiration Date:	07/01/2017
Date of Analysis:	07/01/2014
Storage Conditions:	Frozen
Source:	Bayer CropScience, Frankfurt, Germany

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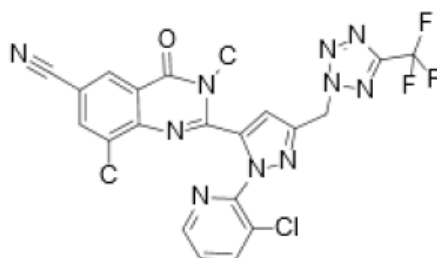
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Molecular Formula:	$C_{22}H_{14}ClF_3N_{10}O$
Molecular Weight:	526.86 g/mol
Standard No.:	K-2118
Purity:	97.7%
Expiration Date:	08/01/2015
Date of Analysis:	08/01/2013
Storage Conditions:	Frozen
Source:	Bayer CropScience, Frankfurt, Germany

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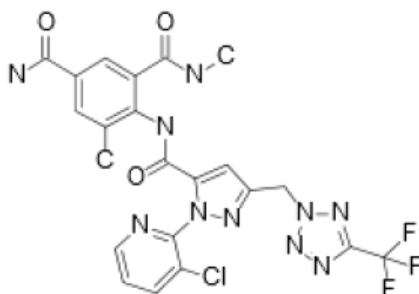
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Molecular Weight:	562.89 g/mol
Standard No.:	K-2090
Purity:	97.9%
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Date of Analysis:	06/20/2013
Storage Conditions:	Frozen
Source:	Bayer CropScience, Frankfurt, Germany

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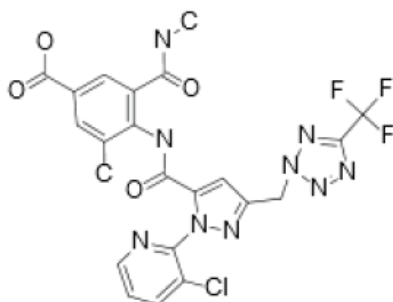
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Molecular Weight:	563.88 g/mol
Standard No.:	K-2117
Purity:	97.2%
Expiration Date:	04/29/2017
Date of Analysis:	05/28/2013
Storage Conditions:	Frozen
Source:	Bayer CropScience, Frankfurt, Germany

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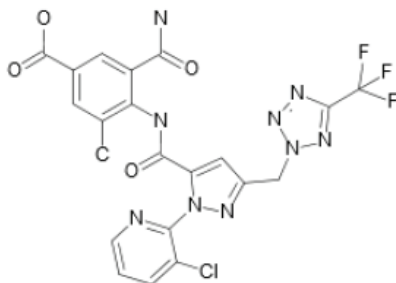
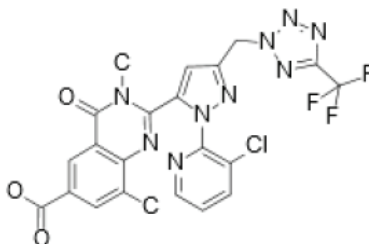|	
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Molecular Formula:	$C_{21}H_{15}ClF_3N_9O_4$
Molecular Weight:	549.85 g/mol
Standard No.:	K-2139
Purity:	0.11% (solution 1.021 mg/mL in 1:1 ACN/water)
Expiration Date:	01/16/2016
Date of Analysis:	01/16/2014
Storage Conditions:	Frozen
Source:	Bayer CropScience, Frankfurt, Germany

BCS-CT30673:

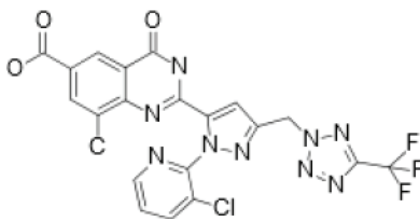|	
CAS Name:	2-[1-(3-chloropyridin-2-yl)-3-{[5-(trifluoromethyl)-2H-tetrazol-2-yl]methyl}-1H-pyrazol-5-yl]-3,8-dimethyl-4-oxo-3,4-dihydroquinazoline-6-carboxylic acid
Molecular Formula:	$C_{22}H_{15}ClF_3N_9O_3$
Molecular Weight:	545.86 g/mol
Standard No.:	K-2066
Purity:	97.9%
Expiration Date:	07/24/2016
Date of Analysis:	07/04/2012
Storage Conditions:	Frozen
Source:	Bayer CropScience, Frankfurt, Germany

BCS-CU81056:

CAS Name:	2-[1-(3-chloropyridin-2-yl)-3-{[5-(trifluoromethyl)-2H-tetrazol-2-yl]methyl}-1H-pyrazol-5-yl]-8-methyl-4-oxo-3,4-dihydroquinaziline-6-carboxylic acid
Molecular Formula:	$C_{21}H_{13}ClF_3N_9O_3$
Molecular Weight:	531.83 g/mol
Standard No.:	K-2091
Purity:	98.3%
Expiration Date:	06/09/2018
Date of Analysis:	06/19/2013
Storage Conditions:	Frozen
Source:	Bayer CropScience, Frankfurt, Germany