

REGISTRATION REPORT

Part B

Section 5

Analytical Methods

Detailed summary of the risk assessment

Product code: **Nordox 75 WG**

Chemical active substance(s):

Copper (I) oxide (Cu_2O), 750 g/kg

NATIONAL ASSESSMENT

Poland

(Authorization in accordance to Art. 43)

Applicant: Nordox AS

Submission date: 31/01/2022

Evaluation date: December 2022

Finalisation date: March 2023

Version history

When	What
31/01/2022	Original version from the applicant Nordox AS for Art. 43 submission. All new data and information are marked in yellow.
12/2022	Version evaluated by zRMS PL
15/02/2022	New data submitted (tomato residue trials), according the RMS comments. All new data and information are marked in green.
03/2023	Version amended by zRMS PL after comments.

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Submission and Evaluation of Copper compounds under Art.43 of 1107/2009

General observation: Deviation from standard Guidance Documents and EFSA conclusion is necessary and unavoidable for Copper.

The RMS and EFSA are held to assess plant protection products according to the existing methodology described in a series of guidance documents (GDs). Those have been developed for synthetic, organic molecules, and are in most cases not applicable to minerals and Copper. This has led to an EFSA conclusion that indicated a number of critical concerns, or assessments that could not be finalized, which do not reflect any realistic risk, but rather illustrate the inappropriateness of the current GDs for the assessment of Copper. This can easily be seen in a number of endpoints that suggest a high risk exists at concentrations below natural background of this essential micronutrient. **This has been recognized by EFSA, the RMS and several MS (see comments from DE and IT in the Peer review Report), and the EU Commission has mandated EFSA with the development with a Copper specific guidance (Mandate No. 2019-0036).**

Art.43 submissions and their evaluation by MS are unfortunately due before this GD will be available. The current EFSA conclusion and list of endpoints could at best be considered as a first tier, and applicants as well as MS are required to deviate from the standard procedures described in the GD for the following reasons:

- The current GD do not consider bio-availability; for an essential, ubiquitous micronutrient that is a metal it is indispensable to provide assessment methodologies that consider the bioavailability and the potentially toxic fraction in each real-world exposure scenario. Total concentrations do not result in any meaningful outcome.
- Data normalisation to enable comparison of toxicological lab and field data as well as data obtained with different bioavailable fractions is a pre-requisite to allow a realistic assessment of potential risk. Simplistic worst-case scenarios will always indicate a high risk already at naturally occurring concentrations.
- For a homeostatically tight controlled essential element the application of assessment factors is meaningless. The question whether an excess exposure or deficiency leads to an adverse disruption of the homeostatic control cannot be approached in this way. Further, the exceptional data richness of the Copper dossier and more than 100 years of experience with the use as fungicide make safety factors unnecessary.

These unique features of Copper are already considered in the assessment of Copper under separate legislation (REACH, BPD). While COM directed EFSA in their mandate to take advantage of those methodologies, TF members have to anticipate their use and in their proposed assessments of the critical areas of concern identified in the EFSA conclusion. This should be reviewed once the new GD is available and no use should be cancelled until then.

Submission and Evaluation of Copper compounds under Art.43 of 1107/2009

General observation: Copper compounds should not be considered as Candidate for Substitution (CfS).

The implementing Regulation (EU) 2018/1981 is renewing the approval of the active substance Copper compounds as candidate for substitution (CfS), in accordance with Regulation (EC) 1107/2009. Whereas (12) considers that Copper compounds are persistent and toxic in accordance with points 3.7.2.1 and 3.7.2.3 of Annex II to Regulation (EC) 1107/2009 (PBT assessment), and fulfil the condition set in the second indent of point 4 of Annex II to Regulation (EC) 1107/2009.

The EUCuTF disagrees with the approval as CfS. The conditions in Annex to Regulation (EC) 1107/2009 lack the exemption of inorganic compounds like Copper minerals from the PBT assessment as it has been established under other chemical legislations like REACH and BPD. As laid down in those legislations, the term persistence is meaningless for an element or mineral, due to its natural occurrence. Persistence per se is therefore not a relevant parameter and consequently a PBT assessment is not carried out for inorganic compounds under REACH and BPD. The recent mandate from COM to EFSA directs the development of a guidance towards methods and procedures available under those legislations better adapted for the assessment of inorganic compounds, where the relevant parameter is their bioavailability. This should include an exempt statement regarding the PBT assessment to harmonize the assessment of the same compounds under different legislations.

It should be noted that persistence of minerals is considered not relevant for being categorized as low-risk active substance according to Regulation (EU) 2017/1432. This is clearly not compatible with the same parameter leading to a classification as CfS under the same Regulation (EC) 1107/2009.

The EUCuTF is of the opinion that Copper compounds should not be considered CfS, and have lodged an action for annulment against Regulation (EU) 2018/1981 and renewing the approval of the active substance Copper compounds as candidate for substitution (case number T-153/19 European Union Task Force v. European Commission).

5 Analytical methods

5.1 Conclusion and summary of assessment

Sufficiently sensitive and selective analytical methods are available for the active substance and relevant impurities in the plant protection product.

Noticed data gaps are:

- None

Sufficiently sensitive and selective analytical methods are available for all analytes included in the residue definitions.

It should be noted that the applicant's dRR was not rewritten by the ZRMS and the RR resulted from the evaluation was prepared by an insertion into the dRR of the ZRMS' comments/corrections on the grey background.

After comments period the applicant amended the section with the method applied in the residue study submitted to B7 (the applicant's changes in green; the study was already evaluated).

Noticed data gaps are:

- None

Commodity/crop	Supported/ Not supported
Plant: high water, high acid, high oil, high protein/high starch content (dry) and difficult matrices	Supported
Animal: Muscle, milk, eggs, fat, liver, kidney	Supported
Soil	Supported
Water	Supported
Air	Supported
Body fluids and tissues	Supported

5.2 Methods used for the generation of pre-authorization data (KCP 5.1)

5.2.1 Analysis of the plant protection product (KCP 5.1.1)

5.2.1.1 Determination of active substance and/or variant in the plant protection product (KCP 5.1.1)

CIPAC methods are available for the determination of total Copper in formulations:

- CIPAC method 44/WP/M/ (Copper in wettable powder formulation)
- CIPAC method 44/DP/M/ (Copper in dustable powder formulation)

Each of the method noted above is based on two alternative procedures, titration or electrolytic, which are applicable to the different Copper matrices. In the first approach, free and combined Copper is converted to sulphate and determined electrolytically. The second approach is based on titration. Cupric ions are

formed by acid digestion. Addition of potassium iodide produces cuprous iodide and iodine. The iodine is titrated with sodium thiosulphate. These CIPAC methods are collaboratively tested and are, therefore, considered applicable for the determination of total Copper in WG formulations containing Copper (I) oxide. In addition, in the following, a study is summarized validating the analytical method for the determination of the copper active ingredient content.

Comments of zRMS:	Accepted. No validation is required for the CIPAC MT method anyway. Nevertheless, the method meets the Sanco/3030/99 rev. 5.
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Reference:	KCP 5.1.1/01
Title:	Nordox Agro Grade: Validation of the Analytical Method for the Determination of the Copper Active Ingredient Content
Report:	Pardo Martinez M.: 2021: CH – 0093/2021
Authority registration No:	-
Guideline(s):	Yes – SANCO/3030/99 rev. 5 dated 22/03/2019
Deviations:	No
GLP/GEP:	Yes
Acceptability:	Yes

Materials and methods

Test item

Name	Nordox Agro Grade
Active ingredient content	>97 % (nominal) as Copper (I) Oxide 87 % w/w (acc. to certificate of analysis)
Batch number	050121
Expiry date	01-2023
Storage condition	Room temperature, dark

Reference item

Name	Copper (I) Oxide
Batch number	0000109180
Purity	88.8 % as Copper
Expiry date	04-02-2022
Storage condition	-20°C in the freezer

The Copper active ingredient determination was performed according to CIPAC, Physico-chemical Methods for Technical and Formulated Pesticides 44/TC/M/3 Copper/3.2 volumetric thiosulphate method (CIPAC E, pag.44).

Specificity

A blank test was conducted with all reagents and following the titration procedure described in the report in order to check possible interferences in the colour changing at the equivalent point of the titration.

Confirmatory

The specificity of CIPAC method and the absence of interferences in the colour changing at the equivalent point of the titration is enough to guarantee this issue.

On the other hand, the confirmatory test for the active ingredient has been verified in the GLP Study No. CH-0097/2021: Spectroscopic Characterisation of Five Batch Samples by IR spectra and X-Ray diffraction analysis.

Linearity

Linear regression analysis was performed using the least squares method. The correlation coefficient was calculated using regression analysis.

To check linearity, six aliquots of Copper (I) Oxide reference material were weighed in triplicate and analyzed as described for the test item in Internal Analytical Method No. 0093/2021.

Repeatability (Precision)

Five test item solutions were analyzed following the titration procedure as described in Internal Analytical Method No. 0093/2021.

Recovery (Trueness)

The accuracy was assessed from the results of the specificity and precision tests.

Results and discussions

	Results
Specificity / Interference	The analytical method was shown to be specific for the active ingredient in test item sample.
Confirmatory	The specificity of CIPAC method and the absence of interferences in the colour changing at the equivalent point of the titration is enough to guarantee this issue.
Linearity	Five Working Standard Solutions: from 50 mg to 150 mg of Copper 1 st titration: $y = 0.1564x - 0.2986$, $R^2 = 0.99967$ 2 nd titration: $y = 0.1569x - 0.4943$, $R^2 = 0.99989$ 3 rd titration: $y = 0.1575x - 0.4480$, $R^2 = 0.99982$
Repeatability (Precision)	Five test item replicates. Copper: 86.8 ± 0.4 % w/w Copper (I) Oxide: 97.7 ± 0.4 % w/w with $RSDr > RSD\%$ and $Hr \leq 1$

Conclusion

The analytical method for determination of the Copper active ingredient content was successfully validated under SANCO/830/99 rev 5.

5.2.1.2 Description of analytical methods for the determination of relevant impurities (KCP 5.1.1)

Relevant impurities in the preparation result from impurities in the technical active substance. Methods for the determination of these impurities in the technical material are summarised in the dRAR (Annex B.5.1.2, France, 2017). Other relevant impurities will not be formed during the formulation process or during storage. In addition, in the following, two studies are summarized validating the analytical method for the determination of the sulphate and chloride significant impurities content as well as the metallic impurities content.

Comments of zRMS:	Accepted
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Reference:	KCP 5.1.1/02
Title:	Nordox Agro Grade: Validation of the Analytical Method for the Determination of the Sulphate and Chloride Significant Impurities Content
Report:	Pardo Martinez M.: 2021: CH – 0094/2021
Authority registration No:	-
Guideline(s):	Yes – SANCO/3030/99 rev. 5 dated 22/03/2019
Deviations:	No
GLP/GEP:	Yes
Acceptability:	Yes

Materials and methods

Test item

Name	Nordox Agro Grade
Active ingredient content	>97 % (nominal) as Copper (I) Oxide 87 % w/w (acc. to certificate of analysis)
Batch number	050121
Expiry date	01-2023
Storage condition	Room temperature, dark

Reference item 1

Name	Chloride
Batch number	BCCCC4654
Purity	9999 g/μL
Expiry date	01-11-2022
Storage condition	Room temperature (5-30°C)

Reference item 2

Name	Sulfate
Batch number	BCCB3694
Purity	9996 g/μL
Expiry date	01-03-2022

Storage condition Room temperature (5-30°C)

Chromatographic conditions

Column	:	IC
Thermo or equivalent	:	Ion pack® AS11-HC, 250 x 4.0 mm
Detector	:	Conductimeter
Gradient	:	5 mM for 0.5 min (curve 5) From 5 mM to 10 mM in 9.5 min (curve 7) 10 mM for 10 min (curve 5) From 10 mM to 40 mM in 10 min (curve 7) 40 mM for 5 min (curve 5) From 40 mM to 5 mM in 0.5 min (curve 7) 5 mM for 4.5 min (curve 5)
Cell Heater Temperature	:	35°C
Column Temperature	:	30°C
Suppressor type	:	ASRS_4mm
Suppressor current	:	149 mA
Flow	:	1.5 mL/min
Volume of injection	:	25 µL
R. T. Chloride	:	about 7.7 min
R. T. Sulphate	:	about 26.1 min
Total Analysis Time	:	40 min

Specificity

The specificity test was conducted injecting the samples in the adjusted chromatographic conditions, comparing the chromatograms to check possible cross contaminations.

Confirmatory

Because of incompatibility with the mass detector, it was not possible to conduct a confirmatory test.

Linearity

Linear regression analysis was performed using the least squares method. The correlation coefficient was calculated using regression analysis.

To check linearity, five working standard solutions were prepared and each solution was analyzed by IC/CD.

Repeatability (Precision)

Five test item solutions were prepared and injected as described in Internal Analytical Method No. 0094/2021.

Recovery (Trueness)

The trueness (recovery) was assessed using the data obtained with the recovery test. The test was performed by spiking test item solution with fortification solution five times at low level (0.05 % w/w) and two times at high level (0.50 % w/w).

Results and discussions

	Results																							
Specificity / Interference	The analytical method results to be specific for Chloride and Sulphate significant impurities in Nordox Agro Grade samples.																							
Confirmatory	Not possible to conduct. Just the specificity of this method and the coherence of the retention times among reference material and test item injected solutions leaves no doubt about the identity of the analytes.																							
Linearity	Five Working Standard Solutions: Nominal injected range from 1.00-75.00 µg/mL, corresponding to a nominal content in the test item of 0.005-0.375 % w/w. Impurity Chloride reference material: $y = 0.1795x - 0.0459$, $R^2 = 0.99998$ Impurity Sulphate reference material: $y = 0.1275x - 0.1430$, $R^2 = 0.99985$																							
L.O.Q.	0.05 % w/w (or 10.00 µg/mL injected)																							
L.O.D.	0.0025 % w/w (or 5 µg/mL injected)																							
Precision	Five test item replicates. Chloride: 0.13 ± 0.01 % w/w Sulphate: 0.12 ± 0.002 % w/w with Hr < 1. The precision of the analytical method was fixed at ± 10 % for each impurity higher than L.O.Q. (0.05 % w/w)																							
Recovery (Trueness)	Test item spiked 5 times at low level and 2 times at high level. For significant impurities, recovery in the correct range for Low and High fortification level. <table><tr><th>Impurity</th><th>Fortification level [% w/w]</th><th>n</th><th>Mean recovery [%]</th><th>RSD [%]</th></tr><tr><td rowspan="2">Chloride</td><td>0.05</td><td>5</td><td>98.2</td><td>1.91</td></tr><tr><td>0.50</td><td>2</td><td>98.9</td><td>0.10</td></tr><tr><td rowspan="2">Sulphate</td><td>0.05</td><td>5</td><td>86.2</td><td>5.00</td></tr><tr><td>0.50</td><td>2</td><td>99.0</td><td>0.12</td></tr></table>	Impurity	Fortification level [% w/w]	n	Mean recovery [%]	RSD [%]	Chloride	0.05	5	98.2	1.91	0.50	2	98.9	0.10	Sulphate	0.05	5	86.2	5.00	0.50	2	99.0	0.12
Impurity	Fortification level [% w/w]	n	Mean recovery [%]	RSD [%]																				
Chloride	0.05	5	98.2	1.91																				
	0.50	2	98.9	0.10																				
Sulphate	0.05	5	86.2	5.00																				
	0.50	2	99.0	0.12																				

Conclusion

The analytical method for determination of the Sulphate and Chloride significant impurities content was successfully validated under SANCO/830/99 rev 5.

Comments of zRMS:	Accepted. The ICP-MS method was used to determine the relevant impurities content. Considering the analytical technique -it is adequate to be used both in the technical material and the PPP. The method is used for analysing the relevant impurities in the technical material and the results are presented in Part C.
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Reference:	KCP 5.1.1/03
Title:	Nordox Agro Grade: Validation of the Analytical Method for the Determination of the Metallic Impurities Content (Arsenic, Cadmium, Lead, Nickel, Chromium, Cobalt, Antimony and Mercury)
Report:	Pardo Martinez M.: 2021: CH – 0095/2021
Authority registration No:	-
Guideline(s):	Yes – SANCO/3030/99 rev. 5 dated 22/03/2019
Deviations:	No
GLP/GEP:	Yes
Acceptability:	Yes

Materials and methods

Test item

Name	Nordox Agro Grade
Active ingredient content	>97 % (nominal) as Copper (I) Oxide 87 % w/w (acc. to certificate of analysis)
Batch number	050121
Expiry date	01-2023
Storage condition	Room temperature, dark

Reference item 1

Name	Multi-element analytical standard solution at about 100 µg/mL of Arsenic (As), Cadmium (Cd), Lead (Pb), Antimony (Sb), Chromium (Cr), Nickel (Ni) and Cobalt (Co)
Batch number	0006529801
Expiry date	31-05-2023
Storage condition	Room temperature (20-25°C)

Reference item 2

Name	Mercury (Hg) ICP standard
Batch number	CT-1449
Purity	1001 ± 2 µg/mL
Expiry date	30-04-2026
Storage condition	Room temperature (20-25°C)

Reference item 3

Name	Trace element fortified sample (As, Cd, Pb, Sb, Cr, Ni and Co)
Batch number	0918

Expiry date	01-07-2021
Storage condition	Refrigerated (about 4°C), dark

Instrumental conditions

Detector	:	ICP/MS
Power	:	1550 W
Carrier gas	:	0.85 L/min
Replicates	:	3 times
Sample introduction setting (peristaltic pump)		
Pump rate	:	0.1 rps

Specificity

The specificity test was conducted by injecting the samples under the described conditions and comparing the signals obtained for the different solutions.

Confirmatory

Since the analysis by ICP/MS gave quantification and identification data, the confirmatory test using another instrumental technique was not necessary.

Linearity

Linear regression analysis was performed using the least squares method. The correlation coefficient was calculated using regression analysis.

To check linearity, six working standard solutions for each metal were prepared and each solution was analyzed by ICP/MS.

L.O.Q. and L.O.D.

Data from the Linearity test were used to calculate the L.O.D. whereas data from the Recovery test were used to calculate the L.O.Q.

Repeatability (Precision)

Five test item solutions were prepared and analyzed as described in Internal Analytical Method No. 0095/2021.

Recovery (Trueness)

The test item was spiked five times at low, medium and high fortification level with the fortification reference material solution (FRMS) corresponding to the following metal additions:

Low level	:	about 1.00 mg/kg for each metal
Medium level	:	about 10.00 mg/kg for each metal
High level	:	about 100.00 mg/kg for each metal

Results and discussions

	Results
Specificity / Interference	The analytical method was shown to be specific for each of 8 metals in test item sample.

	Results																					
Linearity	<p>For each metal: six working standard solutions: from nominal 1 ng/mL to 1000 ng/mL, corresponding to metal nominal content in test item: from 0.1 mg/kg to 100 mg/kg</p> <p>Chromium reference material: $y = 0.5835x + 0.1534$, $R^2 = 0.99996$</p> <p>Nickel reference material: $y = 0.6647x - 2.0638$, $R^2 = 0.99972$</p> <p>Cobalt reference material: $y = 0.9630x - 1.1924$, $R^2 = 0.99994$</p> <p>Arsenic reference material: $y = 0.0840x - 0.0463$, $R^2 = 0.99995$</p> <p>Cadmium reference material: $y = 0.0128x - 0.0121$, $R^2 = 0.99997$</p> <p>Antimony reference material: $y = 0.0139x - 0.0365$, $R^2 = 0.99980$</p> <p>Mercury reference material: $y = 0.0177x - 0.0146$, $R^2 = 0.99994$</p> <p>Lead reference material: $y = 0.0645x + 0.0851$, $R^2 = 0.99998$</p>																					
L.O.Q.	1.00 mg/kg for As, Cd, Cr, Co, Sb, Hg (10.00 ng/mL injected) 10.00 mg/kg for Pb and Ni (100.00 ng/mL injected)																					
L.O.D.	0.5 ng/mL injected, correspondent to 0.05 mg/kg																					
Confirmatory	Since the analysis by ICP/MS gave quantification and identification data, the confirmatory test using another instrumental technique was not necessary.																					
Repeatability (Precision)	For each metal higher than L.O.Q.: five test item replicates with $RSDr > RSD\%$. For each metal lower than L.O.Q.: five test item replicates fortified at Low fortification level with $RSDr > RSD\%$.																					
Recovery (Trueness)	<p>Test item spiked 5 times at low, medium and high level.</p> <p>For each metal: recovery in the correct range for at least two fortification levels.</p> <table><tr><th>Metal</th><th>Fortification level [mg/kg]</th><th>n</th><th>Mean recovery [%]</th></tr><tr><td rowspan="3">Chromium</td><td>1.00</td><td>5</td><td>79.2</td></tr><tr><td>10.01</td><td>5</td><td>90.4</td></tr><tr><td>100.10</td><td>5</td><td>107.4</td></tr><tr><td rowspan="2">Nickel</td><td>1.00</td><td>5</td><td>(1)</td></tr><tr><td>10.01</td><td>5</td><td>84.7</td></tr></table>	Metal	Fortification level [mg/kg]	n	Mean recovery [%]	Chromium	1.00	5	79.2	10.01	5	90.4	100.10	5	107.4	Nickel	1.00	5	(1)	10.01	5	84.7
Metal	Fortification level [mg/kg]	n	Mean recovery [%]																			
Chromium	1.00	5	79.2																			
	10.01	5	90.4																			
	100.10	5	107.4																			
Nickel	1.00	5	(1)																			
	10.01	5	84.7																			

	Results			
		100.10	5	103.8
	Cobalt	1.00	5	84.0
		10.01	5	89.9
		100.10	5	110.2
	Arsenic	1.00	5	108.8
		10.01	5	103.6
		100.10	5	113.2
	Cadmium	1.00	5	91.1
		10.01	5	98.7
		100.10	5	116.3
	Antimony	1.00	5	99.2
		10.01	5	100.2
		100.10	5	117.0
	Mercury	1.00	5	90.1
		10.01	5	102.0
		100.10	5	115.7
	Lead	1.00	5	(1)
		10.01	5	86.5
		100.10	5	104.6
	(1) The mean recovery value was not in compliance with SANCO/3030/99 rev. 5 guideline`s requirement; therefore it was not considered as the L.O.Q. of the validation method.			

Conclusion

The analytical method for determination of the metallic impurities content was successfully validated under SANCO/830/99 rev 5.

5.2.1.3 Description of analytical methods for the determination of formulants (KCP 5.1.1)

Not relevant. None of the formulants are of toxicological concern within the formulation. Please refer to Part C for additional information.

5.2.1.4 Applicability of existing CIPAC methods (KCP 5.1.1)

CIPAC methods are available for the determination of total Copper in formulations (EFSA 2018;16(1):5152):

- CIPAC method 44/WP/M/ (Copper in wettable powder formulation)
- CIPAC method 44/DP/M/ (Copper in dustable powder formulation)

Each of the method noted above is based on two alternative procedures, titration or electrolytic, which are applicable to the different Copper matrices.

These CIPAC methods have been collaboratively tested and are, therefore, applicable for the determination of total Copper in relevant preparations.

These methods are suitable for total Copper determination but not specific to Copper variant. The active substance is determined as total Copper, while the identity of variant is determined by spectroscopic characterisation. The total amount of the variant is then calculated stoichiometrically.

5.2.2 Methods for the determination of residues (KCP 5.1.2)

All information and validation data were provided in the EU review of Copper compounds and were considered adequate.

An overview on the acceptable methods for analysis of residues of Copper compounds for the generation of pre-authorisation data is given in the following table. For the detailed evaluation of new studies please refer to Appendix 2.

Table 5.2-1: Validated methods for the generation of pre-authorization data

Component of residue definition: Total Copper					
Matrix type	Method type	Method LOQ		Principle of method (i.e. GC-MS or HPLC-UV)	Author(s), year / missing / EU evaluated
Grape fruit Must Wet pomace Dry pomace Stems Raisins Wine	Primary: Method MR029/RES	Grape fruit Must Wet pomace Dry pomace Stems Raisins Wine	5,0 mg/kg 1.5 mg/kg 5,0 mg/kg 25,0 mg/kg 40,0 mg/kg 13 mg/kg 0,28 mg/kg	FAAS (measurement at 324.8 nm)	Sicbaldi F. (2002a) report n°00123 EU evaluated dRAR Annex B.5.1.2, France, 2017 Refer to the post-registration method
Blanching water Tomato fruit and canned tomato Wet pomace Tomato leaves Juice	Primary: Method MR029/RES	Blanching water Tomato fruit and canned tomato Wet pomace Tomato leaves Juice	0.3mg/kg 2,0 mg/kg 6 mg/kg 10 mg/kg 12 mg/kg	FAAS (measure-ment at 324.8 nm)	Sicbaldi F. (2002b) report n°00119 EU evaluated dRAR Annex B.5.1.2, France, 2017 Refer to the post-registration method
Cucumber	Primary : acid digestion	calculated LOQ of 0.2 mg/kg		AAS (measure-ment at 324.8	Sicbaldi F., Riccelli S., (2010)

	with a 6:1:2 (v/v) mixture of 65% nitric acid, 30% hydrogen peroxide and distilled water with microwave heating		nm)	report n° RA.09.23 EU evaluated dRAR Annex B.5.1.2, France, 2017 Refer to the post-registration method
Melon	Primary : acid digestion with nitric acid	calculated LOQ of 0.2 mg/kg (Melon peel) and 0.7 mg/kg (Melon)	ICP-AES (measurement at 324.8 nm).	Hansford R.J. (2008a, b) report n° DuPont-22565 and DuPont-22564 Foster A.C. (2006) report n° DuPont-16970 EU evaluated dRAR Annex B.5.1.2, France, 2017
	Primary: acid digestion with nitric acid and heating under reflux	2 mg/kg	ICP-AES	Foster A.C. (2006a, b) report n° DuPont-14536 and DuPont-14542, revision n°1 EU evaluated dRAR Annex B.5.1.2, France, 2017
Melon Whole fruit Pulp Peel leaves	Primary: acid digestion with 65% nitric acid in the heat	calculated LOQ of 10 mg/kg in melon peel, whole fruit, pulp and leaves calculated LOQ of 3 mg/kg in melon peel and 5 mg/kg in melon whole fruit and pulp	ICP-AES (measurement at 327.8 nm)	Goebel O. (2005, 2006) report n° B-05RFLME01 and B-06RFLME01 EU evaluated dRAR Annex B.5.1.2, France, 2017
Tomato	Primary: acid digestion with 65% nitric acid and heating under reflux	0.7 mg/kg	ICP-AES	Foster A.C. (2009) report n° DuPont-22566 EU evaluated within MRL Review (Art. 12, EFSA 2018)
New residue trials				
Cherry		[m/z = 63 (quantification) and m/z = 65 (confirmation)] LOQ = 0.970 mg/kg	ICP-MS	KCA 5.1.2/02 Iffland 2020 (North 2020) S20-01045-L1

Animal products, food of animal origin	No analytical method provided				
Soil, grass and earthworm	Primary: treated with nitric acid, hydrofluoric acid and perchloric acid	40 mg/kg (grass) 20 mg/kg (soil)		ICP-OES (emission wavelengths 324.754 nm)	Klein, O. (2015) report n° 20031343/G1-NFEw EU evaluated dRAR Annex B5.1.2, France, 2017
	Primary: treated with Antifoam B Silicone Emulsion and a mixture of HCl / HNO3 (75:25 v:v) and heated under reflux or with microwave	4 mg/kg (soil) 15 mg/kg (earthworm)		ICP-OES (emission wavelengths 324.754 nm)	
Water	Primary: Total and dissolved Copper analysis	4 µg/L		ICP-MS	Blust, R. & Steven Joosen, S. (2015) report n° F-Cu 2016-2 EU evaluated dRAR Annex B5.1.2, France, 2017
	Primary: Copper ion selective electrode potentiometry	25.3 µg/L		Metrohm 692 pH/Ion Meter fitted with a Metrohm Cupric ion selective electrode and Ag/AgCl reference electrode	
	Primary:	1 µg/L Cu		AAS (GF-AAS) for low concentration levels	Schafers, C. (2000) report n° URA-001/4-50 EU evaluated dRAR Annex B5.1.2, France, 2017
Air	No analytical method provided				
Body fluids and tissues (Toxicology)	VDI, 1997	Receptor fluid	0.1 µg/L	HR-ICP mn-MS	W.J.M. Maas, C. Kunne (2015) report n° 20600/19 and A. Schouten, H.P.M. de Haan (2016) report n° V20801 EU evaluated dRAR Annex B5.1.2, France,
		Receptor/donor wash	0.25 µg/L		
		Skin membrane	0.02 µg		
		Tape strip	0.02 µg		
		Skin Wash	0.05 µg		

				2017
	Primary	5.57 µg/L (receptor fluid)	ICP-AES/ICP-MS for dose solutions, skin wash, tape strips. ICP-MS (field dilutions)/GF-AAS (concentrate) for skin membrane. GF-AAS for receptor fluid, compartment wash and donor compartment wash.	W.J.M. Maas (2012) report n° V9062/final and amendment 01 EU evaluated dRAR Annex B5.1.2, France, 2017

New studies

Data Point:	KCP 5.1.2/02 (<i>submitted under KCA 6.3.11/09</i>)
Report author:	Iffland, D.
Report year:	2020
Report title:	Determination of residues of copper after one application of copper in cherry (outdoor) at 1 sites in Southern Europe 2020
Report No:	S20-01045
Document No:	-
Guidelines followed in study:	SANCO/3029/99, rev. 4 (2000)
Deviations from current test guideline:	None
Previous evaluation:	None
GLP/Officially recognised testing facilities:	Yes, conducted under GLP/Officially recognised testing facility (Eurofins Agrosience Services Ltd)
Acceptability / Reliability:	Reliable without restriction

The study objective was to determine residue levels of copper in the raw agricultural commodity cherry. Homogenised Cherry samples were analysed after microwave digestion using ICP-MS.

I. MATERIALS AND METHODS

Matrix/medium:	Plant matrices	(cherry)
Test item:	Name:	Copper (ICP Standard)

	Supplier:	Merck
	Batch No.	HC74050314
	Expiry date:	September 2020
	Concentration/Purity:	998 mg/L Cu
Instrument and detector:	ICP-MS system Agilent 7700x and Agilent 7800	

II. RESULTS AND DISCUSSION

Characteristics of the analytical method	
Specificity	A highly specific detection system mass spectrometry (MS) was used. Copper was identified by two specific isotopes and their specific mass to charge ratio (m/z). Copper concentrations were determined using a certified reference item as an external standard and scandium as an internal standard was used for quantification. The analytical method can therefore be regarded as highly specific.
Linearity	The linearity of detector response was evaluated using solvent standard solutions. The calibration curves resulting from the injection of ten standards over the concentration range of 0.250 – 750 µg/L, corresponding to 0.042 – 125 mg/kg sample (high concentrated samples were further diluted so they could fit in the range) demonstrated linearity with coefficients of correlation (r) of 1.00.
Accuracy (Recovery) and Precision	The method was validated at the concentration levels of 0.970 and 9.93 mg/kg. At the level of 0.970 mg/kg (LOQ), the mean recovery was 99%, with relative standard deviation of 3%. At the high-level of 9.93 mg/kg, the mean recovery was 101%, with relative standard deviation of 1%. Thus, the criteria of the guideline are met. For details see the table below.
LOQ	0.970 mg/kg.
LOD	0.042 mg/kg.
Storage Stability	Not determined

Table 5.2-2 Accuracy and precision data for analytical method for copper

Matrix	Analyte	Fortification level (mg/kg)	Recovery			
			Range (%)	Mean (%)	RSD (%)	Sample size (n)
cherry	copper	0.970	96 – 103	99	3	5
		9.93	99 – 102	101	1	5
		Overall	96 – 103	-	-	10

Natural background levels in control samples exceeded 30% of LOQ. The mean copper concentration found in untreated control samples was therefore subtracted from respective recovery samples.

III. CONCLUSIONS

The analytical procedure used in the field crop residue study on cherry encompassing the Southern European region has been successfully validated for copper in terms of specificity, linearity, precision, accuracy and LOQ.

5.3 Methods for post-authorization control and monitoring purposes (KCP 5.2)

5.3.1 Analysis of the plant protection product (KCP 5.2)

Analytical methods for the determination of the active substance and relevant impurities in the plant protection product shall be submitted, unless the applicant shows that these methods already submitted in accordance with the requirements set out in point 5.2.1 can be applied.

The applicant confirms that in its expert opinion, the methods submitted under point 5.2.1 can be applied for post-authorization and monitoring and therefore additional methods under this point have not been submitted.

5.3.2 Description of analytical methods for the determination of residues of Copper (KCP 5.2)

5.3.2.1 Overview of residue definitions and levels for which compliance is required

Table 5.3-1: Relevant residue definitions for monitoring/enforcement and levels for which compliance is required

Matrix	Residue definition	MRL / limit	Reference for MRL/level Remarks
Plant, high water content	Total Copper	from 5 mg/kg to 100 mg/kg	Reg. (EC) No. 149/2008
Plant, high acid content		from 5 mg/kg to 50 mg/kg	
Plant, high protein/high starch content (dry commodities)		10 mg/kg	
Plant, high oil content		from 20 mg/kg to 30 mg/kg	
Plant, difficult matrices (hops, spices, tea)		from 40 mg/kg to 100 mg/kg	
Muscle, milk, eggs, fat, liver and kidney	Total Copper	from 2 mg/kg to 30 mg/kg	Reg. (EC) No. 149/2008
Soil (Ecotoxicology)	Total Copper	5 mg/kg	Common limit
Drinking water (Human toxicology)	Dissolved Copper	2 mg/L	Council directive 98/83/EC; general limit for drinking water
Surface water (Ecotoxicology)		0.00112 mg DISSOLVED Cu/L	According to lowest NOEC from aquatic toxicity study
Air	Total Copper	22.5 µg/m ³	AOEL sys: 0.075 mg/kg bw/day
Tissue (meat or liver)	Total Copper	0.1 mg/kg	SANCO/825/00 rev 8.1; general limit Not classified as T / T+
Body fluids		0.05 mg/L	

5.3.2.2 Description of analytical methods for the determination of residues in plant matrices (KCP 5.2)

An overview on the acceptable methods for analysis of Copper compounds in plant matrices is given in the following tables. For the detailed evaluation of additional studies, it is referred to Appendix 2.

Table 5.3-2: Validated methods for food and feed of plant origin (required for all matrix types, “difficult” matrix only when indicated by intended GAP)

Component of residue definition: Total Copper				
Matrix type	Method type	Method LOQ	Principle of method (i.e. GC-MS or HPLC-UV)	Author(s), year / missing / EU evaluated
High water content and High acid content	Primary	5.0 mg/kg in grape	AAS (measurement at 324.8 nm)	Sicbaldi F. (2002a) report n°00123 EU evaluated dRAR Annex B.5.2.1, France, 2017
	Primary	2.0 mg/kg in tomato	AAS (measurement at 324.8 nm)	Sicbaldi F. (2002b) report n°00119 EU evaluated dRAR Annex B5.1.2, France, 2017
	Primary	0.2 mg/kg in cauliflower, strawberry, sugarbeet (roots), canned peaches and beer 0.4 mg/kg in strawberry jam 0.8 mg/kg in cooked peas 2.0 mg/kg in apple dry pomace	AAS (measurement at 324.8 nm)	Sicbaldi F., Riccelli S., (2010) report n° RA.09.23 EU evaluated dRAR Annex B.5.1.2, France, 2017
	Primary	3.0 mg/kg in tomato juice 1.1 mg/kg in melon pulp 0.8 mg/kg melon peel	AAS (measurement at 324.8 nm)	Riccelli S., (2016) report n° RA.16.08 EU evaluated dRAR Annex B.5.1.2, France, 2017
	Primary (matrix effect evaluation)	Not applicable	FAAS (measurement at 324.8 nm)	Riccelli S., (2017) report n° RA.17.02 Appendix 2
	Primary EN 13805	Not applicable	Atomic absorption (flame, electrothermal (ET), hydride, cold-vapour) techniques and ICP-MS	Anon., (2014) report n° EN 13805 EU evaluated dRAR Annex B5.2.1, France, 2017

Component of residue definition: Total Copper				
Matrix type	Method type	Method LOQ	Principle of method (i.e. GC-MS or HPLC-UV)	Author(s), year / missing / EU evaluated
	ILV	Not required		EU evaluated dRAR Annex B5.2.1, France, 2017
	Confirmatory (if required)	Not required		
High oil content	Primary	1.0 mg/kg in oilseed rape	AAS (measurement at 324.8 nm)	Sicbaldi F., Riccelli S., (2010) report n° RA.09.23 EU evaluated dRAR Annex B5.1.2, France, 2017
	Primary	5.5 mg/kg	FAAS (measurement at 324.8 nm)	Riccelli S., (2017) report n° RA.17.02 Appendix 2
	Primary EN 13805	Not applicable	Atomic absorption (flame, electrothermal (ET), hydride, cold- vapour) techniques and ICP-MS	Anon., (2014) report n° EN 13805 EU evaluated dRAR Annex B5.2.1, France, 2017
	ILV	Not required		EU evaluated dRAR Annex B5.2.1, France, 2017
	Confirmatory (if required)	Not required		
High protein/high starch content (dry)	Primary	7.5 mg/kg	FAAS (measurement at 324.8 nm)	Riccelli S., (2017) report n° RA.17.02 KCP 5.2/01 Appendix 2
	Primary EN 13805	Not applicable	Atomic absorption (flame, electrothermal (ET), hydride, cold- vapour) techniques and ICP-MS	Anon., (2014) report n° EN 13805 EU evaluated dRAR Annex B5.2.1, France, 2017
	ILV	Not required		EU evaluated dRAR Annex B5.2.1, France, 2017
	Confirmatory (if required)	Not required		
Difficult (if required, depends on intended use)	Primary	0.5 mg/kg in hops	AAS (measurement at 324.8 nm)	Sicbaldi F., Riccelli S., (2010) report n° RA.09.23 EU evaluated dRAR Annex B5.1.2, France, 2017
	ILV	Not required		EU evaluated dRAR Annex B5.2.1, France,

Component of residue definition: Total Copper				
Matrix type	Method type	Method LOQ	Principle of method (i.e. GC-MS or HPLC-UV)	Author(s), year / missing / EU evaluated
				2017
	Confirmatory (if required)	Not required		

For any special comments or remarkable points concerning the analytical methods for the determination of residues in plant matrices, please refer to Appendix 2.

Table 5.3-3: Statement on extraction efficiency

	Method for products of plant origin
Not required for Copper	The methods of analysis do not rely on an extraction process but utilises acid digestion with AAS or ICP-MS detection.

5.3.2.3 Description of analytical methods for the determination of residues in animal matrices (KCP 5.2)

An overview on the acceptable method for analysis of total Copper in animal matrices is given in the following table.

Table 5.3-4: Validated methods for food and feed of animal origin (if appropriate)

Component of residue definition: Total Copper				
Matrix type	Method type	Method LOQ	Principle of method (i.e. GC-MS or HPLC-UV)	Author(s), year / missing
Milk, eggs, muscle, fat, kidney and liver	Primary EN14082:2003	Not applicable	Atomic absorption spectrometry (AAS)	Anon., (2003) report n° EN14082:2003 EU evaluated dRAR Annex B5.2.2, France, 2017
	ILV	Not required		EU evaluated dRAR Annex B5.2.1, France, 2017
	Confirmatory (if required)	Not required		

Table 5.3-5: Statement on extraction efficiency

	Method for products of animal origin
Not required for Copper	The methods of analysis do not rely on an extraction process but utilises acid digestion with AAS or ICP-MS detection.

5.3.2.4 Description of methods for the analysis of soil (KCP 5.2)

An overview on the acceptable methods for analysis of total Copper in soil is given in the following table.

Table 5.3-6: Validated methods for soil

Component of residue definition: Total Copper			
Method type	Method LOQ	Principle of method (i.e. GC-MS or HPLC-UV)	Author(s), year / missing
Primary	5.0 mg/kg	ICP-AES (bio-available Copper)	Kiefer, (2003) report n°20031084/02-UVX EU evaluated dRAR Annex B5.2.3, France, 2017
	20 mg/kg	AAS	Carey, (1989) report n° 88-003 EU evaluated dRAR Annex B5.2.3, France, 2017
	Soil: 15 mg/kg (following SANCO/825/00) 0.52 mg/kg (following DIN 32645)	ICP-AES	Kiefer, (2004) report n° 20031084/01-UVX EU evaluated dRAR Annex B5.2.3, France, 2017
Confirmatory	Not required		

5.3.2.5 Description of methods for the analysis of water (KCP 5.2)

An overview on the acceptable methods for analysis of dissolved Copper in surface and drinking water is given in the following table.

Table 5.3-7: Validated methods for water

Component of residue definition: Dissolved Copper				
Matrix type	Method type	Method LOQ	Principle of method (i.e. GC-MS or HPLC-UV)	Author(s), year / missing
Drinking water and Surface water	Primary	100 µg/L	ICP-AES	Heintze A., (2000 and 2001) EU evaluated dRAR Annex B.5.2.4, France, 2017
	Primary	30 µg/L	ICP-AES	Carey D.O., (1989) EU evaluated dRAR Annex B.5.2.4, France, 2017
	Primary	0.3 µg/L	ICP-MS	Pardo Martinez, M. (2016) EU evaluated dRAR Annex B.5.2.4, France, 2017
	Primary DIN 38406 Part 7	2 µg/L	AAS	Anon., (1991) EU evaluated dRAR Annex B.5.2.4, France, 2017
	Primary ISO 15586:2003	Not applicable	AAS	Anon., (2004) EU evaluated dRAR Annex B.5.2.4, France, 2017
	ILV	Not required		EU evaluated dRAR Annex B.5.2.4, France, 2017
	Confirmatory	Not required		

5.3.2.6 Description of methods for the analysis of air (KCP 5.2)

An overview on the acceptable methods and possible data gaps for analysis of total Copper in air is given in the following tables. For the detailed evaluation of new studies please refer to Appendix 2.

Table 5.3-8: Validated methods for air

Component of residue definition: Total Copper			
Method type	Method LOQ	Principle of method (i.e. GC-MS or HPLC-UV)	Author(s), year / missing
Primary	Not applicable	AAS (wavelength 324.8 nm)	Anon., (1999) EU evaluated dRAR Annex B.5.2.5, France, 2017
Primary	Not applicable	ICP-OES (wavelength 324.8 nm and 327.4 nm)	Anon., (1997) EU evaluated dRAR Annex B.5.2.5, France, 2017
Primary	0.3 µg/m ³	ICP-MS	Pardo Martinez M., (2018)

Component of residue definition: Total Copper			
Method type	Method LOQ	Principle of method (i.e. GC-MS or HPLC-UV)	Author(s), year / missing
			report n° CH-657/2017 KCP 5.2/02 Appendix 2
Confirmatory	Not required		

For any special comments or remarkable points concerning the analytical methods for air please refer to Appendix 2.

5.3.2.7 Description of methods for the analysis of body fluids and tissues (KCP 5.2)

An overview on the acceptable methods and possible data gaps for analysis of total Copper in body fluids and tissues is given in the following table.

Table 5.3-9: Methods for body fluids and tissues

Component of residue definition: Total Copper			
Method type	Method LOQ	Principle of method (i.e. GC-MS or HPLC-UV)	Author(s), year / missing
Primary	3.0 mg/kg for plasma, 13.9 mg/kg for bile, 359 mg/kg for liver, 841 mg/kg for faeces and 46 mg/kg for carcass	ICP-AES (emission wave-length 324.752 nm)	Himmelstein M.W., (2004) EU evaluated dRAR Annex B.5.2.6, France, 2017
Confirmatory	Not required		

5.3.2.8 Other studies/ information

No other studies available

Appendix 1 Lists of data considered in support of the evaluation

List of data submitted by the applicant and relied on

Data point	Author(s)	Year	Title Company Report No. Source (where different from company) GLP or GEP status Published or not	Vertebrate study Y/N	Owner
KCP 5.1.1/01	Pardo Martinez, M.	2021	Nordox Agro Grade: Validation of the Analytical Method for the Determination of the Copper Active Ingredient Content Company Report No.: 0093/2021 ChemService S.r.l. Controlli e Ricerche, Italy GLP Unpublished	N	Nordox
KCP 5.1.1/02	Pardo Martinez, M.	2021	Nordox Agro Grade: Validation of the Analytical Method for the Determination of the Sulphate and Chloride Significant Impurities Content Company Report No.: 0094/2021 ChemService S.r.l. Controlli e Ricerche, Italy GLP Unpublished	N	Nordox
KCP 5.1.1/03	Pardo Martinez, M.	2021	Nordox Agro Grade: Validation of the Analytical Method for the Determination of the Metallic Impurities Content (Arsenic, Cadmium, Lead, Nickel, Chromium, Cobalt, Antimony and Mercury) Company Report No.: 0095/2021 ChemService S.r.l. Controlli e Ricerche, Italy GLP Unpublished	N	Nordox

Data point	Author(s)	Year	Title Company Report No. Source (where different from company) GLP or GEP status Published or not	Vertebrate study Y/N	Owner
KCP 5.1.2/02 Submitted under KCA 6.3.11/09	North, L.	2021	Determination of residues of copper after one application of copper in cherry (outdoor) at 1 site in Southern Europe 2020 Company Report No: S20-01045 Eurofins AgroScience, Germany GLP Unpublished	N	Nordox AS
KCP 5.1.2/03 Submitted under KCA 6.3.2/07	Foster A.C.	2009	Magnitude of residues of copper in tomatoes (<i>solanaceae</i> -fruiting vegetables) following application of metallic copper (as copper hydroxyde) (DPC-GFJ52) 35WG – Northern Europe, Season 2007-2008 Company Report No: DuPont 22566 Charles River Laboratories (UK) GLP Unpublished	N	European Union Copper Task Force (LoA available)
KCP 5.2/01	Riccelli S.	2017	Method Validation for the determination of Copper in/on dry and oily matrices and Matrix Effect evaluation on dry, oily, high water and acid matrices Company Report No RA.17.02 Isagro – Centro di Saggio BPL GLP Published	N	European Union Copper Task Force
KCP 5.2/02	Pardo Martinez M.	2018	Validation of the Analytical Method for the determination of Copper residues in Air Company Report No: CH-657/2017 ChemService GLP Unpublished	N	European Union Copper Task Force

List of data submitted or referred to by the applicant and relied on, but already evaluated at EU peer review

Please note that all data mentioned as part of DAR, RAR, or EFSA journals are considered as relied on.

Data point	Author(s)	Year	Title Company Report No. Source (where different from company) GLP or GEP status Published or not	Vertebrate study Y/N	Owner
KCP 5.1.2 and KCP 5.2	Sicbaldi, F.	2002a	Analytical method validation for the determination of Copper in/on grapes and their processed fractions Company Report No: 00123 Isagro Ricerca S.r.l GLP Unpublished	N	European Union Copper Task Force
KCP 5.1.2 and KCP 5.2	Sicbaldi, F.	2002b	Analytical method validation for the determination of Copper in/on tomatoes, their processed fractions and leaves. Company Report No: 00119 Isagro Ricerca S.r.l GLP Unpublished	N	European Union Copper Task Force
KCP 5.1.2 and KCP 5.2	Sicbaldi, F., Riccelli S.	2010	Method validation for the reduction of the Limit of Quantification for Copper in representative matrices of plant origin. Company Report No: RA.09.23 Isagro Ricerca S.r.l GLP Unpublished	N	European Union Copper Task Force
KCP 5.1.2	Hansford, R.J.	2008a	Magnitude of residues of Copper in field melons (cucurbits-inedible peel) following applications of metallic Copper (as Copper oxychloride)/Cymoxanil (DPX-KK807) 44WP (9.5:1)-southern Europe, season 2007 Company Report No: DuPont-22565 Charles River Laboratories (UK) GLP Unpublished	N	European Union Copper Task Force

Data point	Author(s)	Year	Title Company Report No. Source (where different from company) GLP or GEP status Published or not	Vertebrate study Y/N	Owner
KCP 5.1.2	Hansford, R.J.	2008b	Magnitude of residues of Copper in protected melons (curcurbits – inedible peel) following applications of metallic Copper (as Copper oxychloride) / cymoxanil (DPX-KK807) 44WP (9.5:1) – Southern Europe, season 2007 Company Report No: DuPont 22564 DuPont GLP Unpublished	N	European Union Copper Task Force
KCP 5.1.2	Foster, A.C.	2006b	Magnitude of residues of Copper and cymoxanil in field melons (fruiting vegetables) following applications of metallic Copper (as Copper oxychloride)/cymoxanil (DPX-KK807) 44WG (9.5:1) under maximum label rates - southern Europe, season 2005 Company Report No: DuPont-16970 Charles River Laboratories (UK) GLP Unpublished	N	European Union Copper Task Force
KCP 5.1.2	Foster, AC	2006c	Magnitude of residues of Copper and cymoxanil in protected melons (fruiting vegetables) following applications of metallic Copper (as Copper oxychloride)/cymoxanil (DPX-KK807) 44WG (9.5:1) under maximum label rates – southern Europe, 2004 Company Report No: DuPont 14536 DuPont GLP Unpublished	N	European Union Copper Task Force
KCP 5.1.2	Foster, A.C.	2006a	Magnitude of residues of Copper and cymoxanil in field melons (fruiting vegetables) following applications of metallic Copper (as Copper oxychloride)/cymoxanil (DPX-KK807) 44WG (9.5:1) under maximum label rates - southern Europe, 2004 Company Report No: DuPont-14542, Revision No. 1 Charles River Laboratories (UK) GLP Unpublished	N	European Union Copper Task Force

Data point	Author(s)	Year	Title Company Report No. Source (where different from company) GLP or GEP status Published or not	Vertebrate study Y/N	Owner
KCP 5.1.2	Goebel, O	2005	Residue determination of Copper in melon after 6 applications of ATOFAP02 (WG 20%) or ATOFAP17NC (WG 40%) Company Report No: B_05RFLME01 Staphyt GLP Unpublished	N	European Union Copper Task Force
KCP 5.1.2	Goebel, O	2006	Residue determination of Copper in melon after 6 applications of ATOFAP02 (Copper - 20% WG) or ATOFAP17NC (Copper - 40% WG) Company Report No: B_06RFLME01 GLP Unpublished	N	European Union Copper Task Force
KCP 5.1.2	Klein, O.	2015	A Field Study to Evaluate the Effects of Copper on the Earthworm Fauna in Central Europe Company Report No: 20031343/G1-NFEw Eurofins GLP Unpublished	N	European Union Copper Task Force
KCP 5.1.2	Blust R., Steven Joosen S	2015	Kinetics and speciation of Copper in Copper based fungicide formulations used in crop protection (Update February 2016) F-Cu 2016-2 Department of Biology, University of Antwerp, Belgium Non-GLP Unpublished	N	European Union Copper Task Force
KCP 5.1.2	Schafers, C.	2000	Community level study with Copper hydroxide 50% WP in aquatic microcosms Company Report No: URA-001/4-50 Fraunhofer - Institut für Umweltchemie und Okotoxikologie - IUCT GLP: Yes Published: No	N	European Union Copper Task Force

Data point	Author(s)	Year	Title Company Report No. Source (where different from company) GLP or GEP status Published or not	Vertebrate study Y/N	Owner
KCP 5.1.2	Maas W.J.M., Kunne C.	2015	In vitro percutaneous absorption of Copper, formulated as Copper hydroxide 50 WP or Copper oxychloride SC, through human and rat skin Company Report No: V20600/19 + Amendment 01 TNO, Zeist, the Netherlands GLP Unpublished	N	European Union Copper Task Force
KCP 5.1.2	Shouten, A., de Haan H.P.M.	2016	Validation of the determination of 65Cu in receptor fluid, stripped skin, tape strips, receptor/donor wash solution and skin wash used in the 'In vitro percutaneous absorption test of Copper through human and rat skin', using a double-focusing high resolution inductively coupled plasma mass spectrometer (HR-ICP-MS) Company Report No: V20801 Triskelion BV GLP Unpublished	N	European Union Copper Task Force
KCP 5.1.2	Maas WJM	2016	In vitro dermal absorption of Copper (Cu) from 8 formulations through human skin Company Report No: V9062 + Amendment 01 TNO, Zeist, the Netherlands GLP Unpublished	N	European Union Copper Task Force
KCP 5.2	Riccelli S.	2016	Method Validation for the determination of Copper in/on tomato juice and melon (pulp and peel) Company Report No: RA.16.08 Isagro GLP Unpublished	N	European Union Copper Task Force

Data point	Author(s)	Year	Title Company Report No. Source (where different from company) GLP or GEP status Published or not	Vertebrate study Y/N	Owner
KCP 5.2	Anon.	2014	Foodstuffs – Determination of trace elements – Pressure digestion Report No.: DIN EN 13805 Deutsches Institut für Normung Non-GLP Published:	N	Public
KCP 5.2	Anon.	2003	Foodstuffs – Determination of trace elements – Determination of lead, cadmium, zinc, Copper, iron and chromium by atomic absorption spectrometry (AAS) after dry ashing Report No.: EN 14082 European committee for standardization Non-GLP Published	N	Public
KCP 5.2	Kiefer, R.	2003	Validation of an analytical method for the determination of bioavailable Copper in soil samples Company Report No: 20031084/02-UVX GAB Biotechnologie GmbH & IFU Umweltanalytik GmbH GLP Unpublished	N	European Union Copper Task Force
KCP 5.2	Carey, D. O.	1989	Method validation report for terrestrial outdoor field dissipation study with Copper-containing pesticides Company Report No: 88-003 Biospherics Inc. GLP Unpublished	N	European Union Copper Task Force
KCP 5.2	Kiefer, R.	2004	Validation of an analytical method for the determination of total Copper in soil samples Company Report No: 20031084/01-UVX GAB Biotechnologie GmbH & IFU Umweltanalytik GmbH GLP Unpublished	N	European Union Copper Task Force

Data point	Author(s)	Year	Title Company Report No. Source (where different from company) GLP or GEP status Published or not	Vertebrate study Y/N	Owner
KCP 5.2	Heintze, A.	2001	Assessment of side effects of URA-13900-F-0-WP on the larvae of the midge, <i>Chironomous riparius</i> with the laboratory test method. Company Report No: 99520/01-ASCr GAB Biotechnologie GmbH & IFU Umweltanalytik GmbH GLP Unpublished	N	European Union Copper Task Force
KCP 5.2	Heintze, A.	2000	Assessment of side effects of URA-08740-F-0-WP on the larvae of the midge, <i>Chironomous riparius</i> with the laboratory test method Company Report No: 99507/01-ASCr GAB Biotechnologie GmbH & IFU Umweltanalytik GmbH GLP Unpublished	N	European Union Copper Task Force
KCP 5.2	Martinez M. P.	2016	Validation of the analytical method for the determination of Copper in surface water Company Report No.: CH-157/2016 ChemService S.r.l. Controlli e Ricerche GLP Unpublished	N	European Union Copper Task Force
KCP 5.2	Anon.	1991	German standard methods for the examination of water, waste water and sludge; Cations (group E); Determination of Copper by atomic absorption spectrometry (AAS) (E 7) Report No.: DIN 38406 Part 7, September 1991 Deutsches Institut für Normung Non-GLP Published	N	Public

Data point	Author(s)	Year	Title Company Report No. Source (where different from company) GLP or GEP status Published or not	Vertebrate study Y/N	Owner
KCP 5.2	Anon.	2004	Water quality. Determination of trace elements using atomic absorption spectrometry with graphite furnace Report No.: DIN EN ISO 15586 Deutsches Institut für Normung Non-GLP Published	N	Public
KCP 5.2	Anon.	1999	Determination of suspended matter in ambient air. Measurement of the concentration by mass of As, Be, Cd, Co, Cr, Cu, Mn, Ni, Pb, Sb, Tl, Zn by atomic absorption spectrometry (AAS) after sampling on filters and digestion in an oxidising acid mixture. Report No.: VDI 2267, Part 1 Verein Deutscher Ingenieure Non-GLP Published	N	Public
KCP 5.2	Anon.	1997	Determination of suspended matter in ambient air. Determination of the mass concentration of Be, Cd, Co, Cr, Cu, Fe, Mn, Ni, Pb, Sb, V, Zn by optical emission spectrometry (ICPOES) after sampling on filters and digestion in an oxidising agent. Report No.: VDI 2267, Part 5 Verein Deutscher Ingenieure Non-GLP Published	N	Public
KCP 5.2	Himmelstein, M. W.	2004	Five Copper substances: Absorption, distribution, and excretion in male rats. Company Report No: DuPont-11784 E.I. du Pont de Nemours and Company GLP Unpublished	N	European Union Copper Task Force

Appendix 2 Detailed evaluation of submitted analytical methods

A 2.1 Analytical methods for Copper compounds

A 2.1.1 Methods used for the generation of pre-authorization data (KCP 5.1)

No new studies have been submitted.

A 2.1.1.1 Analytical method 1

A 2.1.1.1.1 Method validation

Comments of zRMS:	
Reference:	KCP 5.1.2/03, submitted under KCA 6.3.2/07
Title:	Magnitude of residues of copper in tomatoes (<i>solanaceae</i> -fruiting vegetables) following application of metallic copper (as copper hydroxyde) (DPC-GFJ52) 35WG – Northern Europe, Season 2007-2008
Report:	DuPont 22566
Authority registration No:	-
Guideline(s):	European Communities 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 EC Commission Directive 91/414/EEC.
Deviations:	No
GLP/GEP:	Yes
Acceptability:	Yes

Materials and methods

The objective of this study was to determine residues of copper in tomatoes from field samples.

Levels of Copper were determined from dried samples by acidic digestion and reflux heating. After dilution with demineralized water, the mineralized sample was analyzed by ICP-AES (324.8 nm).

The underlying analytical method has been fully validated in study DuPont-16969. Recovery determination has been performed at two fortification levels of 0.7 mg/kg and 7 mg/kg.

Stock, standard solutions and fortification solutions were prepared from an independent source of stock standard.

Results and discussions

The validation of this method by procedural recovery determination was successful and has met the criteria reported in the Guideline Requirements. Method validation data are summarised in Table A 1 and Table A 2.

All mean recoveries were within the required 70-110%. The repeatability found has acceptable %RSD as all values were below the required 20%.

Table A 1: Recovery results from method validation of total Copper using this analytical method

Crop group	Matrix	Fortification level [mg/kg]	n	Mean recovery [%]	RSD [%]	Comments
Tomato	Tomato	0.7	4	93.8	4.2	
		7.0	4	96.5	0.6	
		Overall	8	95.2	3.3	

Table A 2: Characteristics for the analytical method used for validation of total Copper residues in tomatoes

	Total Copper
Specificity	The specificity of the method is given by using ICP-AES analytical technique at the Copper specific wavelength of 324.8 nm. No additional confirmatory method is required. Blank values in control and reagent blank samples, used for method validation, were below 30% of the LOQ.
Calibration (type, number of data points)	The regression equation was generated by the calibration curve in solvent (type 1/x) using the absorbance responses versus the respective concentrations of the calibration standards. Number of data points = 5
Calibration range	Accepted calibration range: Standards prepared in solvent. 0.01 – 10.0 mg/L corresponding to 0.05 - 50 mg/kg. $y = 4.6991E-6 x - 0.007919$, $R^2 = 1.0000$
Assessment of matrix effects is presented	No.
Limit of determination/quantification	LOQ: 0.70 mg/kg LOD: 0.05 mg/kg

Conclusion

The analytical method was successfully validated and meets all guideline criteria to determine residues of total Copper in tomato matrix.

A 2.1.2 Methods for post-authorization control and monitoring purposes (KCP 5.2)

A 2.1.2.1 Description of analytical methods for the determination of residues in plant matrices (KCP 5.2)

A 2.1.2.1.1 Analytical method 1

A 2.1.2.1.1.1 Method validation

Comments of zRMS:	The method has been accepted.
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Reference:	KCP 5.2/01
Title:	Method Validation for the determination of Copper in/on dry and oily matrices and Matrix Effect evaluation on dry, oily, high water and acid matrices
Report:	dart://dart/edition?ed_no=M-201728-01-1Riccelli S.: 2017: RA.17.02
Authority registration No:	-
Guideline(s):	REGULATION (EC) No 1107/2009 OF THE EUROPEAN PARLIAMENT AND OF THE COUNCIL of 21 October 2009 concerning the placing of plant protection products on the market and repealing Council Directives 79/117/EEC and 91/414/EEC. European Commission Guidance Document for Generating and Reporting Methods of Analysis in Support of Pre-Registration data Requirements for Annex II (part A, Section 4) and Annex III (part A, section 5) of directive 91/414, SANCO/3029/99. Guidance document on residue analytical methods; SANCO/825/00 rev. 8.1, European Commission, Directorate General Health and Consumer Protection; 2010-11-16. OECD Guidance Document on Pesticide Residue analytical Methods; ENV/JM/Mono (2007); 2007-08-13
Deviations:	No
GLP/GEP:	Yes
Acceptability:	Yes

Materials and methods

The objective of this study is to determine the matrix effect in dry, oily, high water and acid matrices and to validate the method in dry and oily matrices at limits of quantification (LOQ) established.

Level of Copper are determined from homogenized samples by acidic digestion and microwave heating. The solution containing the mineralized sample was analysed by reading of its absorbance at 324.8 nm, after calibration of the Flame Atomic Absorption Spectrometer (FAAS) with standard solutions in solvent.

The matrices reported in the following table cover the categories object of the study:

Commodity categories	Representative RAC commodities
High water content	Lettuce
High acid content	Grape

Commodity categories	Representative RAC commodities
High oil content	Oilseed rape seed
Dry matrix	Wheat grain

A full validation method was carried out only on oily matrix and dry matrix, while the matrix effect was evaluated for all matrices tested.

Effects of matrix constituents present in the final mineralized sample were assessed by comparing the concentration (mg/L) of Copper obtained from standard solutions in neat solvent with those observed for standards added in mineralized sample of untreated matrices.

Stock, standard solutions and fortification solutions were prepared in water solution of nitric acid 1%.

Results and discussions

- Determine the matrix effect in dry matrix (wheat grain), oily matrix (oilseed rape seed), matrix with high water content (lettuce) and matrix with high acid content (grape):

Matrix Effect was calculated for two levels of the calibration curve: one at a level near to the LOQ level and another at a higher level. The mean matrix effect for each matrix tested is reported below.

Matrix	Mean Matrix Effect (%)
Lettuce	-19.5
Grape	-11.5
Oilseed rape seed	-0.8
Wheat grain	-2.4

The mean effects of matrix on response were not significant for all matrices (<20%).

- Validation of a Copper method in dry matrix (wheat grain) and oily matrix (oilseed rape seed) by FAAS:

The validation of this method was successful and has met the criteria reported in the Guideline Requirements. Method validation data are summarised in Table A 1 and Table A 2.

All mean recoveries were within the required 70-110 % range for both matrices. The repeatability found has acceptable %RSD as all values were below the required 20 %.

Table A 3: Recovery results from method validation of total Copper using this analytical method

Crop group	Matrix	Fortification level [mg/kg]	n	Mean recovery [%]	RSD [%]	Comments
dry matrices	Wheat grain	7.5	7	88.6	4.6	-
		75	5	104.3	2.6	-
oily	Oilseed rape	5.5	7	87.4	10.7	-

Crop group	Matrix	Fortification level [mg/kg]	n	Mean recovery [%]	RSD [%]	Comments
matrices	seed	55	5		3.4	-

Table A 4: Characteristics for the analytical method used for validation of total Copper residues in wheat grain (dry matrices) and oilseed rape seed (oily matrices)

	Total Copper
Specificity	The specificity of the method is given by using the F AAS analytical technique at the Copper specific wavelength of 324.8 nm. No additional confirmatory method is required. Blank values in control samples of the oilseed rape seed, used for method validation, were below 30% of the LOQ. Blank values in control samples of the wheat grain, used for method validation, were 37.8% of the set LOQ.
Calibration (type, number of data points)	The regression equation was generated by the calibration curve in solvent (type 1/x) using the absorbance responses versus the respective concentrations of the calibration standards. Number of data points = 6
Calibration range	Accepted calibration range: Standards prepared in solvent. For grain: 0.01 – 10.0 mg/L corresponding to 0.2 - 200 mg/kg. $y = 0.000509 + 0.110782 x$, $R^2 = 0.9999$ For grain: 0.0125 – 10.0 mg/L corresponding to 0.4 - 400 mg/kg. $y = -0.000391 + 0.101070 x$, $R^2 = 1.0000$
Assessment of matrix effects is presented	Yes. Matrix Effect was calculated for two levels of the calibration curve: one at a level near to the LOQ level and another at a higher level. The mean effects of matrix on response were not significant for all matrices (<20%).
Limit of determination/quantification	For oilseed rape seed (<i>oily matrix</i>) the LOQ tested have been first estimated on the basis of the natural background Copper level in the untreated samples to establish the lowest possible LOQ, based on the background not exceeding 30% of the LOQ. For wheat grain (<i>dry matrix</i>) the tested LOQ was chosen to support a suggested MRL for the cereals at 15 mg/kg. To support an MRL the LOQ should not exceed ~ of the proposed MRL. <u>Tested LOQs:</u> Oilseed rape seed: 5.5 mg/kg Wheat grain: 7.5 mg/kg

Conclusion

The analytical method was successfully validated and meets all guideline criteria to determine residues of total Copper in dry and oily matrices. Also, the matrix effect in dry, oily, high water and acid matrices was assessed as not significant for all matrices (< 20 %).

A 2.1.2.2 Description of analytical methods for the determination of residues in animal matrices (KCP 5.2)

No additional studies have been submitted, please refer to the dRAR for information.

A 2.1.2.3 Description of Methods for the Analysis of Soil (KCP 5.2)

No additional studies have been submitted, please refer to the dRAR for information.

A 2.1.2.4 Description of Methods for the Analysis of Water (KCP 5.2)

No additional studies have been submitted, please refer to the dRAR for information.

A 2.1.2.5 Description of Methods for the Analysis of Air (KCP 5.2)

A 2.1.2.5.1 Analytical method 1

A 2.1.2.5.1.1 Method validation

Comments of zRMS:	The method has been accepted.
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Reference:	KCP 5.2/02
Title:	Validation of the Analytical Method for the determination of Copper residues in Air
Report:	dart://dart/edition?ed_no=M-201728-01-1 Pardo Martinez M.: 2018: CH-657/2017
Authority registration No:	-
Guideline(s):	Yes – SANCO/3029/99 rev 4 and SANCO/825/00 rev 8.1
Deviations:	Yes; 1. Although the matrix effect test was not required in the Study Plan and in the SANCO/825/00 rev. 8.1, it was conducted. 2. The retention capacity of the sampling system (membrane filter) test was required in the Study Plan, it was conducted in this study to be in compliance with EEC guideline SANCO/825/00 rev. 8.1 dated 16/11
GLP/GEP:	Yes
Acceptability:	Yes

Materials and methods

Residues of Copper in air are adsorbed with a membrane filter. The filter holder, 37 mm two-piece cassette, was assembled, preloaded with the GN-4 0.8 um Metrice® membrane filter and the support pad in MCE (mixed cellulose ester). A measured volume of air is drawn through the membrane filter. The membrane filter is dissolved with nitric acid (65-71% % v/v for ultratrace metal analysis) and the determination of Copper residues is performed by Inductively Coupled Plasma-Mass Spectrometry using an external standard.

Its quantification is achieved by comparing the Copper analytical standard intensity signal versus the intensity signal in air samples.

Instrumental conditions

Detector	:	ICP/MS
Power	:	1550 W
Carrier gas	:	0.85 L/min
Replicates	:	3 times
Sample introduction setting (peristaltic pump)		
Pump rate	:	0.1 rps
Copper (Cu)	:	m/z 63 (quantitative)
		m/z 65 (qualitative)
Germanium (Ge)	:	m/z 72 (used as internal standard correction) ⁽¹⁾

⁽¹⁾ Internal standard for Copper quantification.

Experiment:

- 1) For accuracy and precision: The air sampling was performed for 30 minutes at a 10 L/min air flow rate in order to sample a total 0.30 m³ volume of air and therefore to reach the required L.O.Q. of 0.30 µg/m³.
- 2) For retention capacity: The air sampling was performed at temperature of 35°C and relative humidity of 80% on a 0.30 µg/m³ fortified sample for 6 hours with a 10 L/min air flow rate in order to sample a total volume of air higher than 100 L, according to the guideline SANCO/825/00 rev. 8.1.

Results and discussions

Specificity:

Since the analysis performed by ICP/MS is highly specific, and gave both quantification and identification data, a confirmatory test using another instrumental technique was not necessary and specificity and therefore confirmatory were verified with the same injections and instrumental technique. Any interference from the control sample were < 30 % LOQ.

Linearity:

The Copper nominal concentration tested in injected solutions ranged from 1.00 to 50.00 µg/L, corresponding to a Copper concentration ranging from 0.17 µg/m³ to 8.33 µg/m³ in the air sample and was found to be linear for both ⁶³Cu quantifier and ⁶⁵Cu qualifier isotopes (correlation coefficient > 0.99).

No significant memory effect was detected in the washing dilution medium injected after the highest working standard solution and the range tested for Copper was found to be linear (each correlation coefficient > 0.99).

Limit of quantification:

The limit of quantification (L.O.Q.) was the low fortification level at 0.30 µg/m³ for Copper in air samples, corresponding to a final injected solution of 1.80 µg/L.

The limit of detection (L.O.D.), defined as half of the lowest calibration level, was 0.50 µg/L, corresponding to 0.08 µg/m³ for Copper in air samples.

Accuracy and Precision:

The SANCO/825/00 rev. 8.1 guideline requires mean recoveries for each level in the range from 70 to 110% and a RSD% lower than 20 % for each level. From the data obtained, these criteria were fulfilled and therefore precision and accuracy of the analytical method can be considered acceptable for both ⁶³Cu quantifier and ⁶⁵Cu qualifier isotopes.

Table A 5: Recovery results from method validation of Copper using the analytical method

Matrix	Qualifier isotopes	Fortification level	n	Mean recovery [%]	RSD [%]
Air	⁶³ Cu	0.30 µg/m ³	6	83.7%	2.69%
		2.99 µg/m ³	6	83.0%	1.90%
	⁶⁵ Cu	0.30 µg/m ³	6	84.2%	3.32%
		2.99 µg/m ³	6	84.2%	1.50%

Retention capacity:

The obtained recovery value of 87 % was in the acceptable range which demonstrates that the retention capacity was considered sufficient and no significant breakthrough occurred.

Conclusion

The analytical method for Copper in air was successfully validated under SANCO/825/00 rev 8.1.

A 2.1.2.6 Description of Methods for the Analysis of Body Fluids and Tissues (KCP 5.2)

No additional studies have been submitted, please refer to the dRAR for information.

A 2.1.2.7 Other Studies/ Information

No additional studies have been submitted.