Competent Authority Report

ADDENDUM to Document IIIA, Section 4.2

Study Summaries Active Substance

C_{8-18}-TMAC
(CAS no. 61789-18-2)

Product-type 8
(Wood Preservatives)

Rapporteur Member State: Italy

May 2014

This Addendum supplements Doc. IIIA Section 4.2 of the First Draft Competent Authority Report (CAR) which was prepared by the RMS (Italy) according to Directive 98/8/EC for the purpose of the review of the existing biocidal active substance **Quaternary ammonium compounds, coco alkyltrimethyl, chlorides** (C_{8-18}-TMAC, CAS number 61789-18-2) as Wood Preservative (Product Type 8).

Analytical methods for the determination of C_{8-18}-TMAC residues in soil and water (drinking-, ground- and surface-water), along with the relevant study summary, were submitted by Akzo Noble Surface Chemistry AB in May 2012 in order to fill the data gaps which had been remarked by the RMS following the evaluation of the original Dossier.

The RMS conclusions, resulting from the evaluation of the new documentation, are available under the relevant evaluation boxes. Note that, in order to facilitate readability, two separate evaluation boxes have been compiled by the RMS (one for residues in soil, one for water).

Section A4.2a and 4.2c	Analytical Methods for Detection and Identification **Analytical methods including recovery rates and the limits of determination for the active substance, and for residues thereof, and where relevant in/on the following:** Ground water, surface water, tap water and soil	
	1 REFERENCE	**Official use only**
1.1 Reference	[redacted] (2012), TMAC (lyophilised Arquad C-35) Residue Analytical Method for the Determination in Ground, Surface, Tap Water and Soil, DR.U.NOACK-LABORATORIEN, CRA14111 / 101025AH	
1.2 Data protection	Yes	
1.2.1 Data owner	AkzoNobel Surface Chemistry AB.	
1.2.2		
1.2.3 Criteria for data protection	Data submitted to the MS after 13 May 2000 on existing [a.s. / b.p.] for the purpose of its [entry into Annex I/IA / authorisation]	
	2 GUIDELINES AND QUALITY ASSURANCE	
2.1 Guideline study	Yes, SANCO/825/00 rev. 7 (17/03/04)	X
2.2 GLP	Yes	
2.3 Deviations	Yes; the expiry date of the Surface Water was prolonged after sensoric and analytical verification until 2 months and the test item name was changed according to the study "Content of the Active Ingredients and Relevant Impurities " as specified under Literature.	
	3 MATERIALS AND METHODS	
3.1 Preliminary treatment		
3.1.1 Sample preparation	Spiking solutions of the test item I (100 µg/L) = Mixed standard spiking solution diluted by total factor 10000 from the stock solution with acetonitrile II (1000 µg/L) = Mixed standard spiking solution diluted by total factor 1000 from the stock solution with acetonitrile III (2500 µg/L) = Mixed standard spiking solution diluted by total factor 400 from the stock solution with acetonitrile: HPLC water (60:40) + 1 % formic acid IV (25000 µg/L) = Mixed standard spiking solution diluted by total factor 40 from the stock solution with acetonitrile: HPLC water (60:40) + 1 % formic acid Spiking solutions of the standards V (10 µg/L) = Mixed standard spiking solution diluted by total factor 100000 from active ingredient stock solutions with acetonitrile VI (100 µg/L) = Mixed standard spiking solution diluted by total factor 10000 from active ingredient stock solutions with acetonitrile	X*

Section A4.2a and 4.2c	**Analytical Methods for Detection and Identification** **Analytical methods including recovery rates and the limits of determination for the active substance, and for residues thereof, and where relevant in/on the following:** Ground water, surface water, tap water and soil	
	VII (250 µg/L) = Mixed standard spiking solution diluted by total factor 4000 from active ingredient stock solutions with acetonitrile VIII (2500 µg/L) = Mixed standard spiking solution diluted by total factor 400 from active ingredient stock solutions with acetonitrile IX (1000 µg/L) = Mixed standard spiking solution diluted by total factor 1000 from active ingredient stock solutions with acetonitrile Ground Water, Surface Water and Tap Water 1 x LOQ (0.1 µg/L) for the test item 5 samples of 180 mL corresponding water were spiked with 180 µL of the spiking solution I in a 200 mL measuring flask and filled up to 200 mL with methanol. Samples were given over Strata-X cartridges (conditioned with 10 mL acetonitrile + 0.1% TFA, after that 10 mL acetonitrile + HPLC water (10:90)). The cartridges were washed with 5 mL methanol : HPLC water (10:90). After drying, the cartridges were eluted with 10 mL acetonitrile + 0.1 % TFA and evaporated to dryness at 40 °C on the rotary evaporator. The residue was taken up in 1 mL acetonitrile : HPLC water + 1% HCOOH (60:40) under 30 seconds sonification. 10 x LOQ (1.0 µg/L) for the test item 5 samples of 180 mL corresponding water were spiked with 180 µL of the spiking solution II in a 200 mL measuring flask and filled up to 200 mL with methanol. Further steps according to 1x LOQ. 1 x LOQ (0.014 µg/L) for the active ingredients 5 samples of 180 mL corresponding water were spiked with 252 µL of the spiking solution V in a 200 mL measuring flask and filled up to 200 mL with methanol. Further steps according to 1x LOQ of the test item. 10 x LOQ (0.14 µg/L) for the active ingredients 5 samples of 180 mL corresponding water were spiked with 252 µL of the spiking solution VI in a 200 mL measuring flask and filled up to 200 mL with methanol. Further steps according to 1x LOQ of the test item. Control 2 samples of 180 mL corresponding water were prepared each for the test item and compounds as described for the 1 x LOQ. Matrix effect Matrix Control for TMAC (lyophilised Arquad C-35): 50 µL of the spiking solution I were added to 0.45 mL of one control sample	X**

Section A4.2a and 4.2c	**Analytical Methods for Detection and Identification** **Analytical methods including recovery rates and the limits of determination for the active substance, and for residues thereof, and where relevant in/on the following:** Ground water, surface water, tap water and soil	
	after sample preparation. Matrix Control for C8-, C10-, C12-, C14-, C16-, C18- and C18-unsaturated trimethylammonium chloride: 50 µL of the spiking solution VI were added to 0.45 mL of one control sample after sample preparation. Soil 1 x LOQ (0.05 mg/kg) for the test item 5 samples of 5 g soil were weighed into 50 mL centrifuge tubes, 0.505 mL HPLC water, spiked with 100 µL of the spiking solution III and then homogenized. 10 mL acetonitrile + 1 % TFA were added and treated for 10 minutes on the rotary shaker at 80 rounds per minute, then sonificated at room temperature for 5 minutes. After the extraction, the samples get centrifuged for 5 minutes on 3000 rounds per minute and then the supernatant was transferred to a new 50 mL centrifuge tube. This extraction was repeated twice, but the second and third extraction contained 5 mL acetonitrile + 1 % TFA instead of 10 mL. All 3 supernatants were united in the corresponding centrifuge tube filled up to 20 mL with acetonitrile and centrifuged again under same conditions. 10 x LOQ (0.5 mg/kg) for the test item 5 samples of 5 g soil were weighed into 50 mL centrifuge tubes, 0.505 mL HPLC water, spiked with 100 µL of the spiking solution IV and then homogenized. Further steps according to 1x LOQ. 1 x LOQ (0.00714 mg/kg) for the active ingredients 5 samples of 5 g soil were weighed into 50 mL centrifuge tubes, humidified with 0.505 mL HPLC water, spiked with 142.8 µL of the spiking solution VII and then homgenized. Further steps according to 1x LOQ of the test item. 10 x LOQ (0.0714 mg/kg) for the active ingredients 5 samples of 5 g soil were weighed into 50 mL centrifuge tubes, humidified with 0.505 mL HPLC water, spiked with 142.8 µL of the spiking solution VIII and then homgenized. Further steps according to 1x LOQ of the test item. Control 2 samples of 5 g soil were prepared without spiking as decribed for the 1 x LOQ.	

Section A4.2a and 4.2c	**Analytical Methods for Detection and Identification** **Analytical methods including recovery rates and the limits of determination for the active substance, and for residues thereof, and where relevant in/on the following:** Ground water, surface water, tap water and soil		
	Matrix effect Matrix Control for TMAC (lyophilised Arquad C-35): 10 µL of the spiking solution III were added to 0.99 mL of one control sample after sample preparation. Matrix Control for C8-, C10-, C12-, C14-, C16-, C18- and C18-unsaturated trimethylammonium chloride: 10 µL of the spiking solution IX were added to 0.99 mL of one control sample after sample preparation.		X*
3.1.2 Cleanup	Not performed		
3.2 Detection			
3.2.1 Separation method	UPLC-MS/MS		
3.2.2 Detector	Autosampler	Acquity UPLC, Waters	
	Binary Solvent Manager	Acquity UPLC, Waters	
	Column Manager	Acquity UPLC, Waters	
	Detection	Mass selective detector, Xevo, Acquity UPLC, Waters	
	Software	MassLynxTM 4.1, SCN 729, Waters	
	Analytical balance	LA230S, Sartorius	
	Balance	EW3000-2M, Kern	
	SPE cartridges	Strata-X Polymeric RP (200 mg), batch no. S300-144, Phenomenex	
	Rotary evaporator	Büchi	
	Ultrasonic bath	Sonorex Super RK 510 H, Bandelin	
	Rotary shaker	REAX 2, Heidolph	
	Centrifuge	Megafuge 1.0, Heraeus	
	Displacement pipettes	10-100 µL, Gilson 50-250 µL, Gilson 100 -1000 µL, Gilson	
	Piston stroke pipette	2 - 20 µL, Thermo Scientific 20 - 200 µL, Thermo Scientific 200- 1000 µL, Thermo Scientific	
	Reagents	Acetonitrile, gradient grade, VWR HPLC-water, gradient grade, VWR Methanol, gradient grade, VWR Formic acid (HCOOH), 98-100 %, Sigma-Aldrich Trifluoro acetic acid (TFA), peptid analysis, Roth	

Section A4.2a and 4.2c	**Analytical Methods for Detection and Identification** **Analytical methods including recovery rates and the limits of determination for the active substance, and for residues thereof, and where relevant in/on the following:** Ground water, surface water, tap water and soil	

CONDITIONS OF ANALYSIS

Column	Acquity UPLC HSS C18, 1.8 µm, 50 x 2.1 mm, batch 0107, Waters
Column Temperature	30 °C
Mobile Phase	Pump A: HPLC water + 1 % formic acid Pump B: acetonitrile + 1 % formic acid

Gradient table

Time [min]	A [%]	B [%]
0.0	45	55
1.0	45	55
3.0	25	75
5.0	25	75
5.1	5	95
6.0	5	95
6.1	45	55
7.0	45	55

Flow rate	0.5 mL/min
Run time	7.0 minutes
Injection volume	3.0 µL (PLUNO)

CONDITIONS OF DETECTION

Type	Multi Reaction Mode (MRM)
Ionisation mode	Electrospray positive

Detector conditions

Condition	Active Ingredient			
	C8-TMAC	C10-TMAC	C12-TMAC	C-14 TMAC
Precursor ion [Da]	172.24	200.27	228.30	256.33
Product ion, Quan [Da]	60.18	60.17	60.17	60.17
Product ion, Conf [Da]	57.18	57.18	57.18	57.18
Dwell time [s]	0.018	0.018	0.018	0.018
Cone Voltage [V]	34	40	40	46
Collision energy, Quan [eV]	20	22	24	26
Collision energy, Conf [eV]	22	24	26	26

<table>
<tr><td>Section A4.2a and 4.2c</td><td colspan="2">Analytical Methods for Detection and Identification
Analytical methods including recovery rates and the limits of determination for the active substance, and for residues thereof, and where relevant in/on the following:
Ground water, surface water, tap water and soil</td></tr>
<tr><td></td><td colspan="2">
<table>
<tr><th rowspan="2">Condition</th><th colspan="3">Active Ingredient</th></tr>
<tr><th>C16-TMAC</th><th>C18-TMAC unsaturated</th><th>C-18 TMAC</th></tr>
<tr><td>Precursor ion [Da]</td><td>284.43</td><td>310.42</td><td>312.46</td></tr>
<tr><td>Product ion, Quan [Da]</td><td>60.17</td><td>60.17</td><td>60.17</td></tr>
<tr><td>Product ion, Conf [Da]</td><td>57.18</td><td>69.09</td><td>57.18</td></tr>
<tr><td>Dwell time [s]</td><td>0.018</td><td>0.018</td><td>0.018</td></tr>
<tr><td>Cone Voltage [V]</td><td>48</td><td>52</td><td>52</td></tr>
<tr><td>Collision energy, Quan [eV]</td><td>28</td><td>30</td><td>28</td></tr>
<tr><td>Collision energy, Conf [eV]</td><td>28</td><td>30</td><td>30</td></tr>
</table>

Capillary voltage 1 kV
Source temperature 150 °C
Cone gas flow (N_2) 150 L/h
Desolvation gas temperature 400 °C
Desolvation gas flow (N_2) 1000 L/h
Collision gas pressure (Ar) approx. 3.01 x 10^{-3} mbar</td><td></td></tr>
<tr><td>3.2.3 Standard(s)</td><td colspan="2">Octyltrimethylammonium chloride (Lab. ID 14362)
Decyltrimethylammonium chloride (Lab. ID 14363)
Dodecyltrimethylammonium chloride (Lab. ID 14364)
Tetradecyltrimethylammonium chloride (Lab. ID 14365)
Hexadecyltrimethylammonium chloride (Lab. ID 14366)
Stearyltrimethylammonium chloride (Lab. ID 14367)
Arquad O-50 (C18 unsaturated) (Lab. ID 14368)</td><td></td></tr>
<tr><td>3.2.4 Interfering substance(s)</td><td colspan="2">None</td><td></td></tr>
<tr><td>3.3 Linearity</td><td colspan="2"></td><td></td></tr>
<tr><td>3.3.1 Calibration range</td><td colspan="2">1 – 100 µg/L for C8-, C10-, C12-, C14-, C16-, C18- and C18-unsaturated trimethylammonium chloride</td><td>X1</td></tr>
<tr><td>3.3.2 Number of measurements</td><td colspan="2">6 concentrations</td><td>X1</td></tr>
<tr><td>3.3.3 Linearity</td><td colspan="2">The analytical system gave a linear response in the nominal range of 1 – 100 µg/L for C8-, C10-, C12-, C14-, C16-, C18- and C18-unsaturated trimethylammonium chloride. The linearity R^2 was determined to be > 0.992.</td><td>X1</td></tr>
</table>

Section A4.2a and 4.2c

Analytical Methods for Detection and Identification

Analytical methods including recovery rates and the limits of determination for the active substance, and for residues thereof, and where relevant in/on the following:

Ground water, surface water, tap water and soil

3.4 Specificity: interfering substances

Specificity is given by the mass spectrometric method, due to the formation of specific precursor and product ions of the test item for both quantification and confirmation methods. The mean responses of the blank values of the corresponding two control samples were lower than 30 % of LOQ for each test system. Except for tap water, there was a slightly higher amount of C16-TMAC which was subtracted from the mean measured concentrations.

X2

3.5 Recovery rates at different levels

Recovery rates of TMAC (lyophilised Arquad C-35)

Matrix	Component	Method	Concentration	LOQ Level	Mean Recovery Rate [%]
Ground water	C_8-TMAC	Quantification	0.1 µg/L	1x LOQ	< LOQ
			1.0 µg/L	10x LOQ	90
		Confirmation	0.1 µg/L	1x LOQ	< LOQ
			1.0 µg/L	10x LOQ	93
	C_{10}-TMAC	Quantification	0.1 µg/L	1x LOQ	< LOQ
			1.0 µg/L	10x LOQ	93
		Confirmation	0.1 µg/L	1x LOQ	< LOQ
			1.0 µg/L	10x LOQ	91
	C_{12}-TMAC	Quantification	0.1 µg/L	1x LOQ	83
			1.0 µg/L	10x LOQ	93
		Confirmation	0.1 µg/L	1x LOQ	86
			1.0 µg/L	10x LOQ	95
	C_{14}-TMAC	Quantification	0.1 µg/L	1x LOQ	81
			1.0 µg/L	10x LOQ	94
		Confirmation	0.1 µg/L	1x LOQ	84
			1.0 µg/L	10x LOQ	92
	C_{16}-TMAC	Quantification	0.1 µg/L	1x LOQ	< LOQ
			1.0 µg/L	10x LOQ	79
		Confirmation	0.1 µg/L	1x LOQ	< LOQ
			1.0 µg/L	10x LOQ	81
	C_{18}-TMAC	Quantification	0.1 µg/L	1x LOQ	< LOQ
			1.0 µg/L	10x LOQ	72
		Confirmation	0.1 µg/L	1x LOQ	< LOQ
			1.0 µg/L	10x LOQ	80
	C_{18}-TMAC unsat.	Quantification	0.1 µg/L	1x LOQ	< LOQ
			1.0 µg/L	10x LOQ	71
		Confirmation	0.1 µg/L	1x LOQ	< LOQ
			1.0 µg/L	10x LOQ	71

Section A4.2a and 4.2c

Analytical Methods for Detection and Identification

Analytical methods including recovery rates and the limits of determination for the active substance, and for residues thereof, and where relevant in/on the following:

Ground water, surface water, tap water and soil

Matrix	Component	Method	Concentration	LOQ Level	Mean Recovery Rate [%]
Surface water	C_8-TMAC	Quantification	0.1 µg/L	1x LOQ	< LOQ
			1.0 µg/L	10x LOQ	81
		Confirmation	0.1 µg/L	1x LOQ	< LOQ
			1.0 µg/L	10x LOQ	82
	C_{10}-TMAC	Quantification	0.1 µg/L	1x LOQ	< LOQ
			1.0 µg/L	10x LOQ	85
		Confirmation	0.1 µg/L	1x LOQ	< LOQ
			1.0 µg/L	10x LOQ	84
	C_{12}-TMAC	Quantification	0.1 µg/L	1x LOQ	82
			1.0 µg/L	10x LOQ	89
		Confirmation	0.1 µg/L	1x LOQ	82
			1.0 µg/L	10x LOQ	89
	C_{14}-TMAC	Quantification	0.1 µg/L	1x LOQ	81
			1.0 µg/L	10x LOQ	88
		Confirmation	0.1 µg/L	1x LOQ	94
			1.0 µg/L	10x LOQ	88
	C_{16}-TMAC	Quantification	0.1 µg/L	1x LOQ	< LOQ
			1.0 µg/L	10x LOQ	77
		Confirmation	0.1 µg/L	1x LOQ	< LOQ
			1.0 µg/L	10x LOQ	79
	C_{18}-TMAC	Quantification	0.1 µg/L	1x LOQ	< LOQ
			1.0 µg/L	10x LOQ	72
		Confirmation	0.1 µg/L	1x LOQ	< LOQ
			1.0 µg/L	10x LOQ	74
	C_{18}-TMAC unsat.	Quantification	0.1 µg/L	1x LOQ	< LOQ
			1.0 µg/L	10x LOQ	71
		Confirmation	0.1 µg/L	1x LOQ	< LOQ
			1.0 µg/L	10x LOQ	73

Section A4.2a and 4.2c

Analytical Methods for Detection and Identification

Analytical methods including recovery rates and the limits of determination for the active substance, and for residues thereof, and where relevant in/on the following:

Ground water, surface water, tap water and soil

Matrix	Component	Method	Concentration	LOQ Level	Mean Recovery Rate [%]
Tap water	C_8-TMAC	Quantification	0.1 µg/L	1x LOQ	< LOQ
			1.0 µg/L	10x LOQ	88
		Confirmation	0.1 µg/L	1x LOQ	< LOQ
			1.0 µg/L	10x LOQ	94
	C_{10}-TMAC	Quantification	0.1 µg/L	1x LOQ	< LOQ
			1.0 µg/L	10x LOQ	93
		Confirmation	0.1 µg/L	1x LOQ	< LOQ
			1.0 µg/L	10x LOQ	93
	C_{12}-TMAC	Quantification	0.1 µg/L	1x LOQ	81
			1.0 µg/L	10x LOQ	94
		Confirmation	0.1 µg/L	1x LOQ	84
			1.0 µg/L	10x LOQ	94
	C_{14}-TMAC	Quantification	0.1 µg/L	1x LOQ	82
			1.0 µg/L	10x LOQ	95
		Confirmation	0.1 µg/L	1x LOQ	85
			1.0 µg/L	10x LOQ	94
	C_{16}-TMAC	Quantification	0.1 µg/L	1x LOQ	< LOQ
			1.0 µg/L	10x LOQ	85
		Confirmation	0.1 µg/L	1x LOQ	< LOQ
			1.0 µg/L	10x LOQ	84
	C_{18}-TMAC	Quantification	0.1 µg/L	1x LOQ	< LOQ
			1.0 µg/L	10x LOQ	85
		Confirmation	0.1 µg/L	1x LOQ	< LOQ
			1.0 µg/L	10x LOQ	99
	C_{18}-TMAC unsat.	Quantification	0.1 µg/L	1x LOQ	< LOQ
			1.0 µg/L	10x LOQ	76
		Confirmation	0.1 µg/L	1x LOQ	< LOQ
			1.0 µg/L	10x LOQ	78

Section A4.2a and 4.2c

Analytical Methods for Detection and Identification

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Ground water, surface water, tap water and soil

Matrix	Component	Method	Concentration	LOQ Level	Mean Recovery Rate [%]
Soil	C_8-TMAC	Quantification	0.05 mg/kg	1x LOQ	< LOQ
			0.5 mg/kg	10x LOQ	95
		Confirmation	0.05 mg/kg	1x LOQ	< LOQ
			0.5 mg/kg	10x LOQ	95
	C_{10}-TMAC	Quantification	0.05 mg/kg	1x LOQ	< LOQ
			0.5 mg/kg	10x LOQ	96
		Confirmation	0.05 mg/kg	1x LOQ	< LOQ
			0.5 mg/kg	10x LOQ	98
	C_{12}-TMAC	Quantification	0.05 mg/kg	1x LOQ	98
			0.5 mg/kg	10x LOQ	99
		Confirmation	0.05 mg/kg	1x LOQ	95
			0.5 mg/kg	10x LOQ	99
	C_{14}-TMAC	Quantification	0.05 mg/kg	1x LOQ	93
			0.5 mg/kg	10x LOQ	96
		Confirmation	0.05 mg/kg	1x LOQ	95
			0.5 mg/kg	10x LOQ	95
	C_{16}-TMAC	Quantification	0.05 mg/kg	1x LOQ	< LOQ
			0.5 mg/kg	10x LOQ	82
		Confirmation	0.05 mg/kg	1x LOQ	< LOQ
			0.5 mg/kg	10x LOQ	85
	C_{18}-TMAC	Quantification	0.05 mg/kg	1x LOQ	< LOQ
			0.5 mg/kg	10x LOQ	82
		Confirmation	0.05 mg/kg	1x LOQ	< LOQ
			0.5 mg/kg	10x LOQ	81
	C_{18}-TMAC unsat.	Quantification	0.05 mg/kg	1x LOQ	< LOQ
			0.5 mg/kg	10x LOQ	90
		Confirmation	0.05 mg/kg	1x LOQ	< LOQ
			0.5 mg/kg	10x LOQ	90

Section A4.2a and 4.2c

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Ground water, surface water, tap water and soil

Recovery rates of active ingredients

Matrix	Component	Method	Concentration	LOQ Level	Mean Recovery Rate [%]
Ground water	C_8-TMAC	Quantification	0.014 µg/L	1x LOQ	89
			0.14 µg/L	10x LOQ	91
		Confirmation	0.014 µg/L	1x LOQ	95
			0.14 µg/L	10x LOQ	93
	C_{10}-TMAC	Quantification	0.014 µg/L	1x LOQ	95
			0.14 µg/L	10x LOQ	96
		Confirmation	0.014 µg/L	1x LOQ	96
			0.14 µg/L	10x LOQ	94
	C_{12}-TMAC	Quantification	0.014 µg/L	1x LOQ	101
			0.14 µg/L	10x LOQ	95
		Confirmation	0.014 µg/L	1x LOQ	103
			0.14 µg/L	10x LOQ	96
	C_{14}-TMAC	Quantification	0.014 µg/L	1x LOQ	93
			0.14 µg/L	10x LOQ	94
		Confirmation	0.014 µg/L	1x LOQ	96
			0.14 µg/L	10x LOQ	92
	C_{16}-TMAC	Quantification	0.014 µg/L	1x LOQ	99
			0.14 µg/L	10x LOQ	86
		Confirmation	0.014 µg/L	1x LOQ	97
			0.14 µg/L	10x LOQ	86
	C_{18}-TMAC	Quantification	0.014 µg/L	1x LOQ	74
			0.14 µg/L	10x LOQ	72
		Confirmation	0.014 µg/L	1x LOQ	79
			0.14 µg/L	10x LOQ	73
	C_{18}-TMAC unsat.	Quantification	0.014 µg/L	1x LOQ	73
			0.14 µg/L	10x LOQ	79
		Confirmation	0.014 µg/L	1x LOQ	79
			0.14 µg/L	10x LOQ	80

Section A4.2a and 4.2c

Analytical Methods for Detection and Identification

Analytical methods including recovery rates and the limits of determination for the active substance, and for residues thereof, and where relevant in/on the following:

Ground water, surface water, tap water and soil

Matrix	Component	Method	Concentration	LOQ Level	Mean Recovery Rate [%]
Surface water	C_8-TMAC	Quantification	0.014 µg/L	1x LOQ	91
			0.14 µg/L	10x LOQ	89
		Confirmation	0.014 µg/L	1x LOQ	96
			0.14 µg/L	10x LOQ	90
	C_{10}-TMAC	Quantification	0.014 µg/L	1x LOQ	94
			0.14 µg/L	10x LOQ	92
		Confirmation	0.014 µg/L	1x LOQ	101
			0.14 µg/L	10x LOQ	94
	C_{12}-TMAC	Quantification	0.014 µg/L	1x LOQ	101
			0.14 µg/L	10x LOQ	92
		Confirmation	0.014 µg/L	1x LOQ	102
			0.14 µg/L	10x LOQ	95
	C_{14}-TMAC	Quantification	0.014 µg/L	1x LOQ	95
			0.14 µg/L	10x LOQ	92
		Confirmation	0.014 µg/L	1x LOQ	92
			0.14 µg/L	10x LOQ	93
	C_{16}-TMAC	Quantification	0.014 µg/L	1x LOQ	96
			0.14 µg/L	10x LOQ	88
		Confirmation	0.014 µg/L	1x LOQ	87
			0.14 µg/L	10x LOQ	87
	C_{18}-TMAC	Quantification	0.014 µg/L	1x LOQ	76
			0.14 µg/L	10x LOQ	73
		Confirmation	0.014 µg/L	1x LOQ	76
			0.14 µg/L	10x LOQ	72
	C_{18}-TMAC unsat.	Quantification	0.014 µg/L	1x LOQ	74
			0.14 µg/L	10x LOQ	76
		Confirmation	0.014 µg/L	1x LOQ	82
			0.14 µg/L	10x LOQ	76

Section A4.2a and 4.2c

Analytical Methods for Detection and Identification

Analytical methods including recovery rates and the limits of determination for the active substance, and for residues thereof, and where relevant in/on the following:

Ground water, surface water, tap water and soil

Matrix	Component	Method	Concentration	LOQ Level	Mean Recovery Rate [%]
Tap water	C_8-TMAC	Quantification	0.014 µg/L	1x LOQ	92
			0.14 µg/L	10x LOQ	89
		Confirmation	0.014 µg/L	1x LOQ	97
			0.14 µg/L	10x LOQ	92
	C_{10}-TMAC	Quantification	0.014 µg/L	1x LOQ	98
			0.14 µg/L	10x LOQ	94
		Confirmation	0.014 µg/L	1x LOQ	98
			0.14 µg/L	10x LOQ	94
	C_{12}-TMAC	Quantification	0.014 µg/L	1x LOQ	100
			0.14 µg/L	10x LOQ	96
		Confirmation	0.014 µg/L	1x LOQ	102
			0.14 µg/L	10x LOQ	96
	C_{14}-TMAC	Quantification	0.014 µg/L	1x LOQ	96
			0.14 µg/L	10x LOQ	95
		Confirmation	0.014 µg/L	1x LOQ	92
			0.14 µg/L	10x LOQ	94
	C_{16}-TMAC	Quantification	0.014 µg/L	1x LOQ	77
			0.14 µg/L	10x LOQ	92
		Confirmation	0.014 µg/L	1x LOQ	76
			0.14 µg/L	10x LOQ	89
	C_{18}-TMAC	Quantification	0.014 µg/L	1x LOQ	90
			0.14 µg/L	10x LOQ	86
		Confirmation	0.014 µg/L	1x LOQ	87
			0.14 µg/L	10x LOQ	83
	C_{18}-TMAC unsat.	Quantification	0.014 µg/L	1x LOQ	76
			0.14 µg/L	10x LOQ	71
		Confirmation	0.014 µg/L	1x LOQ	80
			0.14 µg/L	10x LOQ	79

Section A4.2a and 4.2c

Analytical Methods for Detection and Identification

Analytical methods including recovery rates and the limits of determination for the active substance, and for residues thereof, and where relevant in/on the following:

Ground water, surface water, tap water and soil

Matrix	Component	Method	Concentration	LOQ Level	Mean Recovery Rate [%]
Soil	C_8-TMAC	Quantification	0.00714 mg/kg	1x LOQ	90
			0.0714 mg/kg	10x LOQ	89
		Confirmation	0.00714 mg/kg	1x LOQ	95
			0.0714 mg/kg	10x LOQ	92
	C_{10}-TMAC	Quantification	0.00714 mg/kg	1x LOQ	91
			0.0714 mg/kg	10x LOQ	94
		Confirmation	0.00714 mg/kg	1x LOQ	93
			0.0714 mg/kg	10x LOQ	92
	C_{12}-TMAC	Quantification	0.00714 mg/kg	1x LOQ	91
			0.0714 mg/kg	10x LOQ	93
		Confirmation	0.00714 mg/kg	1x LOQ	94
			0.0714 mg/kg	10x LOQ	95
	C_{14}-TMAC	Quantification	0.00714 mg/kg	1x LOQ	86
			0.0714 mg/kg	10x LOQ	89
		Confirmation	0.00714 mg/kg	1x LOQ	88
			0.0714 mg/kg	10x LOQ	91
	C_{16}-TMAC	Quantification	0.00714 mg/kg	1x LOQ	80
			0.0714 mg/kg	10x LOQ	88
		Confirmation	0.00714 mg/kg	1x LOQ	85
			0.0714 mg/kg	10x LOQ	90
	C_{18}-TMAC	Quantification	0.00714 mg/kg	1x LOQ	74
			0.0714 mg/kg	10x LOQ	80
		Confirmation	0.00714 mg/kg	1x LOQ	78
			0.0714 mg/kg	10x LOQ	82
	C_{18}-TMAC unsat.	Quantification	0.00714 mg/kg	1x LOQ	89
			0.0714 mg/kg	10x LOQ	93
		Confirmation	0.00714 mg/kg	1x LOQ	97
			0.0714 mg/kg	10x LOQ	96

3.5.1 Relative	Relative standard deviations of TMAC (lyophilised Arquad C-35)

Section A4.2a and 4.2c

Analytical Methods for Detection and Identification

Analytical methods including recovery rates and the limits of determination for the active substance, and for residues thereof, and where relevant in/on the following:

Ground water, surface water, tap water and soil

standard deviation

Matrix	Component	Method	Concentration	LOQ Level	Mean Recovery Rate [%]
Ground water	C_8-TMAC	Quantification	0.1 µg/L	1x LOQ	< LOQ
			1.0 µg/L	10x LOQ	4.05
		Confirmation	0.1 µg/L	1x LOQ	< LOQ
			1.0 µg/L	10x LOQ	6.52
	C_{10}-TMAC	Quantification	0.1 µg/L	1x LOQ	< LOQ
			1.0 µg/L	10x LOQ	2.19
		Confirmation	0.1 µg/L	1x LOQ	< LOQ
			1.0 µg/L	10x LOQ	2.49
	C_{12}-TMAC	Quantification	0.1 µg/L	1x LOQ	4.07
			1.0 µg/L	10x LOQ	2.96
		Confirmation	0.1 µg/L	1x LOQ	3.88
			1.0 µg/L	10x LOQ	2.91
	C_{14}-TMAC	Quantification	0.1 µg/L	1x LOQ	4.14
			1.0 µg/L	10x LOQ	3.47
		Confirmation	0.1 µg/L	1x LOQ	3.97
			1.0 µg/L	10x LOQ	3.93
	C_{16}-TMAC	Quantification	0.1 µg/L	1x LOQ	< LOQ
			1.0 µg/L	10x LOQ	5.51
		Confirmation	0.1 µg/L	1x LOQ	< LOQ
			1.0 µg/L	10x LOQ	6.06
	C_{18}-TMAC	Quantification	0.1 µg/L	1x LOQ	< LOQ
			1.0 µg/L	10x LOQ	9.58
		Confirmation	0.1 µg/L	1x LOQ	< LOQ
			1.0 µg/L	10x LOQ	5.07
	C_{18}-TMAC unsat.	Quantification	0.1 µg/L	1x LOQ	< LOQ
			1.0 µg/L	10x LOQ	5.81
		Confirmation	0.1 µg/L	1x LOQ	< LOQ
			1.0 µg/L	10x LOQ	4.74

Section A4.2a and 4.2c	**Analytical Methods for Detection and Identification** **Analytical methods including recovery rates and the limits of determination for the active substance, and for residues thereof, and where relevant in/on the following:** Ground water, surface water, tap water and soil	

Matrix	Component	Method	Concentration	LOQ Level	Mean Recovery Rate [%]
Surface water	C_8-TMAC	Quantification	0.1 µg/L	1x LOQ	< LOQ
			1.0 µg/L	10x LOQ	4.97
		Confirmation	0.1 µg/L	1x LOQ	< LOQ
			1.0 µg/L	10x LOQ	4.21
	C_{10}-TMAC	Quantification	0.1 µg/L	1x LOQ	< LOQ
			1.0 µg/L	10x LOQ	4.78
		Confirmation	0.1 µg/L	1x LOQ	< LOQ
			1.0 µg/L	10x LOQ	5.27
	C_{12}-TMAC	Quantification	0.1 µg/L	1x LOQ	0.803
			1.0 µg/L	10x LOQ	7.81
		Confirmation	0.1 µg/L	1x LOQ	2.10
			1.0 µg/L	10x LOQ	5.17
	C_{14}-TMAC	Quantification	0.1 µg/L	1x LOQ	1.012
			1.0 µg/L	10x LOQ	5.14
		Confirmation	0.1 µg/L	1x LOQ	2.38
			1.0 µg/L	10x LOQ	4.65
	C_{16}-TMAC	Quantification	0.1 µg/L	1x LOQ	< LOQ
			1.0 µg/L	10x LOQ	3.63
		Confirmation	0.1 µg/L	1x LOQ	< LOQ
			1.0 µg/L	10x LOQ	3.76
	C_{18}-TMAC	Quantification	0.1 µg/L	1x LOQ	< LOQ
			1.0 µg/L	10x LOQ	7.22
		Confirmation	0.1 µg/L	1x LOQ	< LOQ
			1.0 µg/L	10x LOQ	7.23
	C_{18}-TMAC unsat.	Quantification	0.1 µg/L	1x LOQ	< LOQ
			1.0 µg/L	10x LOQ	6.42
		Confirmation	0.1 µg/L	1x LOQ	< LOQ
			1.0 µg/L	10x LOQ	4.58

Section A4.2a and 4.2c

Analytical Methods for Detection and Identification

Analytical methods including recovery rates and the limits of determination for the active substance, and for residues thereof, and where relevant in/on the following:

Ground water, surface water, tap water and soil

Matrix	Component	Method	Concentration	LOQ Level	Mean Recovery Rate [%]
Tap water	C_8-TMAC	Quantification	0.1 µg/L	1x LOQ	< LOQ
			1.0 µg/L	10x LOQ	4.52
		Confirmation	0.1 µg/L	1x LOQ	< LOQ
			1.0 µg/L	10x LOQ	3.06
	C_{10}-TMAC	Quantification	0.1 µg/L	1x LOQ	< LOQ
			1.0 µg/L	10x LOQ	1.57
		Confirmation	0.1 µg/L	1x LOQ	< LOQ
			1.0 µg/L	10x LOQ	6.89
	C_{12}-TMAC	Quantification	0.1 µg/L	1x LOQ	3.63
			1.0 µg/L	10x LOQ	3.19
		Confirmation	0.1 µg/L	1x LOQ	5.91
			1.0 µg/L	10x LOQ	2.85
	C_{14}-TMAC	Quantification	0.1 µg/L	1x LOQ	2.3
			1.0 µg/L	10x LOQ	2.69
		Confirmation	0.1 µg/L	1x LOQ	6.94
			1.0 µg/L	10x LOQ	4.50
	C_{16}-TMAC	Quantification	0.1 µg/L	1x LOQ	< LOQ
			1.0 µg/L	10x LOQ	3.51
		Confirmation	0.1 µg/L	1x LOQ	< LOQ
			1.0 µg/L	10x LOQ	4.79
	C_{18}-TMAC	Quantification	0.1 µg/L	1x LOQ	< LOQ
			1.0 µg/L	10x LOQ	3.42
		Confirmation	0.1 µg/L	1x LOQ	< LOQ
			1.0 µg/L	10x LOQ	3.30
	C_{18}-TMAC unsat.	Quantification	0.1 µg/L	1x LOQ	< LOQ
			1.0 µg/L	10x LOQ	3.29
		Confirmation	0.1 µg/L	1x LOQ	< LOQ
			1.0 µg/L	10x LOQ	4.47

Section A4.2a and 4.2c

Analytical Methods for Detection and Identification

Analytical methods including recovery rates and the limits of determination for the active substance, and for residues thereof, and where relevant in/on the following:

Ground water, surface water, tap water and soil

Matrix	Component	Method	Concentration	LOQ Level	Mean Recovery Rate [%]
Soil	C_8-TMAC	Quantification	0.05 mg/kg	1x LOQ	< LOQ
			0.5 mg/kg	10x LOQ	2.01
		Confirmation	0.05 mg/kg	1x LOQ	< LOQ
			0.5 mg/kg	10x LOQ	5.34
	C_{10}-TMAC	Quantification	0.05 mg/kg	1x LOQ	< LOQ
			0.5 mg/kg	10x LOQ	3.11
		Confirmation	0.05 mg/kg	1x LOQ	< LOQ
			0.5 mg/kg	10x LOQ	3.21
	C_{12}-TMAC	Quantification	0.05 mg/kg	1x LOQ	1.02
			0.5 mg/kg	10x LOQ	1.41
		Confirmation	0.05 mg/kg	1x LOQ	3.49
			0.5 mg/kg	10x LOQ	1.00
	C_{14}-TMAC	Quantification	0.05 mg/kg	1x LOQ	4.14
			0.5 mg/kg	10x LOQ	3.97
		Confirmation	0.05 mg/kg	1x LOQ	2.50
			0.5 mg/kg	10x LOQ	3.75
	C_{16}-TMAC	Quantification	0.05 mg/kg	1x LOQ	< LOQ
			0.5 mg/kg	10x LOQ	3.89
		Confirmation	0.05 mg/kg	1x LOQ	< LOQ
			0.5 mg/kg	10x LOQ	3.75
	C_{18}-TMAC	Quantification	0.05 mg/kg	1x LOQ	< LOQ
			0.5 mg/kg	10x LOQ	5.92
		Confirmation	0.05 mg/kg	1x LOQ	< LOQ
			0.5 mg/kg	10x LOQ	8.01
	C_{18}-TMAC unsat.	Quantification	0.05 mg/kg	1x LOQ	< LOQ
			0.5 mg/kg	10x LOQ	3.24
		Confirmation	0.05 mg/kg	1x LOQ	< LOQ
			0.5 mg/kg	10x LOQ	3.04

Section A4.2a and 4.2c	**Analytical Methods for Detection and Identification** **Analytical methods including recovery rates and the limits of determination for the active substance, and for residues thereof, and where relevant in/on the following:** Ground water, surface water, tap water and soil	

Relative standard deviations of active ingredients

Matrix	Component	Method	Concentration	LOQ Level	Mean Recovery Rate [%]
Ground water	C_8-TMAC	Quantification	0.014 µg/L	1x LOQ	3.28
			0.14 µg/L	10x LOQ	4.43
		Confirmation	0.014 µg/L	1x LOQ	6.17
			0.14 µg/L	10x LOQ	3.12
	C_{10}-TMAC	Quantification	0.014 µg/L	1x LOQ	4.3
			0.14 µg/L	10x LOQ	3.73
		Confirmation	0.014 µg/L	1x LOQ	6.27
			0.14 µg/L	10x LOQ	4.43
	C_{12}-TMAC	Quantification	0.014 µg/L	1x LOQ	8.23
			0.14 µg/L	10x LOQ	3.65
		Confirmation	0.014 µg/L	1x LOQ	4.24
			0.14 µg/L	10x LOQ	3.05
	C_{14}-TMAC	Quantification	0.014 µg/L	1x LOQ	2.7
			0.14 µg/L	10x LOQ	3.24
		Confirmation	0.014 µg/L	1x LOQ	5.17
			0.14 µg/L	10x LOQ	3.69
	C_{16}-TMAC	Quantification	0.014 µg/L	1x LOQ	4.25
			0.14 µg/L	10x LOQ	4.15
		Confirmation	0.014 µg/L	1x LOQ	3.29
			0.14 µg/L	10x LOQ	3.93
	C_{18}-TMAC	Quantification	0.014 µg/L	1x LOQ	3.0
			0.14 µg/L	10x LOQ	1.80
		Confirmation	0.014 µg/L	1x LOQ	7.5
			0.14 µg/L	10x LOQ	2.69
	C_{18}-TMAC unsat.	Quantification	0.014 µg/L	1x LOQ	3.83
			0.14 µg/L	10x LOQ	3.3
		Confirmation	0.014 µg/L	1x LOQ	6.15
			0.14 µg/L	10x LOQ	3.89

Section A4.2a and 4.2c

Analytical Methods for Detection and Identification

Analytical methods including recovery rates and the limits of determination for the active substance, and for residues thereof, and where relevant in/on the following:

Ground water, surface water, tap water and soil

Matrix	Component	Method	Concentration	LOQ Level	Mean Recovery Rate [%]
Surface water	C_8-TMAC	Quantification	0.014 µg/L	1x LOQ	5.24
			0.14 µg/L	10x LOQ	5.38
		Confirmation	0.014 µg/L	1x LOQ	4.90
			0.14 µg/L	10x LOQ	4.06
	C_{10}-TMAC	Quantification	0.014 µg/L	1x LOQ	4.34
			0.14 µg/L	10x LOQ	4.54
		Confirmation	0.014 µg/L	1x LOQ	4.65
			0.14 µg/L	10x LOQ	5.01
	C_{12}-TMAC	Quantification	0.014 µg/L	1x LOQ	5.45
			0.14 µg/L	10x LOQ	4.09
		Confirmation	0.014 µg/L	1x LOQ	2.02
			0.14 µg/L	10x LOQ	5.20
	C_{14}-TMAC	Quantification	0.014 µg/L	1x LOQ	5.19
			0.14 µg/L	10x LOQ	3.98
		Confirmation	0.014 µg/L	1x LOQ	6.94
			0.14 µg/L	10x LOQ	5.65
	C_{16}-TMAC	Quantification	0.014 µg/L	1x LOQ	2.46
			0.14 µg/L	10x LOQ	4.23
		Confirmation	0.014 µg/L	1x LOQ	8.35
			0.14 µg/L	10x LOQ	6.52
	C_{18}-TMAC	Quantification	0.014 µg/L	1x LOQ	2.77
			0.14 µg/L	10x LOQ	2.96
		Confirmation	0.014 µg/L	1x LOQ	5.02
			0.14 µg/L	10x LOQ	5.77
	C_{18}-TMAC unsat.	Quantification	0.014 µg/L	1x LOQ	5.38
			0.14 µg/L	10x LOQ	4.79
		Confirmation	0.014 µg/L	1x LOQ	6.47
			0.14 µg/L	10x LOQ	4.08

Section A4.2a and 4.2c

Analytical Methods for Detection and Identification

Analytical methods including recovery rates and the limits of determination for the active substance, and for residues thereof, and where relevant in/on the following:

Ground water, surface water, tap water and soil

Matrix	Component	Method	Concentration	LOQ Level	Mean Recovery Rate [%]
Tap water	C_8-TMAC	Quantification	0.014 µg/L	1x LOQ	8.52
			0.14 µg/L	10x LOQ	2.57
		Confirmation	0.014 µg/L	1x LOQ	9.19
			0.14 µg/L	10x LOQ	4.33
	C_{10}-TMAC	Quantification	0.014 µg/L	1x LOQ	10.9
			0.14 µg/L	10x LOQ	3.46
		Confirmation	0.014 µg/L	1x LOQ	9.42
			0.14 µg/L	10x LOQ	2.50
	C_{12}-TMAC	Quantification	0.014 µg/L	1x LOQ	9.42
			0.14 µg/L	10x LOQ	3.60
		Confirmation	0.014 µg/L	1x LOQ	7.18
			0.14 µg/L	10x LOQ	3.19
	C_{14}-TMAC	Quantification	0.014 µg/L	1x LOQ	10.5
			0.14 µg/L	10x LOQ	2.66
		Confirmation	0.014 µg/L	1x LOQ	7.48
			0.14 µg/L	10x LOQ	2.86
	C_{16}-TMAC	Quantification	0.014 µg/L	1x LOQ	5.31
			0.14 µg/L	10x LOQ	2.43
		Confirmation	0.014 µg/L	1x LOQ	0.661
			0.14 µg/L	10x LOQ	3.19
	C_{18}-TMAC	Quantification	0.014 µg/L	1x LOQ	10.7
			0.14 µg/L	10x LOQ	1.94
		Confirmation	0.014 µg/L	1x LOQ	12.8
			0.14 µg/L	10x LOQ	2.65
	C_{18}-TMAC unsat.	Quantification	0.014 µg/L	1x LOQ	5.02
			0.14 µg/L	10x LOQ	15.6
		Confirmation	0.014 µg/L	1x LOQ	3.07
			0.14 µg/L	10x LOQ	5.85

Section A4.2a and 4.2c

Analytical Methods for Detection and Identification

Analytical methods including recovery rates and the limits of determination for the active substance, and for residues thereof, and where relevant in/on the following:

Ground water, surface water, tap water and soil

Matrix	Component	Method	Concentration	LOQ Level	Mean Recovery Rate [%]
Soil	C_8-TMAC	Quantification	0.00714 mg/kg	1x LOQ	3.58
			0.0714 mg/kg	10x LOQ	1.56
		Confirmation	0.00714 mg/kg	1x LOQ	7.14
			0.0714 mg/kg	10x LOQ	1.23
	C_{10}-TMAC	Quantification	0.00714 mg/kg	1x LOQ	4.45
			0.0714 mg/kg	10x LOQ	1.09
		Confirmation	0.00714 mg/kg	1x LOQ	7.50
			0.0714 mg/kg	10x LOQ	7.11
	C_{12}-TMAC	Quantification	0.00714 mg/kg	1x LOQ	3.09
			0.0714 mg/kg	10x LOQ	2.48
		Confirmation	0.00714 mg/kg	1x LOQ	5.38
			0.0714 mg/kg	10x LOQ	3.23
	C_{14}-TMAC	Quantification	0.00714 mg/kg	1x LOQ	1.27
			0.0714 mg/kg	10x LOQ	2.43
		Confirmation	0.00714 mg/kg	1x LOQ	4.34
			0.0714 mg/kg	10x LOQ	1.46
	C_{16}-TMAC	Quantification	0.00714 mg/kg	1x LOQ	4.34
			0.0714 mg/kg	10x LOQ	1.75
		Confirmation	0.00714 mg/kg	1x LOQ	4.16
			0.0714 mg/kg	10x LOQ	3.48
	C_{18}-TMAC	Quantification	0.00714 mg/kg	1x LOQ	2.41
			0.0714 mg/kg	10x LOQ	4.12
		Confirmation	0.00714 mg/kg	1x LOQ	7.13
			0.0714 mg/kg	10x LOQ	5.38
	C_{18}-TMAC unsat.	Quantification	0.00714 mg/kg	1x LOQ	3.57
			0.0714 mg/kg	10x LOQ	1.95
		Confirmation	0.00714 mg/kg	1x LOQ	7.88
			0.0714 mg/kg	10x LOQ	2.82

3.6 Limit of determination

The limit of detection (LOD) was confirmed via signal to noise ratio (S/N) >> 3 to be below 1 µg/L of each active ingredient (lowest standard concentration).

3.7 Precision

Section A4.2a and 4.2c	**Analytical Methods for Detection and Identification** **Analytical methods including recovery rates and the limits of determination for the active substance, and for residues thereof, and where relevant in/on the following:** Ground water, surface water, tap water and soil	

3.7.1 Repeatability

Repeatability of Injections of the lowest TMAC Standard (1 µg/L)
Peak area [counts]

	1 µg/L				
	Quantification				
Serial No.	C_8-TMAC	C_{10}-TMAC	C_{12}-TMAC	C_{14}-TMAC	C_{16}-TMAC
1	1390.224	1234.920	1324.344	738.323	908.759
2	1335.232	1256.569	1130.874	890.785	738.899
3	1394.064	1241.206	1150.760	735.395	842.552
4	1366.176	1147.080	1319.281	808.140	840.297
5	1373.500	1172.517	1254.559	909.348	885.822
6	1413.629	1127.185	1269.260	639.645	827.261
Mean ± SD	1379 ± 27.1	1197 ± 54.6	1242 ± 82.9	787 ± 103	841 ± 58.7
RSD [%]	1.97	4.56	6.67	13.1	6.98

	1 µg/L	
	Quantification	
Serial No.	C_{18}-TMAC unsaturated	C_{18}-TMAC
1	421.370	617.211
2	433.231	755.507
3	370.799	695.101
4	357.909	626.601
5	444.026	612.379
6	342.393	643.050
Mean ± SD	392 ± 47.9	658 ± 56.3
RSD [%]	12.2	8.56

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	1 µg/L				
	Confirmation				
Serial No.	**C_8-TMAC**	**C_{10}-TMAC**	**C_{12}-TMAC**	**C_{14}-TMAC**	**C_{16}-TMAC**
1	435.072	418.285	395.896	356.626	365.733
2	405.512	469.199	421.516	335.931	255.208
3	461.215	407.900	414.087	343.815	295.100
4	518.216	431.059	382.002	260.208	223.147
5	431.836	307.118	372.290	420.597	259.193
6	508.429	364.603	435.560	393.283	305.481
Mean ± SD	460 ± 45.0	400 ± 56.6	404 ± 24.3	352 ± 55.1	284 ± 49.8
RSD [%]	9.78	14.2	6.01	15.7	17.5

	1 µg/L	
	Confirmation	
Serial No.	**C_{18}-TMAC unsaturated**	**C_{18}-TMAC**
1	167.146	174.213
2	119.469	150.238
3	191.966	175.092
4	132.772	180.254
5	139.473	201.634
6	142.706	184.388
Mean ± SD	149 ± 26.2	178 ± 16.7
RSD [%]	17.6	9.38

Section A4.2a and 4.2c	**Analytical Methods for Detection and Identification** **Analytical methods including recovery rates and the limits of determination for the active substance, and for residues thereof, and where relevant in/on the following:** Ground water, surface water, tap water and soil	

Repeatability of Injections of the highest TMAC Standard (100 µg/L)
Peak area [counts]

	100 µg/L				
	Quantification				
Serial No.	**C_8-TMAC**	**C_{10}-TMAC**	**C_{12}-TMAC**	**C_{14}-TMAC**	**C_{16}-TMAC**
1	145909.563	125578.898	127553.391	92657.227	83938.797
2	142386.141	125539.898	127097.906	94435.836	86259.641
3	140235.344	125382.789	126603.641	92535.320	86439.828
4	141739.781	124399.094	124723.320	94793.977	87518.289
5	142185.797	125307.328	127174.711	89802.367	84735.445
6	138259.594	125070.797	123251.383	89187.852	88836.336
Mean ± SD	141786 ± 2544	125213 ± 438	126067 ± 1704	92235 ± 2318	86288 ± 1787
RSD [%]	1.79	0.350	1.35	2.51	2.07

	100 µg/L	
	Quantification	
Serial No.	**C_{18}-TMAC unsaturated**	**C_{18}-TMAC**
1	48903.551	76385.922
2	50940.059	78254.578
3	49300.738	76758.727
4	48992.922	74647.250
5	48559.230	72988.461
6	49943.129	75848.422
Mean ± SD	49440 ± 870	75814 ± 1818

Section A4.2a and 4.2c	**Analytical Methods for Detection and Identification** **Analytical methods including recovery rates and the limits of determination for the active substance, and for residues thereof, and where relevant in/on the following:** Ground water, surface water, tap water and soil	

RSD [%]	1.76	2.40

	100 µg/L				
	Confirmation				
Serial No.	**C_8-TMAC**	**C_{10}-TMAC**	**C_{12}-TMAC**	**C_{14}-TMAC**	**C_{16}-TMAC**
1	46449.961	42731.965	41690.816	30625.178	26811.406
2	44995.422	43049.191	41352.711	30451.139	26690.717
3	45453.438	41626.797	41233.141	30617.746	26687.213
4	46184.629	42506.770	40792.184	30820.051	27019.242
5	45540.762	42064.922	42327.242	29612.289	27243.635
6	44983.977	41271.570	40562.996	29217.748	27677.486
Mean ± SD	45601 ± 606	42209 ± 679	41327 ± 634	30224 ± 650	27022 ± 386
RSD [%]	1.33	1.61	1.53	2.15	1.43

	100 µg/L
	Confirmation

Section A4.2a and 4.2c	**Analytical Methods for Detection and Identification** **Analytical methods including recovery rates and the limits of determination for the active substance, and for residues thereof, and where relevant in/on the following:** Ground water, surface water, tap water and soil	

Serial No.	C_{18}-TMAC unsaturated	C_{18}-TMAC
1	18609.471	21225.273
2	19875.627	20805.477
3	18856.027	21332.555
4	19694.783	21343.043
5	19191.760	20563.359
6	19307.389	21099.494
Mean ± SD	19256 ± 482	21062 ± 315
RSD [%]	2.50	1.50

3.7.2 Independent laboratory validation		

Section A4.2a and 4.2c	**Analytical Methods for Detection and Identification** **Analytical methods including recovery rates and the limits of determination for the active substance, and for residues thereof, and where relevant in/on the following:** Ground water, surface water, tap water and soil	
	4 APPLICANT'S SUMMARY AND CONCLUSION	
4.1 Materials and methods	See Part 3	
4.2 Conclusion	For the external standard the analytical system gave a linear response for C8-, C10-, C12-, C14-, C16-, C18- and C18-unsaturated trimethylammonium chloride, between nominal 1 and 100 µg/L. **Ground Water** The results confirm that the described method is suitable for the determination of residues of TMAC (lyophilised Arquad C-35) in **ground water** at limit of quantification 0.1 µg/L. Due to the composition of the test item the C_8 -,C_{10}-, C_{16} -,C_{18}- and C_{18}-unsaturated TMAC components were below the limit of quantification of the analytical system. The other active ingredients gave acceptable recovery and precision for both, quantifier and confirmation trace. For results see 6.4.1. In order to characterise the limit of quantification, even for C_8 -,C_{10}-, C_{16} -,C_{18}- and C_{18}-unsaturated TMAC, an additional set of recovery determinations from **ground water** was performed at limit of quantification of 0.014 µg/L for each active ingredient, respectively. Acceptable recovery and precision for both, quantifier and confirmation trace were obtained, confirming the ability of the method to quantify the active ingredients down to the given limit. **Surface Water** The results confirm that the described method is suitable for the determination of residues of TMAC (lyophilised Arquad C-35) in **surface water** at limit of quantification 0.1 µg/L. Due to the composition of the test item the C_8 -,C_{10}-, C_{16} -,C_{18}- and C_{18}-unsaturated TMAC components were below the limit of quantification of the analytical system. The other active ingredients gave acceptable recovery and precision for both, quantifier and confirmation trace. For results see 6.4.1. In order to characterise the limit of quantification, even for C_8 -,C_{10}-, C_{16} -,C_{18}- and C_{18}-unsaturated TMAC, an additional set of recovery determinations from **surface water** was performed at limit of quantification of 0.014 µg/L for each active ingredient, respectively. Acceptable recovery and precision for both, quantifier and confirmation trace were obtained, confirming the ability of the method to quantify the active ingredients down to the given limit. **Tap Water** The results confirm that the described method is suitable for the determination of residues of TMAC (lyophilised Arquad C-35) in **tap**	

Section A4.2a and 4.2c	**Analytical Methods for Detection and Identification** **Analytical methods including recovery rates and the limits of determination for the active substance, and for residues thereof, and where relevant in/on the following:** Ground water, surface water, tap water and soil	
	water at limit of quantification 0.1 µg/L. Due to the composition of the test item the C_8 -,C_{10}-, C_{16} -,C_{18}- and C_{18}-unsaturated TMAC components were below the limit of quantification of the analytical system. The other active ingredients gave acceptable recovery and precision for both, quantifier and confirmation trace. For results see 6.4.1. In order to characterise the limit of quantification, even for C_8 -,C_{10}-, C_{16} -,C_{18}- and C_{18}-unsaturated TMAC, an additional set of recovery determinations from **tap water** was performed at limit of quantification of 0.014 µg/L for each active ingredient, respectively. Acceptable recovery and precision for both, quantifier and confirmation trace were obtained, confirming the ability of the method to quantify the active ingredients down to the given limit. **Soil** The results confirm that the described method is suitable for the determination of residues of TMAC (lyophilised Arquad C-35) in soil at limit of quantification 0.05 mg/kg. Due to the composition of the test item the C8 -,C10-, C16 -,C18- and C18-unsaturated TMAC components were below the limit of quantification of the analytical system. The other active ingredients gave acceptable recovery and precision for both, quantifier and confirmation trace. For results see 6.4.1. In order to characterise the limit of quantification, even for C8 -,C10-, C16 -,C18- and C18-unsaturated TMAC, an additional set of recovery determinations from soil was performed at limit of quantification of 0.00714 mg/kg for each active ingredient, respectively. Acceptable recovery and precision for both, quantifier and confirmation trace were obtained, confirming the ability of the method to quantify the active ingredients down to the given limit.	
4.2.1 Reliability	Based on the assessment of the method include appropriate reliability indicator 0, 1, 2, 3, 4	X
4.2.2 Deficiencies	No	X

	Evaluation by Competent Authorities
	EVALUATION BY RAPPORTEUR MEMBER STATE
Date	**May 2014**
Materials and methods	**4.2a Soil** An analytical method for the determination of C_{8-18}-TMAC residues in soil by LC-MS/MS is available. Analysis was performed in gradient mode on a C18 reversed-phase column, using external standards. Detection was carried out by an electrospray tandem mass spectrometer in positive mode. For each constituent of C_{8-18}-TMAC, specificity was achieved by formation of a specific precursor ion and two specific product ions (one mass transition used for quantification, the other for identity confirmation).

C_{8-18}-TMAC	Mass transitions
C_8-TMAC	172.24→60.17 (quantification) 172.24→57.18 (confirmation
C_{10}-TMAC	200.27→60.17 (quantification) 200.27→57.18 (confirmation)
C_{12}-TMAC	228.30→60.17 (quantification) 228.30→57.18 (confirmation)
C_{14}-TMAC	256.33→60.17 (quantification) 256.33→57.18 (confirmation)
C_{16}-TMAC	284.43→60.17 (quantification) 284.43→57.18 (confirmation)
C_{18}-TMAC	312.46→60.17 (quantification) 312.46→57.18 (confirmation)

The following information was available in the original study report as regards the investigated soil (certified *LUFA soil No. 2.2, batch Sp2.23311*):

Type	Loamy sand (IS, according to German DIN classification)
Origin	LUFA, Obere Langgasse 40 67346 Speyer "Großer Striet" No 585 D-67374 Hanghofer
pH	5.5
Carbon content [%]	1.87

The method was validated at 0.05 mg C_{8-18}-TMAC/kg and 0.50 mg C_{8-18}-TMAC /kg. For that purpose, lyophilized Arquad C-35 (C_{8-18}-TMAC technical material) was used for the spiking of the soil.

Additional validation was carried out at 0.00714 mg/kg and 0.0714 mg/kg for each individual constituent of C_{8-18}-TMAC (C8-TMAC, C10-TMAC C12-TMAC, C14-TMAC, C16-TMAC and C18-TMAC). Soil fortification was performed using the relevant standards.

Solutions of mixed standards at 7 different concentration levels were used for calibration.

Conclusion	The LC-MS/MS method submitted in May 2012 for the analysis of C_{8-18}-TMAC residues down to a level of 0.05 mg C_{8-18}-TMAC/kg in soil meets the requirements provided for by SANCO/825/00 and the Additional Guidance to TNsG on Data Requirements on analytical methods. The method supports the residue definition. The method is highly specific (LC-MS/MS, with two mass transitions validated), linear over the range 0.008-0.75 mg C_{8-18}-TMAC/kg in soil, accurate (with recovery rates at LOQ and 10xLOQ in the acceptable range 70–120%) and precise (%$RSD_{n=5}$ < 20% for either fortification level). The LOQ (as the lowest validated fortification level) complies with the relevant end-point (< EC_{50} = 11 mg/kg dw soil; read-across from DDAC data). Satisfactory results in terms of recovery rates and %$RSD_{n=5}$ were also obtained for individual constituents of C_{8-18}-TMAC (C8-TMAC, C10-TMAC C12-TMAC, C14-TMAC, C16-TMAC and C18-TMAC) at 0.00714 mg/kg and 0.0714 mg/kg each.
Reliability	2

<table>
<tr><td>Acceptability</td><td>Acceptable</td></tr>
<tr><td>Remarks</td><td>X – Minor deviations from SANCO/825/00 rev. 7 have occurred (see general remark below). Also note that rev. 8.1 (16/11/2010) is now available for the Guideline.

X1 – 7 concentration levels (with single determination) were considered (calibration in solvents). The linearity range of 1–100 µg/L for each constituent of C_{8-18}-TMAC corresponds to 0.008–0.75 mg C_{8-18}-TMAC/kg in soil (inferred by the RMS according to the information on sample preparation available in the study report and taking into account that the % of C12-TMAC in the C_{8-18}-TMAC technical material used for spiking is 53.5%).

Linearity curves:
<table>
<tr><th>C_{8-18}-TMAC</th><th>Mass transition</th><th>Curve equation</th><th>r^2</th></tr>
<tr><td rowspan="2">C_8-TMAC</td><td>172.24→60.17 (quantification)</td><td>y = 1939.74x</td><td>0.999919</td></tr>
<tr><td>172.24→57.18 (confirmation</td><td>y = 647.446x</td><td>0.999884</td></tr>
<tr><td rowspan="2">C_{10}-TMAC</td><td>200.27→60.17 (quantification)</td><td>y = 1953.15x</td><td>0.999961</td></tr>
<tr><td>200.27→57.18 (confirmation)</td><td>y = 690.483x</td><td>0.999882</td></tr>
<tr><td rowspan="2">C_{12}-TMAC</td><td>228.30→60.17 (quantification)</td><td>y = 1835.66x</td><td>0.999945</td></tr>
<tr><td>228.30→57.18 (confirmation)</td><td>y = 629.737x</td><td>0.999758</td></tr>
<tr><td rowspan="2">C_{14}-TMAC</td><td>256.33→60.17 (quantification)</td><td>y = 1597.1x</td><td>0.999744</td></tr>
<tr><td>256.33→57.18 (confirmation)</td><td>y = 509.379x</td><td>0.999456</td></tr>
<tr><td rowspan="2">C_{16}-TMAC</td><td>284.43→60.17 (quantification)</td><td>y = 1543.68x</td><td>0.999779</td></tr>
<tr><td>284.43→57.18 (confirmation)</td><td>y = 477.949x</td><td>0.999838</td></tr>
<tr><td rowspan="2">C_{18}-TMAC</td><td>312.46→60.17 (quantification)</td><td>y = 1296.88x</td><td>0.999556</td></tr>
<tr><td>312.46→57.18 (confirmation)</td><td>y = 370.575x</td><td>0.999068</td></tr>
</table>
X2 – Since two mass transitions have been investigated, the submitted LC-MS/MS can be concluded to be highly specific.

X* – Spiking solutions I to IV of the test item (lyophilized Arquad C-35) are erroneously reported (also in the original study report) to be “mixed standard spiking solutions diluted by a total factor of ... from the stock solution” instead of “dilution by total factor of ... from the stock solution of TMAC”.

X** – To investigate matrix effects, the response of C_{8-18}-TMAC in matrix as well as the response of individual standards in matrix were also considered. According to the original study report, solutions were prepared by fortification of a control sample with lyophilized Arquad C-35 or with the relevant standards, respectively. Satisfactory recovery rates were obtained.

General remarks:

– No spectrum of the product ions has been provided in the original study report. The selection of the ions used for quantification/confirmation has not been justified, either.

– eCA-IT considers C18-unsaturated as not part of the residue definition. Though available in the original study report, validation data relevant to C18-unsaturated have not been taken into account by RMS-IT (ISSUE TO BE DISCUSSED AT WG LEVEL).</td></tr>
<tr><td></td><td>COMMENTS FROM ...</td></tr>
<tr><td>Date</td><td>Give date of comments submitted</td></tr>
<tr><td>Results and discussion</td><td>Discuss additional relevant discrepancies referring to the (sub)heading numbers and to applicant´s summary and conclusion.
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<tr><td>Reliability</td><td>Discuss if deviating from view of rapporteur member state</td></tr>
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</table>

Remarks	

	Evaluation by Competent Authorities
	EVALUATION BY RAPPORTEUR MEMBER STATE
Date	**May 2014**
Materials and methods	**4.2c Water** An analytical method for the determination of C_{8-18}-TMAC residues in ground-, surface- and drinking-water by LC-MS/MS is available. Analysis was performed in isocratic mode on a C18 reversed-phase column, using external standards. Detection was carried out by an electrospray tandem mass spectrometer in positive mode. For each constituent of C_{8-18}-TMAC, specificity was achieved by formation of a specific precursor ion and two specific product ions (one mass transition used for quantification, the other for identity confirmation).

C_{8-18}-TMAC	Mass transitions
C_8-TMAC	172.24→60.17 (quantification) 172.24→57.18 (confirmation
C_{10}-TMAC	200.27→60.17 (quantification) 200.27→57.18 (confirmation)
C_{12}-TMAC	228.30→60.17 (quantification) 228.30→57.18 (confirmation)
C_{14}-TMAC	256.33→60.17 (quantification) 256.33→57.18 (confirmation)
C_{16}-TMAC	284.43→60.17 (quantification) 284.43→57.18 (confirmation)
C_{18}-TMAC	312.46→60.17 (quantification) 312.46→57.18 (confirmation)

The following information was available in the original study report as regards the investigated water matrices:

Type	Ground water	Surface water	Tap water
Origin	Glück auf Straße 32 D-31157 Sarstedt	31180 Ahrbergen Geographical coordinates: 52°13'38.00'' N and 9°5'52.22'' E	At the test facility
pH	7.39	8.79	8.54-8.65
Conductivity [µS/cm]	900	473	168-171
Dissolved oxygen [%]	9.28	8.89	926
DOC [mg/L]	1.55	>100	--
Ammonium-N [mg/L]	0.04	<0.04	--
Nitrate-N [mg/L]	11.8	<0.3	--

The method was validated at 0.1 µg C_{8-18}-TMAC/L and 1.0 µg C_{8-18}-TMAC/L. For that purpose, lyophilized Arquad C-35 (C_{8-18}-TMAC technical material) was used for the spiking of the matrices (ground-, surface- and drinking-water).

Additional validation was carried out at 0.014 µg/L and 0.140 µg/L for each constituent of C_{8-18}-TMAC (C8-TMAC, C10-TMAC C12-TMAC, C14-TMAC, C16-TMAC and C18-TMAC). Water fortification was performed using the relevant standards.

Solutions of mixed standards at 7 different concentration levels were used for calibration.

Conclusion	The LC-MS/MS method submitted in May 2012 for the analysis of C_{8-18}-TMAC residues down to a level of 0.1 µg C_{8-18}-TMAC/L in ground-, surface- and drinking-water meets the requirements provided for by SANCO/825/00 and the Additional Guidance to TNsG on Data Requirements on analytical methods. The method supports the residue definition. The method is highly specific (LC-MS/MS, with two ion transitions validated), linear over the range 0.01–1.04 µg C_{8-18}-TMAC/L in matrix, accurate (with recovery rates at LOQ and 10xLOQ in the acceptable range 70–120%) and precise (%$RSD_{n = 5}$ < 20% for either fortification level). Ground- and drinking-water: the LOQ (as the lowest validated fortification level) complies with the EU water limit of 0.1 µg/L. Surface water: the LOQ (as the lowest validated fortification level) complies with the relevant endpoint (<NOEC = 2.5 µg/L; read-across from C_{12-16}-BKC data). Satisfactory results in terms of recovery rates and %$RSD_{n = 5}$ were also obtained for individual constituents of C_{8-18}-TMAC (C8-TMAC, C10-TMAC C12-TMAC, C14-TMAC, C16-TMAC and C18-TMAC) at 0.014 µg/L and 0.140 µg/L each.
Reliability	2
Acceptability	Acceptable

<table>
<tr><td>Remarks</td><td>X – Minor deviations from SANCO/825/00 rev. 7 have occurred (see general remark below). Also note that rev. 8.1 (16/11/2010) is now available for the Guideline.

X1 – 7 concentration levels (with single determination) were considered (calibration in solvents). The linearity range of 1 – 100 µg/L for each constituent of C_{8-18}-TMAC corresponds to 0.010–1.04 µg C_{8-18}-TMAC/L in matrix (inferred by the RMS according to the information on sample preparation available in the study report and taking into account that the % of C12-TMAC in C_{8-18}-TMAC is 53.5%).

Linearity curves:
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y = 629.737x</td><td>0.999945
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<tr><td rowspan="2">C_{14}-TMAC</td><td>256.33→60.17 (quantification)
256.33→57.18 (confirmation)</td><td>y = 1597.1x
y = 509.379x</td><td>0.999744
0.999456</td></tr>
<tr><td rowspan="2">C_{16}-TMAC</td><td>284.43→60.17 (quantification)
284.43→57.18 (confirmation)</td><td>y = 1543.68x
y = 477.949x</td><td>0.999779
0.999838</td></tr>
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312.46→57.18 (confirmation)</td><td>y = 1296.88x
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X** – To investigate matrix effects, the response of C_{8-18}-TMAC in matrix as well as the response of individual standards in matrix were also considered. According to the original study report, solutions were prepared by fortification of a control sample with lyophilized Arquad C-35 or with the relevant standards, respectively. Satisfactory recovery rates were obtained.

General remarks:

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</table>