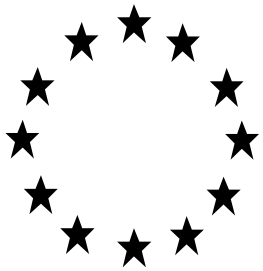
Regulation (EU) No 528/2012 concerning the making available on the market and use of biocidal products

Evaluation of active substances

Assessment Report



Amines, N-C10–C16-alkyltrimethylenedi-, reaction products with chloroacetic acid

Product-type 3

April 2015

Ireland

CONTENTS

1. STATEMENT OF SUBJECT MATTER AND PURPOSE

1.1 PROCEDURE FOLLOWED

This assessment report has been established as a result of the evaluation of Amines, N-C_{10}–C_{16}-alkyltrimethylenedi-, reaction products with chloroacetic acid (which is also known under the synonym Ampholyt) as a product-type 3 (private area and public health area disinfectants), carried out in the context of the work programme for the review of existing active substances provided for in Article 89 of Regulation (EU) No 528/2012, with a view to the possible approval of this substance.

Amines, N-C_{10}–C_{16}-alkyltrimethylenedi-, reaction products with chloroacetic acid or Ampholyt (CAS no. 139734-65-9) was notified as an existing active substance, by Evonik Industries AG (formerly Goldschmidt GmbH), hereafter referred to as the applicant, in product-type 3.

Commission Regulation (EC) No. 1451/2007 of 4 December 2007[1] lays down the detailed rules for the evaluation of dossiers and for the decision-making process.

In accordance with the provisions of Article 7(1) of that Regulation, Ireland was designated as Rapporteur Member State to carry out the assessment on the basis of the dossier submitted by the applicant. The deadline for submission of a complete dossier for Ampholyt as an active substance in product-type 3 was 31st July 2007, in accordance with Annex V of Regulation (EC) No. 2032/2003.

On 30th July 2007, the Irish competent authorities received a dossier from the applicant. The Rapporteur Member State could not at that juncture determine the dossier as complete. The applicant accepted the decision and committed to fulfilling the outstanding requirements as identified by the Rapporteur Member State. The outstanding data were submitted to the Rapporteur Member State on 20th February 2009 and 26th April 2012. The Rapporteur Member State accepted the dossier as complete for the purpose of the evaluation on 14th August 2012.

On 30 August 2013, the Rapporteur Member State submitted to the Commission and the applicant a copy of the evaluation report, hereafter referred to as the competent authority report.

In accordance with Article 16 of Regulation (EC) No 1451/2007, the Commission made the competent authority report publicly available by electronic means. This report did not include such information that was to be treated as confidential in accordance with the provisions of Regulation (EU) No 528/2012.

In order to review the competent authority report and the comments received on it, consultations of technical experts from all Member States (peer review) were organised by the Commission. Revisions agreed upon were presented at technical and competent authority meetings and the competent authority report was amended accordingly.

[1] Commission Regulation (EC) No 1451/2007 of 4 December 2007 on the second phase of the 10-year work programme referred to in Article 16(2) of Directive 98/8/EC of the European Parliament and of the Council concerning the placing of biocidal products on the market. OJ L 325, 11.12.2007, p. 3

1.2 PURPOSE OF THE ASSESSMENT

The aim of the assessment report is to support a decision on the approval of Ampholyt for product-type 3 and should it be approved, to facilitate the authorisation of individual biocidal products in product-type 3 that contain Ampholyt. In the evaluation of applications for product authorisation, the provisions of Regulation (EU) No 528/2012 shall be applied, in particular the provisions of Chapter IV, as well as the common principles laid down in Annex VI.

The conclusions of this report were reached within the framework of the uses that were proposed and supported by the applicant (see Appendix II). Extension of the use pattern beyond those described will require an evaluation at product authorisation level in order to establish whether the proposed extensions of use will satisfy the requirements of Regulation (EU) No 528/2012.

For the implementation of the common principles of Annex VI, the content and conclusions of this assessment report, which is available from the Agency web-site, shall be taken into account.

However, where conclusions of this assessment report are based on data protected under the provisions of Regulation (EU) No 528/2012, such conclusions may not be used to the benefit of another applicant, unless access to these data has been granted.

2. OVERALL SUMMARY AND CONCLUSIONS

2.1 PRESENTATION OF THE ACTIVE SUBSTANCE

2.1.1 Identity, Physico-Chemical Properties and Methods of Analysis

CAS No.: 139734-65-9

EC No.: None available.

Other No. (CIPAC, ELINCS): None available.

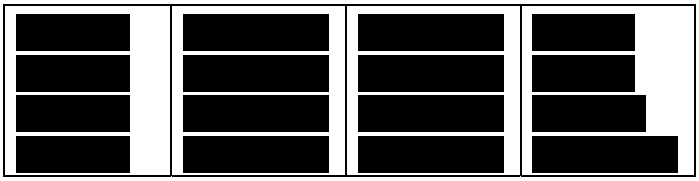
IUPAC Name: Amines, N-C_{10}–C_{16}-alkyltrimethylenedi-, reaction products with chloroacetic acid

CA Name: Amines, N-C_{10}–C_{16}-alkyltrimethylenedi-, reaction products with chloroacetic acid

Common name, synonym: Amines, N-C_{10}–C_{16}-alkyltrimethylenedi-, reaction products with chloroacetic acid; Ampholyt

Molecular formula: No single representative molecular formula available. It is impossible to assign a single molecular formula since Ampholyt is a mixture of various chemicals. Ampholyt contains 24 individual components which are considered to be part of the active substance.

The relevant molecular formulas for the 24 chemical species are presented in the table below:



[redacted]

Purity:
TC - dry material – hypothetical:
Minimum specification of 100% w/w for total active ingredient content.

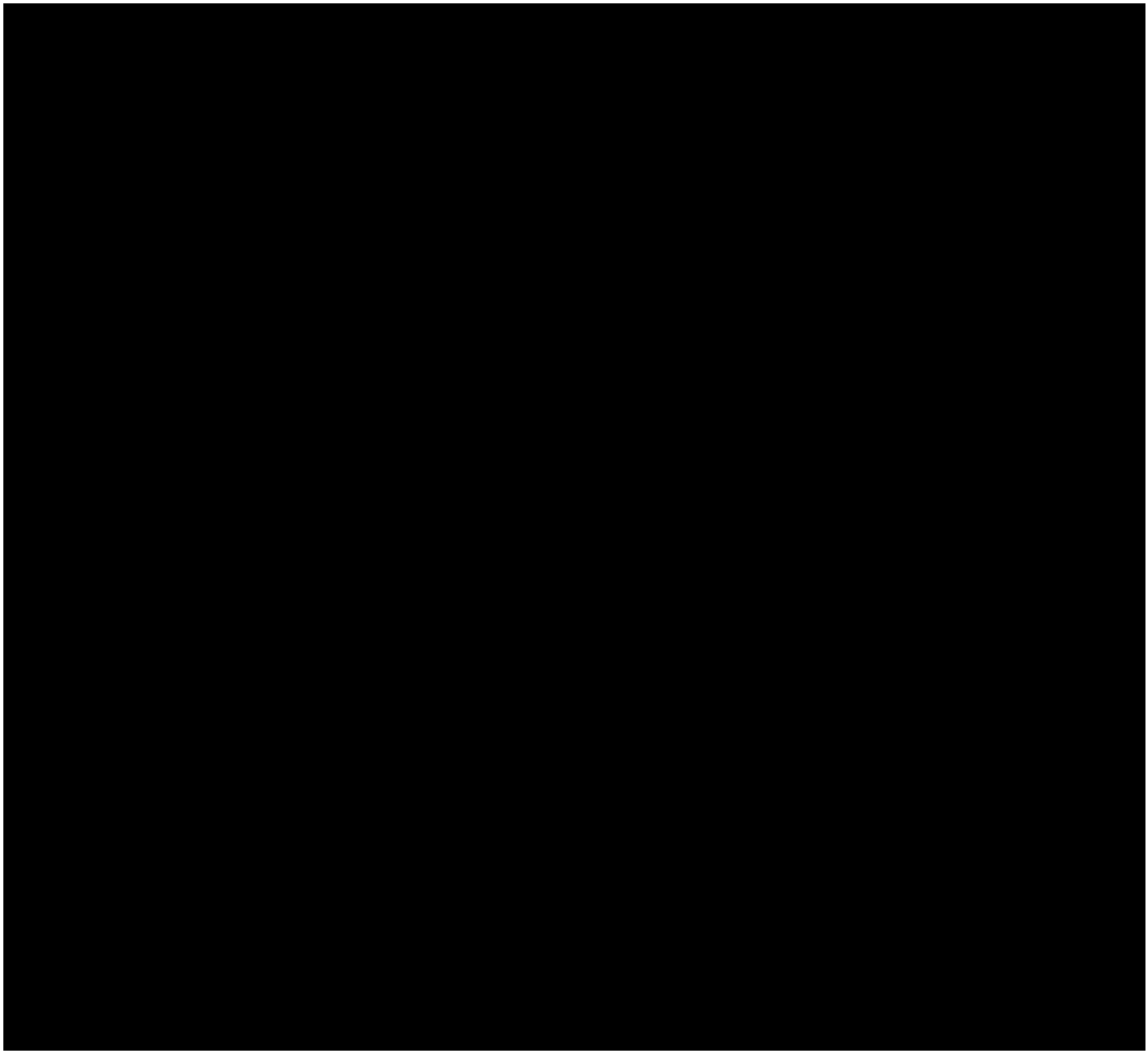
TK – aqueous solution – technical material as manufactured:
Specification range of 16 – 22% w/w (average of 19 % w/w) for total active ingredient content.

The individual specification ranges for the components making up the active ingredient (UVCB substance) in the TC and TK are provided in the Confidential Section of the CAR.

APCP WG III 2014 decided that only the overall min. purity of 100% w/w (TC) will appear in the approval. The exact identity and concentration ranges of the individual components in Ampholyt will not be disclosed in the approval or the non-confidential sections of the CAR.

Structural formula: No single representative structural formula available.

It is impossible to assign a single structural formula since Ampholyt is a mixture of various chemicals, however the structural formulae of the components making up the active substance are provided below:



Note 2: The above structural formulas cover the species making up the UVCB substance. All of these species are in a thermodynamic equilibrium with their protonated and unprotonated forms. The equilibrium is pH dependent. The pH of the solution in water is approximately around 8 – 8.5 in the presence of acetic acid and hydrochloride which is being released from chloroacetic acid after the reaction with the primary and secondary amines.

Molecular weight (g/mol): No single representative molecular weight available.

It is impossible to assign a single molecular weight since Ampholyt is a mixture of various chemicals.
The molecular masses for these 24 chemicals range from 185.4 – 414.63 g/mol.

A "weighted molecular mass" of 280.79 g/mol can also be proposed taking into account the molecular weight of the 24 components and their average composition in the supporting 7-batch analysis.

Ampholyt is a surfactant mixture which is considered to be a UVCB substance (substance of Unknown, Variable Composition, or Biological origin). A number of physical and chemical properties could not be experimentally determined because of the surfactant properties of the mixture - physical and chemical properties for the individual components of the UVCB substance have been determined by QSAR in these specific cases.

The active ingredient is considered to be made up of *ca.* 24 individual components having long chain alkanes (C_{10}-C_{16} with C_{12} and C_{14} predominating) with amine, or amine and carboxyl functional groups. Four of the twenty four components are considered to be "minor constituents". The sum of the "minor constituents" accounts for < 2% of the active ingredient.

Technical Ampholyt is a colourless to yellowish liquid. The purified active substance is a white, crystalline solid. The purified active substance is highly soluble in water (>208 g/L at 20°C– equating to miscible in all proportions) and readily soluble in other polar solvents such as methanol (40 – 50 at 20°C). The purified active is considered to be relatively insoluble in less polar solvents (<10 g/L in p-Xylene , n-Heptane, 1,2-Dichloroethane, Acetone and Ethyl acetate at 20°C.
The active substance decomposes at *ca.* 140°C.

LogPow values for the 20 "major components" range from 0.33 to 6.71. The weighted LogPow value = 3.82 (EpiSuite-KOWWIN).

Log P values for the 24 chemical species making up the active substance ranged from 2.83 – 5.73 (QSAR Calculation using Marvin Sketch – ChemAxon, 2012). However, the Log P QSAR calculations do not take into account that the 24 chemical species are ionisable species at environmental pH. Log P QSAR calculations are only appropriate for neutral molecules.
Log D is a more applicable endpoint for risk assessment. Log D is PH dependent and is applicable to protonated and unprotonated species. The LogD ranges for the 24 chemical species ranged from -4.53 to 2.11 at pH5, -2.37 to 2.33 for pH7 and -3.12 to 3.92 at pH9.

The determination of the dissociation constant is experimentally not feasible. However, the components making up Ampholyt are expected to be ionised in the environmental pH range (4 – 9) based on the presence of acidic (pKa ~ 4) and basic (pKa ~ 10) functional groups in the respective structures.

The surface tension of the purified active was 27.2 mN/m at 20ºC which confirms the surfactant properties of the molecule. The active ingredient is expected to be non-volatile based on a vapour pressure of 1.9 x 10-4 Pa at 20°C. The individual Henry's Law Constants for the "major components" range from 1.29 x 10-9 – 1.22 x 10-2 Pa.m3/mol. The individual Henry's Law Constants for the "minor components" range from 8.94 x 10-10 – 3.05 x 101 Pa.m3/mol. The "minor components" make up <2% of the active ingredient (QSAR Calculations – EPI Suite – HENRYWIN).
The technical material as manufactured will not classify as being flammable, explosive or oxidising.

2.1.1.1. Analysis of the active substance as manufactured

The applicant provided a HPLC-CAD method of analysis for the determination of the active ingredient in the technical material as manufactured. HPLC-MS was used for confirmatory purposes.
Two key deficiencies remain regarding method validation of the method.
The deficiencies relate to:

- the use/synthesis of certain reference standards for method validation and
- the lack of validation data for a number of components which are considered to be part of the active substance.

Ampholyt is a UVCB substance and therefore does not contain impurities as such. The HPLC-CAD method is being relied upon to determine all components of the active ingredient in Ampholyt.

The applicant needs to provide a validated method of analysis for water in Ampholyt.

The applicant has provided a HPLC-UV method (205nm) for the analysis of acetic acid in technical Ampholyt. The applicant should experimentally determine the LOQ down to a level of 0.9% w/w. The other validation parameters (linearity, recovery, precision etc) are considered to be acceptable for the HPLC-UV method.
Further details regarding the HPLC-CAD and HPLC-UV methods can be found in the Confidential Section of the CAR - Section 4.1 of the CAR.

2.1.1.2. Formulation analysis

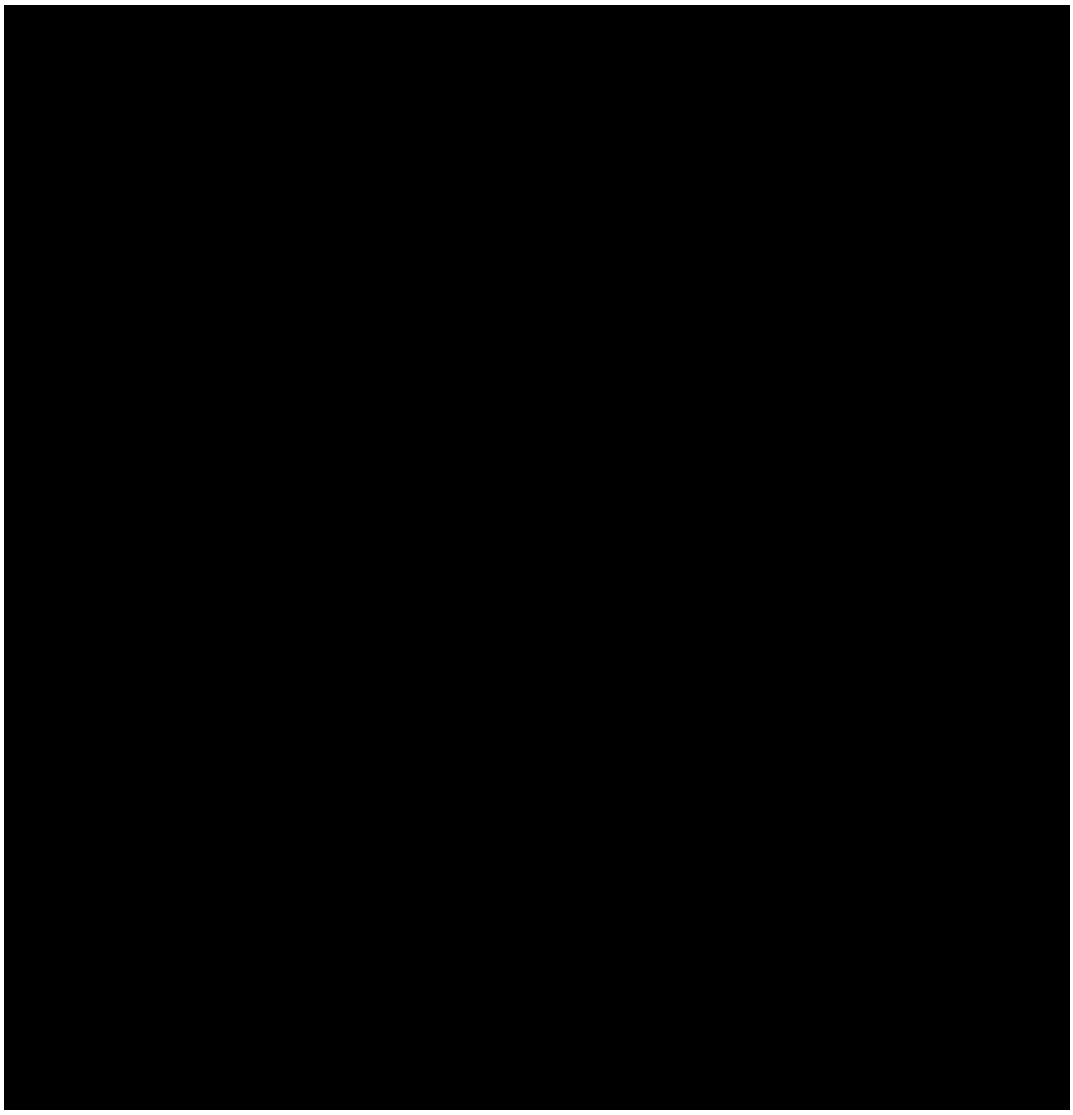
The technical material as manufactured and the formulated product are the same.
See Section 2.1.1.1 for HPLC-CAD method overview.

2.1.1.3. Residue analysis

The applicant has provided LC-MS/MS methods of analysis for Ampholyt in soil, drinking water, food of plant and animal origin (meat, milk, fat, wine and beer), however deficiencies remain outstanding regard method validation. The outstanding information required should be provided to the RMS at least 6 months before the date of entry into force.

<u>Residue definitions for monitoring –</u>
The residue definition for monitoring in all matrices includes "three lead components" [redacted] found in technical Ampholyt:

[redacted]



The RMS notes that we do not know the % of the three lead components or their metabolites in food of plant and animal origin after Ampholyt application because the applicant has not provided metabolism studies for food of plant and animal origin. The applicant needs to elaborate further regarding their choice of lead components for monitoring in food of plant and animal origin.

Soil:
The LC-MS/MS method has been validated for the three lead components.
The LOQ = 0.05 mg/kg for each lead component (critical NO(A)EC in soil = 363 mg/kg soil).

The applicant has only used a single ion transition for method validation. The applicant should validate the lead components for an additional ion transition.
Further details regarding the LC-MS/MS method can be found in the Section A4.2 of the CAR.

Water:
The LC-MS/MS method has been validated for the three lead components in drinking water matrices.
The LOQ = 0.1 µg/L for each lead component in drinking water.

The applicant has only used a single ion transition for method validation. The applicant should validate the lead components for an additional ion transition.

The applicant needs to validate the method in surface water and sediment matrices.
Further details regarding the LC-MS/MS method can be found in the Section A4.2 of the CAR.

Air:
A method of analysis is not required for Ampholyt in air due to insignificant inhalation exposure, however if the type of application changes (foam application) in the future, the request for a method for air may have to be revisited.

Food of plant and animal origin:
The LC-MS/MS method has been validated for the four lead components.
The LOQ = 0.01 mg/kg for each lead component in each matrices (meat, milk, fat, wine and beer).

The applicant has only used a single ion transition for method validation. The applicant should validate the lead components for an additional ion transition.

The applicant will also need to provide an ILV study for the method if MRLs for biocides are required. The request for an ILV will be reconsidered at product authorisation when additional guidance on how to derive MRLs for biocides will be possibly in place.

Further details regarding the LC-MS/MS method can be found in the Section A4.3/04 of the CAR.

Body fluids and tissues:

Reg. 1272/2008
The active substance is classified as Danger (H372).

The applicant will need to provide a validated method of analysis for monitoring in body fluids and tissues with an LOQ which allows determination at the NOAEL.

2.1.2 Intended Uses and Efficacy

The assessment of the biocidal activity of the active substance demonstrates that it has a sufficient level of efficacy against the target organism(s) and the evaluation of the summary data provided in support of the efficacy of the accompanying product, establishes that the product may be expected to be efficacious.

In addition, in order to facilitate the work of Member States in granting or reviewing authorisations, and to apply adequately the provisions of Article 5(1) of Directive 98/8/EC and the common principles laid down in Annex VI of that Directive, the intended uses of the substance, as identified during the evaluation process, are listed in Appendix II.

2.1.2.1 Field of use envisaged / Function and organism(s) to be controlled

Function

The general function of the active substance is for use as a disinfectant.

The more specific function of the active substance is for use as a:

- bactericide,
- yeasticide and,
- a limited virucide (effective against enveloped viruses) and active against Adenovirus

Field of Use

The active substance field of use as described in Annex V is as follows:

- Main Group 1 (MG01): Disinfectants
  - Product-type 3 (PT3): Veterinary hygiene

The use of the product is for the protection of principally both humans and animals. Application of the product to achieve the protection claims are treatments to surfaces, walls, and floors in various areas in industry (i.e. the food/feed industry) as well as in public health or veterinary areas. The mode of application for the PT3 area includes application by low pressure spraying, use in footbaths and also a ready-to-use trigger spray for localised spot treatments. All applications assessed are intended for professionals only. In use concentrations are 0.1–0.2 % active matter as aqueous solution, i.e. 0.5–1 % TEGO 2000 (1:200–1:100) or RTU (0.2% a.s.).

Organisms to be Controlled

Ampholyt is used as a disinfectant, antimicrobial cleaner and detergent sanitizer to prevent the spread of various micro-organisms. The spectrum of antimicrobial activity is focused on the destruction of gram-positive and gram-negative bacteria, yeasts, as well as a limited viruicide activity against enveloped viruses and against the non-enveloped adenovirus.

Specifically, the target organisms where data for Ampholyt has shown effectiveness are:

Bacteria: effective against *Staphylococcus aureus*, methycillin resistant *S. aureus* (MRSA), *Pseudomonas aeruginosa*, *Enterococcus hirae*, *Esherichia coli*, *Streptococcus faecium*, *Proteus mirabilis* at various concentrations (v/v) Ampholyt.

Phase 1 tests according to EN 1040 were completed on *S. aureus and P. aeruginosa*; a minimum of 0.125% (v/v) Ampholyt was required to achieve a reduction in bacterial

viability of more than 10^5 after 5 mins in both species. Phase 2/step 1 tests according to EN Standards were completed on *S. aureus*, *P. aeruginosa*, *E. hirae*, *E. coli and* methycillin resistant *S. aureus* (MRSA) ; a minimum of 0.25% (v/v) Ampholyt was required to achieve a reduction in bacterial viability of more than 10^5 after 5 mins under clean conditions, concentrations of 0.5% or higher (v/v) of Ampholyt was required under dirty conditions. Phase 2/step 2 tests according to EN Standard 13697 were completed on *S. aureus*, *P. aeruginosa*, *E. hirae*, *E. coli and* methycillin resistant *S. aureus* (MRSA). In the phase 2/step 2 test (EN 13697) tests bactericidal efficacy could not be demonstrated under dirty conditions.

Fungi: None of the experiments on fungicidal activity claims were fully satisfactory (B5.10.2/04 failed against *A niger*, B5.10.2/10-B5.10.2/14 did not test all obligatory fungal strains). However, Ampholyt demonstrated effectiveness at 0.125% v/v and at 0.25% v/v against the yeast *Candida albicans*.

Viruses: From the results presented on *Vaccinia virus*, Bovine viral diarrohea, *Herpes simplex*, *Bovine coronavirus*, avian influenza, human rota virus, Poxvirsus, Orthomyxovirus, Adenovirus, Enterovirus (Poliovirus), Rhabdovirus and hepatitis B. The strains tested represent both DNA and RNA viruses, and enveloped and non-enveloped viruses. Ampholyt had limited virucidal activity against enveloped viruses and against the non-enveloped adenovirus but was effective at concentrations between 0.2% and 1.0% on the viruses tested, with the exception of the tests on Poliovirus type 1 strain (a non-enveloped RNA virus). In these tests Ampholyt appeared to be ineffective against the virus.

Overall, the effectiveness of Ampholyt observed in tests under a range of conditions on bacteria, moulds and viruses to demonstrate innate activity of the active substance against a selection of representative target organisms indicated effective concentrations in the ranges 0.125-0.5%, 0.125-0.25% and 0.2-1.0%, respectively.

2.1.2.2 Effects on target organism(s)

The microbicidal action of Ampholyt has been attributed to the induction of changes of the cell wall structure, leading to a disruption of the bacterial cell, or the viral envelope, respectively. Since microbicidal amphoterics are cationic, they are readily attracted and adsorbed on surfaces possessing a negative charge, such as proteins or the microbial cell wall.

The adsorption to the microbial cell components is followed by a perforation by tubular protuberances of the outer lamellas; it is assumed that the amphoteric molecules with their amino acid like structures integrate into the cell wall and cytoplasma membrane, thus penetrate the cell wall and affecting cell permeability. The cytoplasm is coagulated and reaction occurs with the carboxylic acid group of enzymes. This leads to crystallisation like processes inside the cell and cytoplasma membrane, and results in disorganisation of metabolic pathways and structural disaggregation of the cells.

Experimental data on the effectiveness of Ampholyt against target organisms are provided in Document IIA.

2.1.2.3 Humaneness

Not applicable

2.1.2.4 Resistance

Specific resistance to Ampholyt has not been recorded to date.

Resistance to amphoteric disinfectants is not expected due to the relatively unspecific mode of action of amphoterics, which is at least partly based on surface activity. Amphoteric surfactants integrate into the cell wall of bacteria (or the envelope of viruses) and thereby cause leakage of intracellular components or impair the viral function. Furthermore, the intended uses, including performance of mechanical cleaning procedures, limit the number of targets and thereby additionally reduce the likelihood of development of resistance.

Bacteria or other micro-organisms may generally have an intrinsic or natural capacity of developing resistance to an antimicrobial agent. Resistance may in principle also be acquired by adaptation. Resistance may be mediated by resistance genes, which insert in specific sequences or by acquisition of plasmids or transposons, encoding a mechanism to disable a specific antimicrobial action. Microbial resistance to antimicrobial agents – or more generally biocides – is favoured by frequent use of sublethal concentrations and misuse of the agents which imposes a selective pressure.

However, since the mode of action of amphoteric surfactants is relatively unspecific, including (as the name implies) surface activity, selection for specific resistance genes is hardly conceivable. Instead, because of the multiply charged character of the molecule, amphotheric agents effectively bind to cellular or viral surfaces, and disrupt the barrier that ensures impermeability. The interaction with membrane components may further disorganise signalling. These effects lead to the very effective decrease in cell viability and viral infectivity. In conclusion, it is therefore considered unlikely that microorganisms or viruses develop resistance against Ampholyt. Recent literature searches have not revealed any information indicating that resistance to Ampholyt may have occurred.

2.1.3 Classification and Labelling

2.1.3.1 Proposal for the classification and labelling of the active substance

Proposal for the classification and labeling of the active substance according to CLP Regulation (EC) No 1272/2008

Pictogram: (for labelling)		
Signal word:	DANGER	
Hazard Statements: (for labelling)	H302 H314 H361f H372 H410 EUH401	Harmful if swallowed Causes severe skin burns and serious eye damage Suspected of damaging fertility (Category 2) Causes damage to (eyes, mesenteric lymph nodes, male/female genital systems) through prolonged or repeated exposure (STOT RE Cat 1) Very toxic to aquatic life with long lasting effects To avoid risks to human health and the environment, comply with the instructions for use
Precautionary Statements: (for labelling)	P273 P391 P501	Avoid release to the environment Collect Spillage Dispose of contents/ container in accordance with applicable regulations
M-Factors	M = 10 (acute) M = 1 (chronic)	

Justification for the proposal:

Physical-Chemical Properties:
The substance is not explosive, flammable or oxidising. The substance is not a compressed gas. The applicant should provide a study or scientific statement in relation to corrosivity to metals

Human Health:

Acute Toxicity
The lyophilized formulation of Ampholyt (100% technical material) was determined (Prinsen, 2003) to have a Rat Oral LD_{50} value between 300 and 2,000 mg/kg body weight. According to the requirements specified by Regulation (EC) 1272/2008 and subsequent regulations, Ampholyt requires classification for acute oral toxicity with classification Category 4, signal word "warning" and the hazard phrase Warning H302: Harmful if swallowed" in accordance with the classification criteria of Regulation 1272/2008. It is proposed that the active substance obtains a health hazard classification based on *in vivo* corrosivity and irritation studies on rabbit. Results indicate that the active substance should be classified as Danger H314: Causes severe skin burns and serious eye damage.

Sub-chronic and Reproductive Toxicity
In addition, repeated dose studies on the 90-day rat study, the 90-day dog study and the two year mouse study indicate that a proposal of H372: STOT RE Cat 1 Causes damage to organs (eyes, mesenteric lymph nodes, male/female genital systems) through prolonged or repeated exposure is warranted. Furthermore, reproductive toxicity studies

on the 90-day dog and 18-month mouse study indicate that a proposal of H361f Cat 2: Suspected of damaging fertility is applicable.

Environment:

Because of the acute toxicity to aquatic organisms, 72 hr E_rC_{50} ≤ 1 mg/L, Ampholyt classifies under the CLP Regulation (EC) No 1272/2008 as *Acute Cat. 1*. This carries the Hazard Statement H400: *Very toxic to aquatic life*. Additionally, the lowest chronic aquatic endpoint, the 21d NOEC of 0.0023 mg a.s/L, results in a further classification of *Chronic Cat. 1,* as the NOEC ≤ 0.01 mg/L. This carries the Hazard Statement H410: *Very toxic to aquatic life with long lasting effects*.
The CLP Regulation (EC) No 1272/2008 allows for the 10-day window condition to be waived for UVCB or complex, multi-constituent substances with structurally similar constituents. This category includes multicomponent substances whose structure suggests that they will have surface active properties, as is the case for Ampholyt. In the case of amines, hydrogenated tallow alkyl, the 10-day window condition was waived, and for the purposes of classification it considered to be readily biodegradable[2].

Therefore, the acute M-factor for Ampholyt is 10, whereas the chronic M-factor is 1.

2.1.3.2 Proposal for the classification and labelling of the product(s)

The proposed classification for the product (TEGO 2000) containing Ampholyt is:

Proposed classification for the biocidal product (TEGO 2000) according to CLP Regulation (EC) No 1272/2008

Pictogram: (for labelling)		
Signal word:	DANGER	
Hazard Statements: (for labelling)	H314 H361f H373 H410 EUH401	Causes severe skin burns and serious eye damage Suspected of damaging fertility (Category 2) May cause damage to organs (**eyes, mesenteric lymph nodes, male/female genital systems)** through prolonged or repeated exposure (STOT RE Cat 2) Very toxic to aquatic life with long lasting effects To avoid risks to human health and the environment, comply with the instructions for use
Precautionary Statements: (for labelling)	P262 P305+351+338 P337+313	Do not get in eyes, on skin, or on clothing IF IN EYES: Rinse cautiously with water for several minutes. Remove contact lenses, if present and easy to do. Continue rinsing. If eye irritation persists: Get medical advice/attention Wear protective gloves/protective clothing/eye protection/face protection

[2] Committee for Risk Assessment RAC Annex 1 Background document to the Opinion proposing harmonised classification and labelling at Community level of amines, coco alkyl ECHA/RAC/CLH-O-0000002198-71-01/F EC number: 262-976-6 CAS number: 61788-45-2, December 2011

	P273 P391 P501	Avoid release to the environment Collect Spillage Dispose of contents/ container in accordance with applicable regulations
M-Factors	M = 10 (acute) M = 1 (chronic)	

Justification for the proposal:

Physical-Chemical Properties:
The product is not explosive, flammable or oxidising. Neither is the product a compressed gas. The applicant should provide a corrosivity study for metals or provide a scientific statement with regards to the products corrosivity to metals.

Human Health:

Acute Toxicity
Ampholyt has a Rat Oral LD_{50} value of 3300 and 4318 mg/kg body weight for males and female rats respectively. Classification according to CLP Regulation (EC) 1272/2008 is not warranted for the product.

It is proposed that the biocidal product does obtain a health hazard classification based on *in vivo* corrosivity and irritation studies on rabbit. Results indicate that the active substance should be classified as Danger H314: Causes severe skin burns and serious eye damage.

Sub-chronic and Reproductive Toxicity
In addition, repeated dose studies on the 90-day rat study, the 90-day dog study and the two year mouse study indicate that a proposal of H373 STOT RE Cat 2: May Cause damage to organs (eyes, mesenteric lymph nodes, male/female genital systems) through prolonged or repeated exposure is warranted. Furthermore, reproductive toxicity studies on the 90-day dog and 18-month mouse study indicate that a proposal of H361f Cat 2: Suspected of damaging fertility is applicable.

Environment:
Because of the acute toxicity to aquatic organisms, 72 hr $E_rC_{50} \leq 1$ mg/L, Ampholyt classifies under the CLP Regulation (EC) No 1272/2008 as *Acute Cat. 1*. This carries the Hazard Statement H400: *Very toxic to aquatic life*. Additionally, the lowest chronic aquatic endpoint, the 21d NOEC of 0.0023 mg a.s/L, results in a further classification of *Chronic Cat. 1*, as the NOEC ≤ 0.01 mg/L. This carries the Hazard Statement H410: *Very toxic to aquatic life with long lasting effects*.

The CLP Regulation (EC) No 1272/2008 allows for the 10-day window condition to be waived for UVCB or complex, multi-constituent substances with structurally similar constituents. This category includes multicomponent substances whose structure suggests that they will have surface active properties, as is the case for Ampholyt. In the case of amines, hydrogenated tallow alkyl, the 10-day window condition was waived, and for the purposes of classification it considered to be readily biodegradable[3].

Therefore, the acute M-factor for Ampholyt is 10, whereas the chronic M-factor is 1.

[3] Committee for Risk Assessment RAC Annex 1 Background document to the Opinion proposing harmonised classification and labelling at Community level of amines, coco alkyl ECHA/RAC/CLH-O-0000002198-71-01/F EC number: 262-976-6 CAS number: 61788-45-2, December 2011

2.2 SUMMARY OF THE RISK ASSESSMENT

2.2.1 Human Health Risk Assessment

2.2.1.1 Hazard Identification

The data requirements for identification of the potential health hazard of Ampholyt (100% technical material) and Ampholyt have been investigated.

2.2.1.1.1 Toxicokinetics, Metabolism and Distribution

Toxicokinetics, metabolism and distribution was performed in accordance with OECD 417 and the routes of inhalation, oral adsorption and dermal adsorption investigated. In plasma, only parent substances but no transformation products could be identified. A first-pass effect following intestinal absorption is therefore unlikely. In urine and faeces, parent substances as well as transformation products were detected. Metabolites in both urine and faeces were characterised by oxidation (hydroxylation) of the parent and some other metabolites like dehydrogenated and acetylated compounds, especially for C12 PDA. The most abundant compounds in urine were the oxidation products. The elimination half-life was approx. 74 h both in the p.o. and i.v. group. The systemic bioavailability, assessed by comparison of plasma radioactivity after oral and intravenous administration, was determined to be 34%. The recovery of radioactivity in the examined organs was greatest in the liver (less than 4 %). In all other tissues the radioactive recovery was low (≤ 1.00 %).

The following endpoints are considered relevant based on the Absorption, Distribution, Metabolism and Excretion:

Dermal Absorption
2 % for the high exposure (undiluted Ampholyt-100% technical material), and 15 % (15.1%) for the low exposure scenario (1 % Ampholyt, i.e. 0.2 % active substance).

Oral Absorption

Oral availability was determined through a bioavailability study where the plasma concentration post i.v. injection was compared to the plasma concentration p.o. administration. Oral availability was determined to be 34%.

Inhalation absorption
Assumed to be 100%

Bioavailability
The systemic bio-availability, assessed by comparison of plasma radioactivity after oral and intravenous administration was determined to be 34%.

2.2.1.1.2 Acute Toxicity

Oral Toxicity

The Acute Oral Toxicity (Rat) study undertaken by Meisel (1984) found Ampholyt to have an LD_{50} of value between 3,300 and 4,318 mg/kg body weight. The lyophilized formulation of Ampholyt (100% technical material) was determined (Prinsen, 2003) to have a Rat Oral LD_{50} value between 300 and 2,000 mg/kg body weight. According to the requirements specified by Regulation (EC) 1272/2008 and subsequent regulations, Ampholyt requires classification for acute oral toxicity with classification Category 4, signal word "warning"

and the hazard phrase "H302: Harmful if swallowed" in accordance with the classification criteria of Regulation 1272/2008.

Acute Oral Toxicity Classification:
Ampholyt (100% technical material) requires classification:

- Warning H302: "Harmful if swallowed" in accordance with the classification criteria of CLP Regulation 1272/2008.

Ampholyt has a Rat Oral LD_{50} value of 3300 and 4318 mg/kg body weight for males and female rats respectively. Classification according to CLP Regulation (EC) 1272/2008 is not warranted for the product.

Skin and Eye Irritation

According to the requirements Ampholyt does not requires classification for dermal toxicity (LD_{50}, dermal, rat > 2000 mg/kg). The active substance is a 20% aqueous solution of the active matter ("product by process"), from which the active ingredient is not volatile (vapour pressure < 1.9×10^{-4} Pa). The active substance is neither a powder nor is it to be included in powdery preparations. Furthermore, the substance is not intended to be applied in a manner leading to generation of aerosols, particles or droplets in the inhalable size range (MMAD < 50 µm). Therefore, the generation of data on the inhalation toxicity is not considered to be required.

Skin and eye corrosive properties were observed during the testing of Ampholyt and Ampholyt (100% technical material), and therefore, classification with the following is warranted.

Skin Irritation and Corrosivity Classification:

- Danger H314 "Causes severe skin burns and serious eye damage" according to CLP Regulation 1272/2008.

Skin and eye corrosive properties were observed in the tests using Ampholyt (TEGO 2000) and Ampholyt (100% technical material). Therefore, the classification of Ampholyt as H314 "Causes severe skin burns and serious eye damage", according to CLP Regulation 1272/2008, warrants the incorporation of a qualitative local risk assessment to address the potential risks associated with its use to the skin and to the eye. Study data suggests that the technical active substance and products may cause corrosion to skin and eyes under the normal conditions of use through the mixing and loading phase of an application process. The use of PPE including protective eyewear is recommended because of corrosive and irritant local effects.
The potential risk of local toxicity from use is presented, in accordance with the Guidance for Human Health Risk Assessment Version 1.0 – December 2013, in the Qualitative Assessment below.

Guidance for the concluding qualitatively on the acceptability of the risk for the General Public

Hazard		Exposure Information		
Hazard Category	**Effects**	**Frequency & Duration of Potential Exposure***	**Degree of Potential Exposure under Best Practice Conditions**	**Relevant RMMs (PPE not relevant)**
High	Skin Corrosion 1C (H314)	Equal to or less than once per week and equal to or less than few minutes per day	Practically no exposure	Labelling, instructions for use; Child proof closure; Packaging eliminating exposure.

*Duration of potential exposure might be significantly lower than the duration of task/use/process

Guidance for the concluding qualitatively on the acceptability of the risk for Professional Exposure

High Hazard				
Hazard	**Exposure Information**			
Effects	**Frequency & Duration of Potential Exposure***	**Degree of Potential Exposure under Best Practice Conditions**	**Relevant RMMs**	**PPE**
Skin Corrosion 1C (H314)	Few minutes per day or less	High level of containment, practically no exposure; no splashes, no hand to eye transfer, no (liquid or solid) aerosol formation e.g. exposure below or similar to brief contact with technical RMM and PPE as touching of contaminated surfaces	**Measures to ensure well controlled exposure, such as:** **1). Technics** Containment as appropriate; Segregation of the emitting process; Effective contaminant extraction; Good standard of general ventilation; Minimisation of manual phases; Regular cleaning of equipment and work area; Avoidance of contact with contaminated tools and objects; **2). Organisation** Minimise number of staff exposed; Management/supervision in place to check that the RMMs in place are being used correctly and OCs followed; Training for staff on good practice; Good standard of personal hygiene.	**Typical PPE may include:** Substance/task appropriate gloves; Skin coverage with appropriate barrier material based on potential for contact with the chemicals; Substance/task appropriate respirator; Optional face shield; Eye protection

*Duration of potential exposure might be significantly lower than the duration of task/use/process

Skin Sensitisation

No skin reactions were observed after the challenge procedure using the lyophilised form of the active substance. Thus, no classification for skin sensitisation of Ampholyt is required. Consequently, Ampholyt, which is a 20 % (w/w) aqueous solution of the active matter, likewise does not require classification for skin sensitising properties.

2.2.1.1.3 Sub-chronic Toxicity

Based on the results of the studies in rats and dogs, the lowest NO(A)EL for sub-chronic toxicity of Ampholyt was 2.5 mg a.s./kg bw/day (rats, 90 days). The target organ appeared to be the mesenteric lymph nodes in rats. In dogs, changes in haematological and biochemical parameters were observed as the critical effects, leading to unspecific organ alterations like discolouration (kidneys, liver, pancreas, spleen and/or lungs) and body weight loss. Atrophy of both male and female genital systems was seen from 75 mg/kg bw/day equivalent to 15 mg a.s./kg bw/day.

The overall lowest NO(A)EL for sub-chronic toxicity in terms of active substance is thus derived from the oral 90-d study in rats. Thus, the following endpoints for repeated dose toxicity may be forwarded to the risk assessment:

$NOAEL_{subchronic}$ = 2.5 mg a.s./kg bw/d

$NOAEL_{chronic}$ = 1.03 mg a.s./kg bw/d

2.2.1.1.4 Genotoxicity

Ampholyt was tested in a battery of three *in vitro* assays measuring several endpoints of potential genotoxicity such as gene mutation and chromosomal aberration.

Ampholyt was determined to not exhibit potential to induce gene mutation as exhibited by the overall negative results of the chosen bacterial *in vitro* genotoxicity testing processes undertaken. In the chromosomal aberration test with CHO cells, no indication for a clastogenic effect was observed either in the presence or the absence of exogenous metabolic activation. In addition there was no evidence of gene mutation in bacterial or mammalian cells in culture. Taken together, the weight of evidence indicates the absence of genotoxic effects of Ampholyt on chromosomes.

Based on the overall negative results of the recommended *in vitro* genotoxicity test battery system, Ampholyt may be regarded as void of genotoxic potential and, in accordance with the COM Guidance on a Strategy for Genotoxicity Testing of Chemical Substances and the EFSA Scientific Committee Guidance on Genotoxicity Testing Strategy (EFSA, 2008B), it is not deemed necessary to undertake further Tier 2 *in vivo* testing.

2.2.1.1.5 Chronic Toxicity

The chronic NOAEL derived from the combined chronic toxicity and carcinogenicity study in mice is established at 2 mg a.s./kg bw/d in males and was not identified in females (histological change to muscle at 4 mg a.s./kg bw/d).

The lowest NOAEL for chronic toxicity was 3.8 mg Ampholyt 15 DL/kg bw/d. In terms of the active substance (Ampholyt), the lowest chronic NOAEL is therefore 1.03 mg a.s./kg bw/d (male rats, 2-years, read-across from Ampholyt 15).

$NOAEL_{chronic}$ = 1.03 mg a.s./kg bw/d

Chronic Toxicity Classification:
Ampholyt (100% technical material): Suggested classification of Danger H372 STOT RE 1 "Causes damage to organs through prolonged or repeated exposure" according to CLP Regulation 1272/2008.

Ampholyt: Suggested classification of Warning H373 STOT RE 2 "May cause damage to organs through prolonged or repeated exposure" according to CLP Regulation 1272/2008.

2.2.1.1.6 Reproductive and Developmental Toxicity

Rat Studies
Dams treated with 250 mg/kg b.w./day showed maternal toxicity comprising a decrease in body weight and body weight gain, which were statistically significant on gestation days 13 to 17 and 11 to 17, when compared to the control group. Females treated with 250 mg/kg bw/day showed a statistically significant decrease in food consumption on gestation days 6 to 17. Four animals out of 24 in the high dose group showed generalised clinical findings including hunched posture, laboured respiration, rales, brown discolouration of the snout, salivation and piloerection. The maternal NO(A)EL is set at 100mg/kg bw/day (20 mg a.i. /kg bw/day equivalents).

External examination of the foetuses did not reveal any findings among foetuses of litters treated with TEGO 2000 that were considered to be an adverse effect of the test substance. There are minor morphological changes amongst all litters across all dose groups as well as the control. In general there were no indications of any consistent treatment or dose related effects on any of the parameters investigated. However, in the 100 and 250 mg/kg dose groups there appeared to be a treatment related increase in the incidence of unilateral hydroureter compared with the concurrent controls in conjunction with interparietal and supraoccipital delayed ossification. There was also a slight increase in unilateral renal pelvic cavitation with treatment. There were no increases recorded in the incidences of bilateral hydroureter or bilateral renal pelvic cavitation. Oral administration of TEGO 2000 to pregnant Wistar rats during the period of organogenesis at dose levels up to 40 mg/kg bw/day (8 mg/kg bw/day 100% a.i. equivalent) did not result in any adverse effects during foetal development

Parental toxicity consisted of clinical signs (only P-animals), effects on body weights and food consumption for P- and F1-animals treated at 100 mg/kg body weight/day. Reproductive toxicity consisted of a reduced number of implantation sites at necropsy and living pups on day 1 of lactation for P-animals treated at 100 mg/kg body weight/day. The development of the pups was not affected up to 100 mg/kg bw/day. Based on the results of this study, the parental and reproductive NOAEL was established at 30 mg/kg bw/day. The developmental NOAEL was established at 100 mg/kg bw/day.

Dog Study
Dose-related atrophy was observed in the male and female genital systems concomitant with but compounded by significant body weight loss at the highest dose. The a.i. dose equivalents were 0, 5, 15, 45 mg/kg bw/day (corresponding 0, 25, 75, 225 mg/kg bw/day TEGO 2000, a synonym of Ampholyt). Males treated at 45 mg a.s./kg bw/d exhibited prostates that were downsized, but no weight measurements were reported. Atrophy of the prostate was seen in 2/4, 4/4 and 4/4 animals of the low, mid and high dose groups respectively. In high dose females a reduction in the size of the uterus, both relative (to bw) and in absolute weight (g) was recorded.

The histopathological report revealed a dose-related atrophy in the male and female genital system in intermediate and high dosed animals. Atrophy of the male and female genital system (cervix, epididymis, ovary, prostrate, uterus and vagina from 15 mg/kg bw/day and atrophy of the germinal epithelium in the testes at 45 mg/kg bw/day only). Confounding effect: all animals administered 45 mg a.s./kg bw/d showed emesis and

salivation on most treatment days. Significant reductions in body weight observed at week 6 and 13 for both males and females.

Vacuolisation of epithelial cells in the epididymides of most mice were recorded in both the interim and final sacrifice groups in high dose (20/7 mg a.i. eq./kg bw/day) animals and mid-dose animals (6 mg a.i. eq./kg bw/day) that had prematurely died.

Mouse

A consistent reduction in body weight and body weight gain was observed and is considered adverse. A dose-related vacuolisation of the epithelial cells lining the epididymides was observed in males. The a.i. dose equivalents in males for the combined chronic toxicity and carcinogenicity study were 0, 2, 6, 20/7 mg/kg bw/day (equivalent to 0, 10, 30, 100/35 mg/kg bw/day TEGO 2000).

Reproductive Toxicity Classification

Classification for developmental effects were deemed not to be warranted as no clear effects were seen on the reproductive organs of rats from either the 2-gen study or the 90 dietary rat study. Classification for fertility effects were deemed warranted based upon the findings from the 90 day dog dietary study (Leuschner 2008) and supported by findings in the mouse 18 month dietary study by Hansen et al., 2012 (both studies are described above in the *Repeat dose Toxicity* sub-section).

Ampholyt and Ampholyt 20 (TEGO 2000) are to be classified as follows:

- "Warning" Repro. Category 2 H361f: Suspected of damaging fertility according to CLP Regulation (EC) 1272/2008.

2.2.1.1.7 Neurotoxicity

A neurotoxicity study with Ampholyt is not considered to be required due to the chemical structure of the compound, since neurotoxicological effects would not be expected.

2.2.1.1.8 Hazard Summary

Test	Result	Classification
Acute Oral Toxicity	Rat Oral LD_{50} ≥ 300 and 2,000 mg/kg body weight	No classification
Acute Dermal Toxicity	LD_{50}, dermal, rat > 2000 mg/kg	No classification
Acute Skin Irritation	Causes severe burns and eye damage	Danger: H314
Acute Eye Irritation		Danger: H318
Skin Sensitization	No skin reactions were observed after the challenge procedure	No classification
Genotoxicity	Void of Genotoxic potential *in vitro* – no *in vivo* tests conducted based on the findings of the *in vitro* test battery	Void of genotoxic potential *in vitro*
Sub-chronic Toxicity Chronic Toxicity	$NOAEL_{subchronic}$ = 2.5 mg a.s./kg bw/d $NOAEL_{chronic}$ = 1.03 mg a.s./kg bw/d	**Ampholyt (active ingredient):** Danger STOT RE1: H372 **Ampholyt 20 (TEGO 2000):** Warning STOT RE2: H373

Reproductive and Development Toxicity	NOAEL maternal (rat): 20 mg a.i./kg bw/day NOAEL developmental (rat): 8 mg a.i./kg bw/day NOAEL maternal and developmental (rabbit):10 mg a.i./kg bw/day NOAEL reproductive: 30 mg/kg bw/d	Warning: H361f (Repro. Cat 2)
Neurotoxicity	Not applicable due to the chemical structure of the compound (not an organophosphorus compound)	-

2.2.1.2 Effects Assessment

The following short- (acute), medium- and long-term AELs were derived in relation to the effects assessment and for determination of the risk characterisation for human health. All three values were adjusted for oral adsorption of 34%.

AEL$_{long-term}$
The 2-year feeding study in the rat produced an NOAEL of 1.03 mg/kg bw/day and this can be used to establish a systemic AEL long-term reference value. This value becomes 0.35 mg/kg bw/day when adjusted for 34% oral absorption. The AEL long-term reference value is derived by dividing the adjusted NOAEL value by an overall safety factor of 100. This gives an AEL long-term reference value of 0.0035 mg/kg bw/day.

AEL$_{acute}$
The rat developmental toxicity study is the acute study used to provide an NOAEL value that can be used to establish a systemic AEL acute reference value. The NOAEL from this study is 8 mg/kg bw/day, which becomes 2.72 mg/kg bw/day when adjusted for oral absorption of 34%. An AEL acute reference value is derived by dividing the adjusted value by an overall assessment factor of 100. This gives an AEL acute reference value of 0.027 mg/kg bw/day.

AEL$_{medium-term}$
The 90-day feeding study in the rat produced an NOAEL of 2.5 mg/kg bw/day and this can used to establish systemic AEL medium-term reference values. The NOAEL from this study is 2.5 mg/kg bw/day, which becomes 0.85 mg/kg bw/day when adjusted for oral absorption of 34%. The AEL medium-term reference value is derived by dividing the adjusted value by an overall assessment factor of 100. This gives an AEL medium-term reference value of 0.0085 mg/kg bw/day (rounded).

2.2.1.3 Exposure Assessment

Product Type 3: Veterinary hygiene biocidal products

A summary of the scenarios evaluated for the exposure/effect assessment and risk characterisation under PT3 (Veterinary hygiene biocidal products) are outlined below.

Scenario	Summary description of scenario
Spraying Application	Primary exposure; Exposed group: Professionals only; Phases of exposure assessed include: **Mixing and loading**: Handling containers and diluting with water for low pressure spraying. Low-pressure spraying: Spray Model 1 (TNsG 2002) incorporates mixing and loading with an exposure time combining mixing and loading and application of 120 min. Tier I with no PPE. Tier II with PPE (coveralls and gloves, 10%). **Application**: Low-pressure spraying: Spray Model 1 (TNsG 2002) which incorporates mixing and loading with an exposure time for mixing and loading and application of 120 min. Tier I with no PPE. Tier II with PPE (coveralls and gloves, 10%). **Post-application**: Wash-down, disposal of the remaining cleaning solution/waste water and disposal of empty containers. Post-application time assumed: 9 minutes. Tier I with no PPE. Tier II with PPE (coveralls and gloves, 10%).0%). **Combined exposure**: Mixing and loading, application and post application (coveralls and gloves, 10%) for low-pressure spraying
Footbath Application:	Primary exposure; Exposed group: Professionals only; Phases of exposure assessed include: **Mixing and loading for preparation**: handling containers and diluting with water for footbath preparation and, where relevant, pouring of the prepared solution from a mixing bucket to the footbath tray. EUROPOEM II DB for manual pouring and loading (up to 20L). Tier I with no PPE. Tier II with PPE (coveralls and gloves, 10%). **Application**: Negligible exposure. **Post-application**: Wash-down, disposal of the remaining cleaning solution/waste water and disposal of empty containers. Tier I with no PPE. Tier II with PPE (coveralls and gloves, 10%). **Combined exposure**: mixing and loading, application and post application (coveralls and gloves, 10%) for footbaths
RTU Application: Combined exposure	Primary exposure; Exposed group: Professionals only; Phases of the combined exposure assessed for an RTU include: **Application**: Model TNsG Consumer Spraying and Dusting Model 2 – hand held trigger spray. **Post-application**: Wash-down, disposal of the remaining cleaning solution/waste water and disposal of empty containers. Application time assumed: 30 minutes (total exposure including application and post-application). Wiping step (Surface disinfection model 1): 15 minutes. Tier I with no PPE. Tier II with PPE (coveralls and gloves, 10%).
Secondary exposure	Secondary exposure; Exposed group: Child; Modelled scenario: Child in contact (oral and dermal exposure) with residues following disinfection procedure.

Primary (direct) exposure

Spraying and footbath scenarios
The systemic doses for mixing and loading, application and post application procedures are 0.1726 mg/kg bw/day for Tier 1 (worst case, unprotected) and 0.0128 mg/kg bw/d for Tier 2 (coveralls and gloves) from application as a veterinary hygiene disinfectant by spraying. The footbath disinfection method leads to a total systemic exposure of 0.06

mg/kg bw/d for Tier 1(unprotected) and 0.006 mg/kg bw/day for Tier 2 (coveralls and gloves) for footbath application.

The risk characterisation indicates that typical disinfection procedures for footbath (passive) application using TEGO 2000 (i.e. active ingredient Ampholyt) are not a risk to trained professional workers, wearing the appropriate prescribed PPE, applying the disinfectant as a surface disinfectant (Product Type 3, reasonable worst case) when compared to the $AEL_{medium\text{-}term}$ (0.0085 mg/kg bw/day). However, a safe operator exposure dose for low-pressure spraying of stables with Product Type 3, when compared to the $AEL_{medium\text{-}term}$ (0.0085 mg/kg bw/day), could not be established.

Evaluators note:

The exposure times for the above modeled processes are the retained exposure times agreed by the Member States at the Biocides Working Group (WG) Meeting in Helsinki in June of this year (2014). Based upon the exposure times chosen and incorporation of the appropriate PPE (coveralls and gloves) no safe use could be achieved for PT3 – Footbath and PT3 – Spraying at 120 minutes.

RTU- handheld trigger spray products

In-use survey data was subsequently submitted, following the WG meeting in June, to the RMS by the Applicant for a ready-to-use spraying product (0.2% a.s.). This new data was modeled according to TNsG Consumer Spraying and Dusting Model 2 – hand held trigger spray (TNsG Human Exposure, part 2, p. 197) and Surface Disinfection Model 1 (TNsG Human Exposure, part 2, p. 173) to incorporate a 15 minute wiping step. A total systemic dose of 0.091 mg/kg bw/day (worst case) was determined for unprotected application (spraying and wiping) and post application procedures for the ready-to-use product (0.2% a.s.), as described in detail in Document II-B. Of higher relevance is the Tier 2 (coated coveralls and gloves) assessment for the spraying application considering the obligate protection of workers handling the disinfectant, being 0.0026 mg/kg bw/day.
The risk characterisation indicates that typical disinfection procedures for spraying using the ready-to-use (0.2% a.s.) product does not present a risk to trained professional workers while wearing the appropriate prescribed PPE and as such a safe operator exposure dose for trigger spraying of veterinary hygiene areas with Product Type 3, when compared to the $AEL_{long\text{-}term}$ (0.0035 mg/kg bw/day) and $AEL_{medium\text{-}term}$ (0.0085 mg/kg bw/day), was established.

Secondary (Indirect) exposure

When applied as product type 3, indirect exposure for small children when crawling on floors in a disinfected area (dermal contact and hand-to-mouth transfer, see Doc. II-B) was calculated. Thus, secondary exposure via the dermal and oral route of children or infants was considered.

The exposure scenario for product type 3, presented in detail in Doc. II-B, results in a maximum dose of 6.42×10^{-2} mg/kg bw/day (crawling infant, dermal route) and 2.91×10^{-5} mg/kg bw/d (oral, hand-to-mouth transfer), which gives a total exposure of 6.42×10^{-2} mg/kg bw/d and is considered as an absolute worst case. In relation to the acute NOAEL of 2.7 mg a.i./kg bw/d a MOS of 42 is derived. A MOS below 100 is not considered to be safe. Therefore, secondary exposure (crawling infant) following application of PT 3 was not found to be safe when modeled for oral and dermal exposure to a toddler when compared to the acute NOAEL (0.027 mg/kg bw/day).

A risk of secondary exposure is identified based on the worst case exposure of a crawling child on a floor in contact (oral and dermal exposure) with residues following hard surface

disinfection. However, for industrial or cross-contamination prevention application scenarios this situation of a crawling child is not considered appropriate.

Table 2.2.1.3-1 MOS values for the critical effects concerning the workplace exposure

Workplace operation	PPE	Exposure path	Body dose [mg/kg bw/d] (reasonable worst case)	Repeated dose toxicity ($NOAEL_{systemic}$ = 0.85 mg/kg bw/d)
				MOS
PT3, spraying	Coveralls and gloves (Tier 2)	Dermal, all phases	0.0128	66.34
PT3, footbath preparation/application	Coveralls and gloves (Tier 2)	Dermal, all phases	0.006	141.84

Table 2.2.1.3-2 Exposure/AEL values for the critical effects concerning the workplace exposure

Workplace operation	PPE	Exposure path	Body dose [mg/kg bw/d] (reasonable worst case)	Repeated dose toxicity ($AEL_{systemic}$ = 0.0085 mg/kg bw/d)
				Exposure/ $AEL_{systemic}$
PT3, spraying	Coveralls and gloves (Tier 2)	Dermal, all phases	0.0128	1.51
PT3, footbath preparation/application	Coveralls and gloves (Tier 2)	Dermal, all phases	0.006	0.706

Table 2.2.1.3-3 MOS and Exposure/AEL values for the critical effects concerning workplace exposure to the ready-to-use product (0.2% a.i.)

Workplace operation	Exposure path	Body dose [mg/kg bw/d] (reasonable worst case)	Repeated dose toxicity ($NOAEL_{systemic}$ = 0.85 mg/kg bw/d)	
			Repeated dose toxicity ($AEL_{systemic}$ = 0.0085 mg/kg bw/d)	
PT3 – trigger spray (Tier 2)	Dermal, all phases	0.0026	MOS = 327	
PT3 – trigger spray (Tier 2)	Dermal, all phases	0.0026	Exposure/AEL = 0.31	

2.2.1.4 Risk Characterisation

Product Type 3

Primary (direct) exposure

The risk characterisation, based on Tier 2 exposure estimates, indicates an acceptable use for trained professional workers applying the disinfectant as a surface disinfectant for footbath disinfection (product type 3, Tier 2) and the ready-to-use product (0.2%) when compared to the $NOAEL_{systemic}$ (0.85 mg/kg bw/day). Appropriate PPE (coveralls and gloves) are worn during both disinfection processes. For Tier 2 the margin of safety (MOS)

for the $NOAEL_{systemic}$ resulted in a MOS of 142 (coveralls and gloves; footbath) and 327 (coveralls and gloves; hand-held trigger spray).
The risk characterisation for low-pressure spraying application for stables did not return a safe use for trained professional workers when applying the disinfectant when compared to the $AEL_{medium-term}$ (0.0085 mg/kg bw/day) which resulted in an Exposure/ $AEL_{medium-term}$ of 1.51. A value >1 is not considered safe.

The risk characterisation for footbath and trigger spray disinfection resulted in an Exposure/ $AEL_{medium-term}$ of 0.706 and 0.31 respectively and therefore are considered safe.

Secondary (Indirect) exposure

When applied as product type 3, indirect exposure for small children when crawling on floors in a disinfected area was calculated (dermal contact and hand-to-mouth transfer, see Doc. II-B). Thus, secondary exposure via the dermal and oral route of children or infants was considered.

The exposure scenario for product type 3, presented in detail in Doc. II-B, results in a maximum dose of 6.42×10^{-2} mg/kg bw/day (crawling infant, dermal route) and 2.91×10^{-5} mg/kg bw/d (oral, hand-to-mouth transfer), which gives a total exposure of 6.42×10^{-2} mg/kg bw/d and is considered as an absolute worst case. In relation to the acute NOAEL of 2.7 mg a.i./kg bw/d a MOS of 42 is derived. A MOS below 100 is not considered to be safe. Therefore, secondary exposure (crawling infant) following application of PT 3 was not found to be safe when modeled for oral and dermal exposure to a toddler when compared to the acute NOAEL (0.027 mg/kg bw/day).

A risk of secondary exposure is identified based on the worst case exposure of a crawling child on a floor in contact (oral and dermal exposure) with residues following hard surface disinfection. However, for industrial or cross-contamination prevention application scenarios this situation of a crawling child is not considered appropriate.
Although exposure of this type must be regarded as acute or at most sub-chronic in view of its irregularity (particularly since stable disinfection occurs at most three times per year), even comparison with a chronic endpoint indicates that residues of Ampholyt after disinfection are of no particular concern.

Table 2.2.1.3-4 MOS values for the critical effects concerning the workplace exposure towards active substance.

Workplace operation	PPE	Exposure path	Body dose [mg/kg bw/d] (reasonable worst case)	Repeated dose toxicity ($NOAEL_{systemic}$ = 0.85 mg/kg bw/d)
				MOS
PT3, trigger spraying, low-pressure spraying or wiping	NA	Dermal and oral(Child crawling on treated floors)	6.42×10^{-3}	42

Table 2.2.1.3-5 Exposure / AEL values for the critical effects concerning the workplace exposure towards active substance.

Workplace operation	PPE	Exposure path	Body dose [mg/kg bw/d] (reasonable worst case)	Repeated dose toxicity ($AEL_{systemic}$ = 0.0085 mg/kg bw/d)
				Exposure / $AEL_{systemic}$
PT3, trigger spraying, low-pressure spraying or wiping	NA	Dermal and oral(Child crawling on treated floors)	6.42×10^{-3}	238

Exposure modelling yielded a safe use for Ampholyt for operators for footbath (passive) application using the substance as Product Type 3. The applicant has employed a chronic exposure function when calculating primary exposure to PT3. In addition an additional protection factor for "coveralls and gloves" at Tier 2 has also been employed for this application scenario. A safe-use was also achieved for Product Type 3 ready-to-use (0.2% a.i.) trigger spray product, when additional protection factors (coveralls and gloves), were incorporated into the modelling. However, exposure modelling for PT3-low pressure spraying application in stables did not yield a safe use for Ampholyt for operators.

Exposure to a child crawling on a treated floor was modelled and taken as worst case and resulted in a MOS of 42 when compared to the acute NOAEL. A risk of secondary exposure is identified based on the worst case exposure of a crawling child on a floor in contact (oral and dermal exposure) with residues following hard surface disinfection. However, for industrial or cross-contamination prevention application scenarios this situation of a crawling child is not considered appropriate.

Qualitative Risk Assessment

There is a need to undertake a qualitative risk assessment, discussed at the BPC WG Meeting in June 2014, due to the potential for human exposure arising from the transfer of residues and contaminated food or feedstuffs following use of the product types in food and feed areas or areas in which indirect or secondary exposure could arise.

The qualitative risk of exposure to Ampholyt arising from residues or contaminated food or feed stuff is expected to be limited given the intrinsic properties of the substance (Ampholyt is a highly water soluble disinfectant) and also due to the risk mitigation steps incorporated into the disinfection process via the washing down and rinsing of the treated area with sterile water post-application. In addition, the likelihood of the entire product residue being transferred to food or feed products is low. This combined with the intrinsic properties of the substance and the wash-down step post-disinfection the predicted transfer of residues and contamination of food and feedstuffs are likely to be negligible. However, the accuracy of this cannot be concluded due to the absence of relevant guidance or data to quantitatively confirm this.

Aggregate Exposure

The need to discuss the aggregated exposure arising when an active substance is approved across multiple product types has been discussed during the substance peer review process. As such, it was proposed that an assessment would be required for Ampholyt. However, in the absence of robust guidance that would allow a quantitative assessment to be carried out, it was agreed that a qualitative analysis would be conducted. To summarise there are three possible situations where aggregated exposure relating to human health may be expected:

Usage of the products in a PT2, 3 and 4 use work as a professional.

Additional indirect or secondary exposure may also aggregate to the above from the exposure to residues from treated surfaces.

2.2.1.5 Evaluators note

The exposure assessment and risk characterisation for footbath application of product type 3, is based upon an exposure to 2kg of the product using EUROPOEM II database for assessment. The in-use survey data, subsequently submitted to the RMS by the Applicant for a ready-to-use spraying product at 20%, was modelled according to TNsG Consumer Spraying and Dusting Model 2 – hand held trigger spray (TNsG Human Exposure, part 2, p. 197) for a total exposure period of 45 minutes (30 minutes spraying; 15 minutes wiping tasks). The risk characterisation for both footbath application (passive) and the ready-to-use product (0.2% a.s.) returned safe-uses using coveralls and gloves (90% exposure reduction) when compared to the $AEL_{medium\text{-}term}$ (0.0085 mg/kg bw/day).

The risk characterisation for low-pressure spraying using TEGO 2000, as product type 3, did not result in a safe operator exposure dose at the duration of 120 minutes (total value for mixing and loading, application and post-application) when compared to the $AEL_{medium\text{-}term}$ (0.0085 mg/kg bw/day). It is therefore not considered safe for operator use in this exposure scenario (i.e. low-pressure spraying of stables). There is the potential to reduce these values if extra risk mitigation factors are incorporated (i.e. new gloves and impermeable coveralls, 95% or 99%).

Safe uses for PT3 have been established for footbath application and hand-held trigger spray ready-to-use product (0.2% a.s.) and at the specified times and volume/weight, when compared to the $AEL_{medium\text{-}term}$ (0.0085 mg/kg bw/day).

A risk of secondary exposure is identified based on the worst case exposure of a crawling child on a floor in contact (oral and dermal exposure) with residues following hard surface disinfection. However, for industrial or cross-contamination prevention application scenarios this situation of a crawling child is not considered appropriate.

2.2.2 Environmental Risk Assessment

2.2.2.1 Fate and Distribution in the Environment

Ampholyt[4]/Ampholyt/100 was rapidly removed in test systems designed to measure ready biodegradability (4.02-8.02 mg/L DOC). Ampholyt is considered as ready biodegradable (not fulfilling the 10-day window) based on a weight of evidence approach. Ampholyt/100 was also rapidly *removed* in STP simulation systems. Ampholyt was identified as non-biodegradable under anaerobic conditions because of its potentially toxic effects to bacteria, at least at the concentrations recommended by the test guideline for anaerobic degradation.

The active substance appears to be hydrolytically stable; and direct photolysis in water is not considered to contribute significantly to abiotic degradation in aqueous systems under environmentally relevant conditions. Any Ampholyt reaching the air compartment is expected to be rapidly eliminated via photo-oxidative reactions (half-life is 0.125 d, 24 hr, 5 x 10^5 molec/cm^3).

Due to the fact that Ampholyt is not a pure substance, but a complex mixture it is hard to obtain robust distribution coefficients. In the adsorption isotherms, the Ka_{OC} for the mixture ranged from 31,660 to 86,743 $\square g^{1-1/n}(cm^3)^{1/n}\ g^{-1}$with an average value of 62,031.6 $\square g^{1-1/n}(cm^3)^{1/n}\ g^{-1}$. 1/n varied from 1.0986 to 1.1745. The desorption kinetics experiments showed that the adsorption was only marginally reversible. Only 4.7–7.7 % of the adsorbed Ampholyt/100 were desorbed. The CA notes the adsorption behaviour of Ampholyt/100 was determined by measuring the total ^{14}C-radioactivity without differentiation of the single components of the test substance. Ampholyt is comprised of approximately 20 constituents. Some of the components have difunctional amine structures with caboxylation grades ranging from 0 – 2. The ionic or even amphoteric nature of the Ampholyt constituents implies that the charge of each individual molecule will be strongly pH dependent. Following discussions at the ECHA WG III meeting (Environmental session) 2014 it was agreed to perform the environmental exposure assessment with the lowest measured Koc (15,431.8 L/kg) to cover the lower range of Kocs and the highest permitted Koc in EUSES (1 x 10^6 L/kg) to cover the higher range of Kocs exhibited by some components of Ampholyt.

2.2.2.2 Effects Assessment

Effects on aquatic organisms

With LC/EC_{50} values below 1 mg/L for fish, algae and aquatic invertebrates, Ampholyt is considered as highly toxic to aquatic organisms.

The E_rC_{50} was 0.0237 mg a.s./L for algae indicates that Ampholyt is highly toxic to aquatic organisms.

The long-term effects of Ampholyt on aquatic organisms are presented for fish, daphnia and algae. By comparison of the NOEC values, *Daphnia magna* (2.3 µg a.s./L) is the most sensitive species with respect to chronic effects, based on cumulative offspring per *Daphnia* and intrinsic rate of increase. *Oncorhynchus mykiss*, appears to be less sensitive, with no observed effects on any test parameter up to the highest tested concentration of 52.3 µg a.s./L. The 72 hour algal test can be considered an acute and chronic study,

[4] Ampholyt 20/100 is the lyophilised active substance
Ampholyt 20 is a trade name and synonym for "TEGO 2000", obtained as a "product by process", i.e., a 20% aqueous solution of the pure active.

therefore the growth rate of unicellular algae was inhibited at a test concentration of NOE_rC = 9.55 µg/L.

The lowest chronic toxicity NOEC with a value of 2.3 µg a.s./L was observed in *Daphnia magna*.

PNEC for aquatic organisms

Chronic toxicity data for three trophic levels of aquatic species are available. The PNEC for aquatic organisms is derived by dividing the NOEC from the chronic Daphnia magna reproduction toxicity study (2.3 µg a.s./L) by an assessment factor of 10, according to the TGD on risk assessment.

PNECaquatic = 2.3 × 10–4 mg/L

PNEC for sediment-dwelling organisms

Specific data on the toxicity of Ampholyt 20 to sediment-dwelling organisms are not available in view of the limited exposure. Nevertheless, assuming equal susceptibility of sediment-dwelling and aquatic organisms to the given chemical, a PNEC for sediment organisms can be estimated using the equilibrium partitioning method (EPM). Two PNECsediment values were derived for Ampholyt 20, representing the two Koc's that were assessed for environmental exposure, these were performed according to the Guidance provided in the TGD. The Koc of 106 L/kg results in a Ksusp-water of 2.5 X104 m3/m3,while the Koc of 15,432.8 L/kg results in a Ksusp-water of 387 m3/m3. The RHOsusp is 1,150 kg/ m3 . Therefore the PNECs for both Koc scenarios are derived as follows:

$$PNECsediment = \frac{Ksusp\text{-}water \cdot PNECwater \cdot 1000}{RHOsusp}$$

PNECsediment = 5 mg/kg wwt (Koc 106 L/kg)
PNECsediment 0.0774 mg/kg wwt (Koc 15,432 L/kg)

According to the TGD p. 112 'the formula only considers uptake via the water phase. However, uptake may also occur via other exposure pathways like ingestion of sediment and direct contact with sediment. This may become important, especially for adsorbing chemicals, for example those with a logKow greater than 3'. Because Ampholyt 20 is an amphoteric molecule and measured Koc values indicate a potentially strong adsorption or binding behaviour, an additional safety factor of 10 is applied to the PEC/PNEC ratio. Therefore the PNECsediment values applied in the risk assessment are reduced by 10 resulting in the following:

PNECsediment = 0.5 mg/kg wwt (Koc 106 L/kg)
PNECsediment 0.00774 mg/kg wwt (Koc 15,432 L/kg)

The effect of Ampholyt on the respiration inhibition of micro-organisms was examined in two activated sludge studies. One study was carried out utilising sludge from a municipal STP. The lower EC_{50} will be carried forward to the risk assessment. The results of the studies presented show that the EC_{50} for Ampholyt is 22 mg a.s/L.

Atmosphere

Ampholyt is rapidly degraded in the atmosphere by photo-oxidative processes. The maximum numerical half-life is 0.125 d (2.993 hr). Due to the low vapour pressure of the active matter of Ampholyt, this degradation path is considered to be of limited relevance in view of the limited exposure to air.

Effects on terrestrial organisms

A chronic toxicity/carcinogenicity study in rats showed treatment related changes in the mesenteric lymph nodes of high dose animals only. Based on the alterations in the mesenteric lymph nodes observed for both sexes, the lowest NOAEL observed was 1.03 mg/kg bw/d for males. Ampholyt is not toxic to mammals.

PNEC for soil organisms

Effect concentrations > 1000 mg/kg for microorganisms and earthworms.
The PNEC for soil organisms is derived from three terrestrial toxicity studies covering three trophic levels. The lowest endpoint was observed for soil microflora, namely a NOEC of 64 mg/kg dwt,which equates 56.6 mg/kg wwt.

Determination of the conversion factor for soil (according to the TGD):

$$\mathrm{CONV_{soil}} = \frac{\mathrm{RHO_{soil}}}{\mathrm{F_{solid}} \times \mathrm{RHO_{solid}}}$$

RHOsoil = 1,700 [kgwwt x m-3]
Fsolid (for soil) = 0.6 [msolid3 x msoil-3]
RHOsolid = 2,500 [kgdwt x msolid-3]
CONVsoil = 1.13 [kgwwt x kgdwt-1]

This results in the following EC50 for wet weight soil:

$$\mathrm{EC_{50\ wwt}} = \frac{\mathrm{EC_{50\ dwt}}}{\mathrm{CONV_{soil}}}$$

No normalisation of the organic content of the soil is performed because according to the TGD, this is only appropriate for non-ionic compounds.

Therefore the PNECsoil can be derived following the selection of the appropriate Assessment Factor (AF) according to Table 20. Taking an AF of 100 (NOEC for one long-term toxicity test), results in a

PNECsoil = 0.57 mg/kg wwt

The lowest EC_{50}-value was observed in *Lactuca sativa* (363 mg a.s./kg). Thus, the lowest relevant EC_{50} value to be forwarded to the risk assessment was identified for *Lactuca sativa*, determined at 363 mg a.s./kg dry soil.

A toxicity study on birds was not carried out and justification for non-inclusion of a study is acceptable. Based on the toxicity of Ampholyt to rats in a 90 days feeding study, Ampholyt is not toxic to mammals.

2.2.2.3 PBT and POP Assessment (including Endocrine Disruption Assessment)

PBT Assessment

Persistence

Ampholyt is considered as ready biodegradable (not fulfilling the 10-day window) based on a weight of evidence approach and does not fulfil the P criteria nor the vP criteria.

Bioaccumulation

Ampholyt is not considered to be of concern with regard to bioaccumulation in either the aquatic or terrestrial food chain.

Whilst no studies were submitted investigating the potential hazards of Ampholyt on non-target aquatic/terrestrial organisms via secondary poisoning, Ampholyt has been shown to be both highly water-soluble and readily biodegradable (failing the 10-d window) in the environment. In addition, Log D values were calculated using a number of software models for the most hydrophobic component of Ampholyt (C16-PDA) (see Document IIA, Section 4.1.2) to determine its potential to bioaccumulate (i.e. lipophilicity). These results showed a maximum Log D value of 4.4 at pH 9, indicating that Ampholyt does not bioaccumulate (criteria agreed at WG III 2014 was Log D ≥ 4.5 at pH 4-9). Additionally, further supporting information provided in the weight of evidence approach including BCF model estimation, environmental degradation and mammalian metabolic pathways support the conclusion that bioaccumulation and secondary poisoning are not of concern for Ampholyt.

Ampholyt does not fulfil the B or vB criteria.

Toxicity

The NOEC value *Daphnia magna* (2.3 µg a.s./L) is the most sensitive species with respect to chronic effects based on *Daphnia magna*. Therefore Ampholyt classifies as toxic.

Other data from chronic toxicity for mammalian toxicity also suggests that the T criterion is met, since the active substance is considered to classify as a STOT RE Cat. 1 according to Regulation EC No. 1272/2008.

Ampholyt is considered to fulfil the T criterion.

Overall PBT conclusion:

Ampholyt is not considered a PBT substance.

Ampholyt is considered not to fulfil the criteria for persistence nor bioaccumulation.

Ampholyt is considered to fulfil the criteria for toxicity.

POP Assessment

Persistence
Ampholyt is considered as ready biodegradable (not fulfilling the 10-day window) based on a weight of evidence approach and is not considered to be persistent.

Bioaccumulation

Ampholyt is not considered to be of concern with regard to bioaccumulation in either the aquatic or terrestrial food chain.

Whilst no studies were submitted investigating the potential hazards of Ampholyt on non-target aquatic/terrestrial organisms via secondary poisoning, Ampholyt has been shown to be both highly water-soluble and readily biodegradable (failing the 10-d window) in the environment. In addition, Log D values were calculated using a number of software models for the most hydrophobic component of Ampholyt (C16-PDA) (see Document IIA, Section 4.1.2) to determine its potential to bioaccumulate (i.e. lipophilicity). These results showed a maximum Log D value of 4.4 at pH 9, indicating that Ampholyt does not bioaccumulate (criteria agreed at WG III 2014 was Log D ≥ 4.5 at pH 4-9). Additionally, further supporting information provided in the weight of evidence approach including BCF model estimation, environmental degradation and mammalian metabolic pathways support the conclusion that bioaccumulation and secondary poisoning are not of concern for Ampholyt.
Ampholyt does not fulfil the B or vB criteria.

Long-range environmental transport

Ampholyt is rapidly degraded in the atmosphere by photo-oxidative processes. The maximum numerical half-life is 0.125 d (2.993 hr). Due to the low vapour pressure of the active matter of Ampholyt, this degradation path is considered to be of limited relevance in view of the limited exposure to air. It is not expected that the substance will fulfil the screening criteria (DT_{50air} 2d) for the potential for long-range environmental transport. Furthermore, there is no monitoring data available or other evidence indicating potential for long-range environmental transport.

Adverse effects (includes endocrine disruption assessment)

The NOEC value for *Daphnia magna* (2.3 µg a.s./L), the most sensitive species with respect to chronic effects, is based on cumulative offspring per *Daphnia* and intrinsic rate of increase and indicates that Ampholyt is toxic to aquatic invertebrates. Therefore Ampholyt classifies as toxic.

In relation to mammalian toxicology, guidance on Scientific criteria for identification of Endocrine disruptors (EFSA, 2013) states that a substance that is Toxic for Reproduction Category 2 and which have toxic effects on the endocrine organs may be considered as having such endocrine disrupting properties. As the classification proposal for Ampholyt suggests "H372: STOT RE Cat 1 Causes damage to organs (eyes, mesenteric lymph nodes, male/female genital systems) through prolonged or repeated exposure" and "H361f Cat 2: Suspected of damaging fertility" identification as an ED may appear justified. However, effects on organ systems precipitating the STOT RE Cat 1 suggest a systemic toxicity mediated by perturbations in the lymphatic system. The male and female genital systems are not selectively impacted but rather are part of a group of organs impacted by Ampholyt's systemic toxicity. When the appropriate ED guidance is in place, these effects will be reconsidered in light of whether Ampholyt is to be identified as an ED.

Overall POP conclusion:

Ampholyt is not considered to fulfil the requirements for it to be identified as a POP substance.In relation to endocrine disruption, it should be noted that, Ampholyt could be considered a possible endocrine disruptor (ED). When the appropriate ED guidance is in place, these effects will need to be reconsidered in light of whether Ampholyt is to be identified as an ED.

2.2.2.4 Exposure Assessment

Environmental Exposure

Product Type 3

TEGO 2000 (Ampholyt) is intended to be used in veterinary hygiene as defined by the manufacturer as follows:

- Disinfectants used in areas in which animals are housed, kept or transported
- General disinfectants used in footbaths for animals for prevention of cross contamination

The main emission route to the environment for PT 3 disinfectants is to manure/slurry, air and agricultural soil (from spreading of manure/slurry). Run-off as well as leaching processes after manure/slurry application to agricultural soil can also lead to exposure of biocides to surface water and groundwater, respectively.

Emissions and PECs were calculated in accordance with the new ESD (2011) for product type 3 for the following scenarios which covers the intended uses:

(3.1) Disinfection of animal housing
(3.2) Disinfection of vehicles used for animal transport
(3.4) Disinfection for veterinary hygiene: footwear and animals'

Application of ampholyt by ready-to-use (RTU) products used for the spot treatment of surfaces was not considered for the PT3 scenarios in the environment but it should be noted that such uses could be applicable for applications at product authorisation as an in-use survey was subsequently submitted to the RMS by the applicant, as part of the WG ad hoc process for human health, for a ready-to-use spraying product.. For the disinfection of animal housing the applicant has only considered the following scenarios: sows in groups, fattening pigs and broilers in free range. For another product type 3 active substance the Swedish CA showed the scenario "veal cattle" represents the worst case with respect to grassland and the scenario "ducks" with respect to arable land. Furthermore the scenario for "turkey" is worst case with respect to emissions to STP. Consequently, the current exposure assessment only covers use in animal housing containing sows in groups, fattening pigs and broilers in free range. Additional uses would need to be evaluated at product authorisation stage.

Aggregate Exposure

The applicant proposes to use Ampholyt as a veterinary hygiene product (PT3) to disinfect:

- Animal housing
- Vehicles used to transport mammals/poultry
- Footwear associated with animal housing
- Animal feet entering/leaving milking parlours.

Several uses can result in exposure to the same environmental compartment. Consequently, it is necessary to consider whether these emissions can overlap in time and space and if an aggregate exposure assessment is warranted. The disinfection of animal vehicles (365 emission days) and footwear of personnel working in animal houses (365 emission days) release emissions on a daily basis and may end up in the same STP. These emissions could overlap in time and space. On some occasions the daily emission rate could be enhanced due to i). emissions from the disinfection of animal hooves/feet which occurs on a weekly basis and ii)occasionally from emissions from animal housing associated with broilers. As a worst case scenario an aggregate assessment was performed for all uses within PT3 for which authorisation is sought that make emissions to the STP via the facility drain by the summation of local concentrations of all single uses.

Three of the four proposed PT3 uses can make emissions to the environment via the slurry + manure → soil → groundwater → surface water. The disinfection of footwear of personnel working in animal houses and the disinfection of the animal housing could make emissions to the same manure storage facility. These emissions could potentially overlap in time and space as i). the emissions associated with the disinfection of footwear occur on a daily basis over a period of 365 d and ii). the manure/slurry is stored for the same period of time in each scenario. Consequently, aggregate exposure could occur. To assess the degree of aggregate exposure, the amount of active ingredient in the manure or slurry after the relevant number of biocide applications for the manure/slurry application for both scenarios was added. In accordance with the ESD for PT3 for the calculation of the concentration in soil, the amount of biocide present in the manure was related to the nitrogen content and the nitrogen load, allowed under the nitrogen immission standard. Disinfection of animals' hooves is generally associated with cows entering or leaving a milking parlour. The applicant has only assessed exposure arising from the disinfection of stables associated with poultry and pigs. In this case emissions to manure/slurry are not considered to overlap in time and space as both scenarios are likely to occur on separate farms. Consequently, this route of exposure was not considered in the aggregate exposure assessment. However, emissions from this route of exposure may need to be considered at product authorisation if it is proposed to use the product in a different type of animal housing e.g. dairy cows. In the latter case emissions could potentially overlap.

2.2.2.5 Risk Characterisation

Appropriate risk assessments were carried out based on two exposure scenarios in an attempt to address the different distribution potentials of the Ampholyt constituents in the environment. These are delineated by applying Koc's of both 10^6 L/kg and 15,431.8 L/kg.

Product Type 3

PT 3.1: Disinfection of animal housing

The disinfection of animal housing with no direct emissions to STP can be considered to be acceptable in the case of housing for sows and fattening pigs. The risk quotients for the soil (arable, grassland), surface water and sediment compartments following application of slurry/manure from housing for sows and fattening pigs are acceptable.

The use of Ampholyt in the disinfection of housing for broilers poses a risk to the surface water and sediment as a result of emission of waste via the STP. Reliable and enforceable risk mitigation measures to contain this risk would need to be in place to support safe use. This could possibly be attained by directing the emission to slurry/manure rather than to the STP. The slurry and manure would be subsequently directed to soil where the risk quotients for the soil (arable, grassland), surface water and sediment compartments following application of slurry/manure from broilers are considered acceptable.

Acceptable risks for Ampholyt in the disinfection of animal housing for sows and fattening pigs were identified. Where emissions from broilers are directed to manure storage or slurry tanks use in broiler housing may be permitted. At product authorisation any uses outside of these safe uses must be assessed for risk to the environment.

Disinfection of animal housing - sows in groups

Compartment	Unit	PEC (Koc 15,43	PEC (Koc 10^6	PNEC (Koc	PNEC (Koc 10^6 L/kg)	PEC/ PNEC	PEC/ PNEC

		1.8 L/kg)	L/kg)	15,431.8 L/kg)		(Koc 15,431.8 L/kg)	(Koc 10^6 L/kg)
Slurry/manure application							
Grassland							
PIEC Soil (grassland; N)	[mg/kg $_{wwt}$]	3.00E-02		0.57	0.57	5.26E-02	5.26E-02
PECsoil (4 applications/yr)	[mg/kg $_{wwt}$]	8.03E-02		0.57	0.57	1.41E-01	1.41E-01
PECsoil after 10 yrs of slurry/manure application	[mg/kg $_{wwt}$]	9.14E-02		0.57	0.57	1.6E-01	1.6E-01
PECsoil after 10 yrs of slurry/manure application(TWA)	mg/kg$_{wwt}$]	8.39E-02		0.57	0.57	1.47E-01	1.47E-01
Groundwater (grassland; N)	[mg/L]	2.09E-04	3.22 E-06				
Surface water (grassland; N)	[mg/L]	2.04E-05	1.29 E-07	2.30E-04	2.30E-04	8.87E-02	5.61E-04
Sediment (grassland; N)	[mg/kg wwt]	6.87E-03	2.80 E-03	0.00774	0.5	8.88E-01	5.60E-03
Arable land							
PIEC Soil (arable land; N)	[mg/kg $_{wwt}$]	2.25E-02		0.57	0.57	3.95E-02	3.95E-02
PEC soil (1 app/yr)	[mg/kg $_{wwt}$]	2.25E-02		0.57	0.57	3.95E-02	3.95E-02
PECsoil after 10 yrs of slurry/manure application	[mg/kg $_{wwt}$]	2.56E-02		0.57	0.57	4.49E-02	4.49E-02
PECsoil after 10 yrs of slurry/manure application (TWA)	[mg/kg $_{wwt}$]	2.35E-02		0.57	0.57	4.12E-02	4.12E-02
Groundwater (arable land; N)	[mg/L]	5.85E-05	9.02 E-07				
Surface water (arable land; N)	[mg/L]	5.71E-06	3.61 E-08	2.30E-04	2.30E-04	2.48E-02	1.57E-04
Sediment (arable; N)	[mg/kg wwt]	1.92E-03	7.85 E-04	0.00774	0.5	2.48E-01	1.57E-03

For veterinary hygiene products used in stables, the entry pathway into the environment is: application in stables → slurry + manure → soil → groundwater → surface water
STP is not a relevant route of exposure

Risk quotients for the use of Ampholyt 20 for disinfection of animal housing – fattening pigs

Compartment	Unit	PEC (Koc 15,431.8 L/kg)	PEC (Koc 10^6 L/kg)	PNEC (Koc 15,431.8 L/kg)	PNEC (Koc 10^6 L/kg)	PEC/ PNEC (Koc 15,431.8 L/kg)	PEC/ PNEC (Koc 10^6 L/kg)
Slurry/manure application							
Grassland							

PIEC Soil (grassland; N)	[mg/kg$_{wwt}$]	2.20E-02		0.57	0.57	3.86E-02	3.86E-02
PECsoil (4 applications/yr)	[mg/kg$_{wwt}$]	5.89E-02		0.57	0.57	1.03E-01	1.03E-01
PECsoil after 10 yrs of slurry/manure application	[mg/kg$_{wwt}$]	6.71E-02		0.57	0.57	1.63E-01	1.63E-01
PECsoil after 10 yrs of slurry/manure application	[mg/kg$_{wwt}$]	6.16E-02		0.57	0.57	1.08E-01	1.08E-01
Groundwater (grassland; N)	[mg/L]	1.53E-04	2.36E-06				
Surface water (grassland; N)	[mg/L]	1.50E-05	9.45E-08	2.30E-04	2.30E-04	6.52E-02	4.11E-04
Sediment (grassland; N)	[mg/kg wwt]	5.03E-03	2.05E-03	0.00774	0.5	6.50E-01	4.10E-03
Arable							
PIEC Soil (arable land; N)	[mg/kg$_{wwt}$]	1.10E-02		0.57	0.57	1.93E-02	1.93E-02
PEC soil (1 app/yr)		1.10E-02		0.57	0.57	1.93E-02	1.93E-02
PECsoil after 10 yrs of slurry/manure application	[mg/kg$_{wwt}$]	1.25E-02		0.57	0.57	2.19E-02	2.19E-02
PECsoil after 10 yrs of slurry/manure application (TWA)	[mg/kg$_{wwt}$]	1.15E-02		0.57	0.57	2.02E-02	2.02E-02
Groundwater (arable land; N)	[mg/L]	2.86E-05	4.41E-07				
Surface water (arable land; N)	[mg/L]	2.79E-06	1.76E-08	2.30E-04	2.30E-04	1.21E-02	1.45E-06
Sediment (arable; N)	[mg/kg wwt]	9.40E-04	3.84E-04	0.00774	0.5	1.21E-01	7.68E-03

For veterinary hygiene products used in stables, the entry pathway into the environment is: application in stables → slurry + manure → soil → groundwater → surface water
STP is not a relevant route of exposure

Risk quotients for the use of Ampholyt for disinfection of animal housing – broilers in free range with litter floor

Compartment	Unit	PEC (Koc 15,431.8 L/kg)	PEC (Koc 10^6 L/kg)	PNEC (Koc 15,431.8 L/kg)	PNEC (Koc 10^6 L/kg)	PEC/ PNEC (Koc 15,431.8 L/kg)	PEC/PNEC (Koc 10^6 L/kg)
STP emission#							

STP	[mg/L]	0.0111	5.40E-03	0.22	0.22	0.05	2.45E-02
Surface water	[mg/L]	1.08E-03	2.16E-04	2.30E-04	2.30E-04	**4.70**	9.39E-01
Sediment	[mg/kg_{wwt}]	0.365	4.7	0.00774	0.5	**47.16**	**9.40E+00**
Soil	[mg/kg_{wwt}]	0.134	0.228	0.57	0.57	0.24	4.00E-01
Groundwater	[mg/L]	3.33E-04	8.77E-06				
Slurry/manure application							
Grassland*							
PIEC Soil (grassland; N)	[mg/kg_{wwt}]	5.88E-03		0.57	0.57	1.03E-02	1.03E-02
PECsoil (4 applications/yr)	[mg/kg_{wwt}]	1.57E-02		0.57	0.57	2.75E-02	2.75E-02
PECsoil after 10 yrs of slurry/manure application	[mg/kg_{wwt}]	1.79E-02		0.57	0.57	3.14E-02	3.14E-02
PECsoil after 10 yrs of slurry/manure application	[mg/kg_{wwt}]	1.65E-02		0.57	0.57	2.89E-02	2.89E-02
Groundwater (grassland; N)	[mg/L]	4.09E-05	6.31E-07				
Surface water (grassland; N)	[mg/L]	4.00E-06	2.53E-08	2.30E-04	2.30E-04	1.74E-02	1.10E-04
Sediment (grassland; N)	[mg/kg wwt]	1.35E-03	5.49E-04	0.00774	0.5	1.74E-01	1.10E-03
Arable							
PIEC Soil (arable land; N)	[mg/kg_{wwt}]	7.35E-03		0.57	0.57	1.29E-02	1.29E-02
PEC soil (1 app/yr)	[mg/kg_{wwt}]	7.35E-03		0.57	0.57	1.29E-02	1.29E-02
PECsoil after 10 yrs of slurry/manure application	[mg/kg_{wwt}]	8.37E-03		0.57	0.57	1.47E-02	1.47E-02
PECsoil after 10 yrs of slurry/manure application	[mg/kg_{wwt}]	7.68E-03		0.57	0.57	1.35E-02	1.35E-02
Groundwater (arable land; N)	[mg/L]	1.91E-05	2.95E-07				
Surface water (arable land; N)	[mg/L]	1.87E-06	1.18E-08	2.30E-04	2.30E-04	8.13E-03	5.13E-05
Sediment (arable; N)	[mg/kg wwt]	6.28E-04	2.56E-04	0.00774	0.5	8.11E-02	5.12E-04

[#]For veterinary biocidal products used in stables, the entry pathway into to environment is: application in stables → slurry + manure → soil → groundwater → surface water
Please refer to the previous table for an outline of how the PECs were calculated.

* Indirect emissions of biocidal active substances into the environment via the sewage system after use. These PECs were calculated using EUSES 2.1.2

PT 3.2: Disinfection of vehicles used for animal transport

The risk assessment for Ampholyt use for disinfection for animal transport, mammals and poulty, indicated a risk to the environment at both Tiers of assessment. In the absence of risk mitigation measures the current proposed use of Ampholyt for the disinfection of vehicles used in animal transport poses a risk to the environment as assessed at both Tiers.

Disinfection of vehicles used for animal transport-mammal transport

Environmental Compartment	PNEC	Koc 15,431.8 L/kg		Koc 10^6 L/kg	
		PEC	PEC/PNEC	PEC	PEC/PNEC
STP	0.22	0.14	0.64	0.0681	0.31
Surface water	2.30E-04	0.0137	**59.57**	2.73E-03	**11.87**
Sediment	0.5/0.00774*	4.6	**594.32**	59.2	**118.40**
Soil	0.57	1.69	**2.96**	2.88	**5.05**
Groundwater	---	4.2E-03	**---**	1.11E-04	**---**

Disinfection of vehicles used for animal transport-poultry transport

Environmental Compartment	PNEC	Koc 15,431.8 L/kg		Koc 10^6 L/kg	
		PEC	PEC/PNEC	PEC	PEC/PNEC
STP	0.22	0.137	0.62	0.0669	0.30
Surface water	2.30E-04	0.0134	**58.26**	2.68E-3	**11.65**
Sediment	0.5/0.00774*	4.52	**583.98**	58.2	**116.40**
Soil	0.57	1.66	**2.91**	2.83	**4.96**
Groundwater	---	4.13E-03	**---**	1.09E-04	**---**

PT 3.4: Disinfection for veterinary hygiene: footbaths (for humans)

The disinfection of footwear where there are no direct emissions to STP can be considered acceptable in the case of footbaths used when entering housing for fattening pigs, sows and broilers. The risk quotients for sediment following indirect release to the environment via slurry/manure for fattening pigs and sows are marginally higher that the trigger value of 1. However, considering the conservative approach to the derivation of both the PNEC and PECs (see Doc IIA and IIB for details) for the risk assessment the CA concludes that this risk is acceptable.

The use of Ampholyt for footwear disinfection in housing for broilers, sows and fattening pigs poses a risk to sediment as a result of emission of waste water via the STP. Reliable and enforceable risk mitigation measures to contain this risk would need to be in place to support safe use. This could possibly be attained by directing the emission to slurry/manure rather than to the STP. The slurry and manure would be subsequently directed to soil where the risk quotients for the soil (arable, grassland), surface water and sediment compartments following application of slurry/manure from broilers are considered acceptable.

Acceptable risks for Ampholyt in the disinfection of footwear using footbaths for humans in animal housing for sows and fattening pigs were identified. Where emissions from broilers, sows and fattening pigs are directed to manure storage or slurry tanks use for disinfection of footwear in these situations may be permitted. At product authorisation any uses outside of these safe uses must be assessed for risk to the environment.

Risk quotients for the use of Ampholyt for veterinary hygiene – footwear: sows in groups

Compartment	Unit	PEC (Koc 15,431.8 L/kg)	PEC (Koc 10^6 L/kg)	PNEC (Koc 15,431.8 L/kg)	PNEC (Koc 10^6 L/kg)	PEC/PNEC (Koc 15,431.8 L/kg)	PEC/PNEC (Koc 10^6 L/kg)

STP emission#							
STP	[mg/L]	1.71E-03	8.31E-4	0.22	0.22	7.78E-03	3.78E-03
Surface water	[mg/L]	1.67E-04	3.32E-5	2.30E-04	2.30E-04	7.26E-01	1.44E-01
Sediment	[mg/kg_{wwt}]	0.0561	0.723	0.00774	0.5	**7.25**	**1.45**
Soil	[mg/kg_{wwt}]	0.0206	3.51E-02	0.57	0.57	3.61E-02	6.16E-02
Groundwater	[mg/L]	5.12E-05	1.35E-06				
Slurry/manure application							
Grassland							
PIEC Soil (grassland; N)	[mg/kg_{wwt}]	1.07E-02		0.57	0.57	1.88E-01	1.88E-01
PECsoil (4 applications/yr)	[mg/kg_{wwt}]	2.87E-01		0.57	0.57	5.04E-01	5.04E-01
PEC soil 10 yr of slurry/manure application	[mg/kg_{wwt}]	3.26E-01		0.57	0.57	5.72E-01	5.72E-01
PEC soil 10 yr of slurry/manure application (TWA)	[mg/kg_{wwt}]	2.99E-01		0.57	0.57	5.25E-01	5.25E-01
Groundwater (grassland; N)	[mg/L]	7.44E-04	1.15E-05				
Surface water (grassland; N)	[mg/L]	7.28E-05	4.60E-07	2.30E-04	2.30E-04	3.17E-01	2.00E-03
Sediment (grassland; N)	[mg/kg wwt]	2.45E-02	9.99E-03	0.00774	0.5	**3.17E+00**	2.00E-02
Arable							
PIEC Soil (arable land; N)	[mg/kg_{wwt}]	1.07E-01		0.57	0.57	1.88E-01	1.88E-01
PECsoil (1 applications/year)	[mg/kg_{wwt}]	1.07E-01		0.57	0.57	1.88E-01	1.88E-01
PEC soil 10 yr of slurry/manure application	[mg/kg_{wwt}]	1.22E-02		0.57	0.57	2.14E-02	2.14E-02
PEC soil 10 yr of slurry/manure application (TWA)	[mg/kg_{wwt}]	1.12E-01		0.57	0.57	1.96E-01	1.96E-01
Groundwater (arable land; N)	[mg/L]	2.78E-04	4.29E-06				
Surface water (arable land; N)	[mg/L]	2.72E-05	1.72E-07	2.30E-04	2.30E-04	1.18E-01	7.48E-04
Sediment (arable; N)		9.15E-03	3.73E-03	0.00774	0.5	**1.18E+00**	7.46E-03

Risk quotients for the use of Ampholyt for veterinary hygiene – footwear: fattening pigs

Compartment	Unit	PEC (Koc 15,431.8 L/kg)	PEC (Koc 10^6 L/kg)	PNEC (Koc 15,431.8 L/kg)	PNEC (Koc 10^6 L/kg)	PEC/PNEC (Koc 15,431.8 L/kg)	PEC/PNEC (Koc 10^6 L/kg)

STP emission#							
STP	[mg/L]	1.71E-03	8.31E-4	0.22	0.22	7.78E-03	3.78E-03
Surface water	[mg/L]	1.67E-04	3.32E-5	2.30E-04	2.30E-04	7.26E-01	1.44E-01
Sediment	[mg/kg_{wwt}]	0.0561	0.723	0.00774	0.5	**7.25**	**1.45**
Soil	[mg/kg_{wwt}]	0.0206	3.51E-02	0.57	0.57	3.61E-02	6.16E-02
Groundwater	[mg/L]	5.12E-05	1.35E-06				
Slurry/manure application							
Grassland							
PIEC Soil (grassland; N)	[mg/kg_{wwt}]	8.22E-02		0.57	0.57	1.44E-01	1.44E-01
PECsoil (4 applications/yr)	[mg/kg_{wwt}]	2.20E-01		0.57	0.57	3.86E-01	3.86E-01
PEC soil 10 yr of slurry/manure application	[mg/kg_{wwt}]	2.51E-01		0.57	0.57	4.40E-01	4.40E-01
PEC soil 10 yr of slurry/manure application (TWA)	[mg/kg_{wwt}]	2.30E-01		0.57	0.57	4.04E-01	4.04E-01
Groundwater (grassland; N)	[mg/L]	5.72E-04	8.83E-06				
Surface water (grassland; N)	[mg/L]	5.59E-05	3.53E-07	2.30E-04	2.30E-04	2.43E-01	1.53E-03
Sediment (grassland; N)	[mg/kg wwt]	1.88E-02	7.68E-03	0.00774	0.5	**2.43E+00**	1.54E-02
Arable							
PIEC Soil (arable land; N)	[mg/kg_{wwt}]	8.22E-02		0.57	0.57	1.44E-01	1.44E-01
PECsoil (1 applications/year)	[mg/kg_{wwt}]	8.22E-02		0.57	0.57	1.44E-01	1.44E-01
PEC soil 10 yr of slurry/manure application	[mg/kg_{wwt}]	9.36E-02		0.57	0.57	1.64E-01	1.64E-01
PEC soil 10 yr of slurry/manure application (TWA)	[mg/kg_{wwt}]	8.59E-02		0.57	0.57	1.51E-01	1.51E-01
Groundwater (arable land; N)	[mg/L]	2.14E-04	3.30E-06				
Surface water (arable land; N)	[mg/L]	2.09E-05	1.32E-07	2.30E-04	2.30E-04	9.09E-02	5.74E-04
Sediment (arable; N)		7.03E-03	2.87E-03	0.00774	0.5	9.08E-01	5.74E-03

Risk quotients for the use of Ampholyt for veterinary hygiene – footwear: broilers in free range with litter floor

Compartment	Unit	PEC	PEC (Koc 10^6 L/kg)	PNEC (Koc	PNEC (Koc 10^6 L/kg)	PEC/PNEC (Koc	PEC/PNEC (Koc 10^6 L/kg)

		(Koc 15,431.8 L/kg)		**15,431.8 L/kg)**		**15,431.8 L/kg)**	
STP emission							
STP		1.71E-03	8.31E-04	0.22	0.22	7.77E-03	3.78E-03
Surface water		1.67E-04	3.32E-05	2.30E-04	2.30E-04	7.26E-01	1.44E-01
Sediment		0.0561	0.723	0.00774	0.5	**7.25E+00**	**1.45E+00**
Soil		0.0206	3.51E-02	0.57	0.57	3.61E-02	6.16E-02
Groundwater		5.12E-05	1.35E-06				
Slurry/manure application							
Grassland							
PIEC Soil (grassland; N)	[mg/kg$_{wwt}$]	3.21E-02		0.57	0.57	5.63E-02	5.63E-02
PECsoil (4 applications/yr)	[mg/kg$_{wwt}$]	8.60E-02		0.57	0.57	1.51E-01	1.51E-01
PEC soil 10 yr of slurry/manure application	[mg/kg$_{wwt}$]						
PEC soil 10 yr of slurry/manure application (TWA)	[mg/kg$_{wwt}$]	8.98E-02		0.57	0.57	1.58E-01	1.58E-01
Groundwater (grassland; N)	[mg/L]	2.23E-04	3.45E-06				
Surface water (grassland; N)	[mg/L]	2.18E-05	1.38E-07	2.30E-04	2.30E-04	9.48E-02	6.00E-04
Sediment (grassland; N)	[mg/kg wwt]	7.35E-03	3.00E-03	0.00774	0.5	9.50E-01	6.00E-03
Arable							
PIEC Soil (arable land; N)	[mg/kg$_{wwt}$]	3.21E-02		0.57	0.57	5.63E-02	5.63E-02
PEC soil (1 app/yr)	[mg/kg$_{wwt}$]	3.21E-02		0.57	0.57	5.63E-02	5.63E-02
PEC soil 10 yr of slurry/manure application	[mg/kg$_{wwt}$]	3.65E-02		0.57	0.57	6.40E-02	6.40E-02
PEC soil 10 yr of slurry/manure application (TWA)	[mg/kg$_{wwt}$]	3.35E-02		0.57	0.57	5.88E-02	5.88E-02
Groundwater (arable land; N)	[mg/L]	8.34E-05	1.29E-06				
Surface water (arable land; N)	[mg/L]	8.15E-06	5.15E-08	2.30E-04	2.30E-04	3.54E-02	2.24E-04
Sediment (arable; N)	[mg/kg wwt]	2.74E-03	1.12E-03	0.00774	0.5	3.54E-01	2.24E-03

PT 3.4: Disinfection for veterinary hygiene: animals' hooves (dairy cows)

A risk has been identified for the disinfection of animals' feet hooves. If release to STP can be prevented and the waste be directed to the slurry/manure for disposal the risk to the

environment is reduced. The risk quotients for soil (grassland) and sediment following indirect release to the environment via slurry/manure exceed the trigger value of 1. However, considering the highly conservative approach to the derivation of both the PNEC and PECs (see Doc IIA and IIB for details) for the risk assessment the eCA concludes that this risk is acceptable.

Veterinary hygiene – animals': hooves (dairy cows)

Compartment	Unit	PEC (Koc 15,431.8 L/kg)	PEC (Koc 10^6 L/kg)	PNEC (Koc 15,431.8 L/kg)	PNEC (Koc 10^6 L/kg)	PEC/PNEC (Koc 15,431.8 L/kg)	PEC/PNEC (Koc 10^6 L/kg)
STP emission							
STP*	[mg/L]	0.207	0.101	0.22	0.22	9.41E-01	4.59E-01
Surface water*	[mg/L]	0.0203	4.04E-03	2.30E-04	2.30E-04	**8.83E+01**	**1.76E+01**
Sediment*	[mg/kg_{wwt}]	6.81	87.8	0.00774	0.5	**8.80E+02**	**1.76E+02**
Soil*	[mg/kg_{wwt}]	2.51	4.27	0.57	0.57	**4.40E+00**	**7.49E+00**
Groundwater*	[mg/L]	6.23E-03	1.64E-04				
Slurry/manure application							
Grassland							
PIEC Soil (grassland; N)	[mg/kg_{wwt}]	5.41E-01		0.57	0.57	9.49E-01	9.49E-01
PECsoil (4 applications)	[mg/kg_{wwt}]	1.45E+00		0.57	0.57	**2.54E+00**	**2.54E+00**
PEC soil 10 yr TWA	[mg/kg_{wwt}]	1.51E+00		0.57	0.57	**2.65E+00**	**2.65E+00**
Groundwater (grassland; N)	[mg/L]	3.76E-03	5.81E-05				
Surface water (grassland; N)	[mg/L]	3.68E-04	2.32E-06	2.30E-04	2.30E-04	**1.60E+00**	1.01E-02
Sediment (grassland; N)	[mg/kg wwt]	1.24E-01	5.05E-02	0.00774	0.5	**1.60E+01**	1.01E-01
Arable							
PIEC Soil (arable land; N)	[mg/kg_{wwt}]	5.07E-01		0.57	0.57	8.89E-01	8.89E-01
PEC soil (1 app/yr)	[mg/kg_{wwt}]	5.07E-01		0.57	0.57	8.89E-01	8.89E-01
PEC soil 10 yr TWA	[mg/kg_{wwt}]	5.30E-01		0.57	0.57	9.30E-01	9.30E-01
Groundwater (arable land; N)	[mg/L]	1.32E-03	2.03E-05				
Surface water (arable land; N)	[mg/L]	1.29E-04	8.13E-07	2.30E-04	2.30E-04	5.61E-01	3.53E-03
Sediment (arable; N)		4.33E-02	1.77E-02	0.00774	0.5	**5.59E+00**	3.54E-02

Summary conclusion

The risk assessment and characterisation for the environment identified an acceptable use for products containing Ampholyt for the footbath (for use with footwear) and animal housing scenarios only where emissions were directed to slurry/manure containers for subsequent spreading to the land.

Aggregate Exposure

Two aggregate exposure assessments were conducted for PT 3 use of Ampholyt. Assessment looked at aggregated emission to STP, and the other for indirect emission via slurry/manure.

However, the need to discuss the aggregated exposure arising when an active substance is approved across multiple product types has been discussed during the substance peer review process. As such, it was proposed that an assessment would be required for Ampholyt. However, in the absence of robust guidance that would allow a quantitative assessment to be carried out, it was agreed that a qualitative analysis would be conducted.

To summarise there are possible situations where aggregated exposure relating to the environment may be expected:

Overlap in time and space is possible in usage of products within the same PT at the same facility and may also occur for the PT2, 3 and 4 products where discharges to a municipal STP result in a loading to the different environmental compartments (SW, soil, sludge, sed and GW). However, where risk mitigation measures are in place for any scenario the PECs for that scenario reduce to zero and the aggregate exposure would alter appropriately. Specifically, in relation to the aggregate quantitative exposure assessment which will need to be updated/changed when guidance is agreed. Please note the following provisional information:

Aggregate exposure to environmental compartments from emissions to a STP

The aggregate exposure assessment indicated a risk to the environment due to direct emission to the STP. Therefore risk mitigation measures need to be considered and implemented to reduce this risk. In the case where a mitigation measure can be in place the related PECs would be reduced to zero. The tables are therefore presented to give an overall picture of the outcome whereby no mitigation measures are in place.

Aggregate exposure to environmental compartments from emissions from slurry/manure

In some cases the risk quotients for the sediment compartment marginally exceed the trigger value of 1. The highest value obtained was 4.07. Considering the highly conservative approach to the derivation of the PNEC and PECs (see IIA and IIB for details) for the risk assessment in this compartment the CA concludes that this risk is acceptable.

Risk of secondary poisoning

Ampholyt is not considered to be of concern with regard to secondary poisoning in either the aquatic or terrestrial food chain.

Whilst no studies were submitted investigating the potential hazards of Ampholyt on non-target aquatic/terrestrial organisms via secondary poisoning, Ampholyt has been shown to be both highly water-soluble and readily biodegradable failing the 10-d window in the environment. In addition, Log D values were calculated using a number of software models for the most hydrophobic component of Ampholyt (C16-PDA) (see Document IIA, Section 4.1.2) to determine its potential to bioaccumulate (i.e. lipophilicity). These results showed a maximum Log D value of 4.4 at pH 9, indicating that Ampholyt does not bioaccumulate (criteria agreed at WG III 2014 was Log D ≥ 4.5 at pH 4-9). Additionally, further supporting information provided in the weight of evidence approach including BCF model estimation, environmental degradation and mammalian metabolic pathways support the conclusion that secondary poisoning is not of concern for Ampholyt.

2.2.3 List of Endpoints

In order to facilitate the work of Member States in granting or reviewing authorisations the most important endpoints, as identified during the evaluation process, are listed in Appendix I.

2.2.4 Conclusions and Decision of the Assessment Report

Further data may be required, in particular regarding the physical and chemical properties and efficacy, to support products and should be provided by applicants at the product authorisation stage.

The outcome of the assessment for Ampholyt in product-type 3 is specified in the BPC opinion following discussions at the tenth meeting of the Biocidal Products Committee (BPC). The BPC opinion was adopted for Ampholyt PT3 on 15 April 2015 and is available from the ECHA web-site.

APPENDIX I: LIST OF ENDPOINTS

CHAPTER 1: IDENTITY, PHYSICAL AND CHEMICAL PROPERTIES, CLASSIFICATION AND LABELLING

Active substance (ISO Common Name)	Amines, N-C_{10}–C_{16}-alkyltrimethylenedi-, reaction products with chloroacetic acid
Product-type	PT3 – Veterinary hygiene

Identity

Chemical name (IUPAC)	Amines, N-C_{10}–C_{16}-alkyltrimethylenedi-, reaction products with chloroacetic acid
Chemical name (CA)	Amines, N-C_{10}–C_{16}-alkyltrimethylenedi-, reaction products with chloroacetic acid
CAS No.	=139734-65-9
EC No.	None
Other substance No.	None
Minimum purity of the active substance as manufactured (g/kg or g/l)	TC - dry material – hypothetical: Minimum specification of 100% w/w for total active ingredient content. TK – aqueous solution – technical material as manufactured: Specification range of 16 – 22% w/w (average of 19 % w/w) for total active ingredient content. The concentration ranges of the individual components making up Amoholyt 20 can be found in the Confidential section of the CAR.
Identity of relevant impurities and additives (substances of concern) in the active substance as manufactured (g/kg)	None.
Molecular formula	Impossible to assign a single molecular formula being a mixture of 20+ chemical species. The relevant molecular formulas for the 24 chemical species are presented below: [redacted] It should be noted that only 16 molecular formulas are presented in the table because some of the 24 chemical species are structural isomers (same molecular formula).
Molecular mass	It is impossible to assign a single molecular mass being a mixture of 24 chemical species.

	The molecular masses for these 24 chemicals range from 185.4 – 414.63 g/mol. A "weighted molecular mass" of 280.79 g/mol can also be proposed taking into account the molecular weight of the 24 components and their average composition in the supporting 7-batch analysis.
Structural formula	It is impossible to assign a single structural formula being a mixture of various chemicals, however the structural formulae of the components making up the active substance are provided below: "Major components":  "Minor constituents" :  Note 2: The above structural formulas cover the species making up the UVCB substance. All of these species are in a thermodynamic equilibrium with their protonated and unprotonated forms. The equilibrium is pH dependent. The pH of the solution in water is approximately around 8 – 8.5 in the presence of acetic acid and hydrochloride which is being released from chloroacetic acid after the reaction with the primary and secondary amines.

Physical and Chemical Properties

Melting point (state purity)	It was not possible to determine a melting point. The active substance decomposed at 140-145 °C (Purity: 99%)
Boiling point (state purity)	It was not possible to determine a boiling point. The active substance decomposed at 140-145 °C (Purity: 99%)
Temperature of decomposition	140-145 °C (Purity: 99%)
Appearance (state purity)	Technical material: colourless to yellowish liquid (Purity 20 %) 19.3% Purified technical material: a white to light yellow solid (Purity: 99%)
Relative density (state purity)	1.027 at 23°C (Purity: 99%)
Surface tension	27.2 mN/m (20ºC, 1 g/l solution) (Purity: 99%)
Vapour pressure (in Pa, state temperature)	1.9×10^{-4} Pa at 20°C & 4.0×10^{-4} Pa at 25°C (extrapolated from experimental data)
Henry's law constant (Pa m^3 mol^{-1})	It is impossible to assign a single Henry's Law Constant given that the active substance is a mixture of 24 chemical species. The individual Henry's Law Constants for the "major components" range from 1.29×10^{-9} – 1.22×10^{-2} Pa.m3/mol. The individual Henry's Law Constants for the "minor components" range from 8.94 x 10-10 – 3.05×10^{1} Pa.m3/mol. The "minor components" make up <2% of the active ingredient. (QSAR Calculations – EPI Suite – HENRYWIN).
Solubility in water (g/l or mg/l, state temperature)	pH 4: >208 g/l at 20°C (Purity: 99%) pH 7: >208 g/l at 20°C (Purity: 99%) pH 9: >208 g/l at 20°C (Purity: 99%)
Solubility in organic solvents (in g/l or mg/l, state temperature)	40-50 g/L (10oC), 40-50 g/L (20oC) and 160 - 200 g/L (30oC) in Methanol & <10 g/L in p-Xylene , n-Heptane, 1,2-Dichloroethane, Acetone and Ethyl acetate at 10, 20 and 30°C (Purity: 99%)
Stability in organic solvents used in biocidal products including relevant breakdown products	Not applicable - the active substance as manufactured is not delivered in an organic solvent.
Partition coefficient (log P_{OW}) (state temperature)	LogPow values for the 20 "major components" range from 0.33 to 6.71. The weighted LogPow value = 3.82 (EpiSuite-KOWWIN).

	Log P values for the 24 chemical species making up the active substance ranged from 2.83 – 5.73 (QSAR Calculation using Marvin Sketch – ChemAxon, 2012). However, the Log P QSAR calculations do not take into account that the 24 chemical species are ionisable species at environmental pH. Log P QSAR calculations are only appropriate for neutral molecules. Log D is a more applicable endpoint for risk assessment. Log D is PH dependent and is applicable to protonated and unprotonated species. The LogD ranges for the 24 chemical species are provided below: pH 5: Log D = -4.53 to 2.11 pH 7: Log D = -2.37 to 2.33 pH 9: Log D = -3.12 to 3.92
Hydrolytic stability (DT_{50}) (state pH and temperature)	pH 4: Hydrolytically stable at 50 °C pH 7: Hydrolytically stable at 50 °C pH 9: Hydrolytically stable at 50 °C
Dissociation constant	The determination of the dissociation constant is experimentally not feasible. However, the components making up Ampholyt are expected to be ionised in the environmental pH range (4 – 9) based on the presence of acidic (pKa ~ 4) and basic (pKa ~ 10) functional groups in the respective structures.
UV/VIS absorption (max.) (if absorption > 290 nm state ε at wavelength)	Molar absorption coefficient ε (290 nm): pH < 2: 4.4 L mol-1cm-1 pH 5: 2.8 L mol-1cm-1 pH 7: 2.2 L mol-1cm-1 pH 9: 3.0 L mol-1cm-1 pH > 12: 2.8 Lmol-1cm-1 (Purity 99%)
IR Spectral data	The active substance is composed of 24 chemical species. The IR spectrum is representative of the structures of these species. 3228 cm-1: NH2+, NH (broad) 2800-3000 cm-1: CH_2 (stretch) 1586 cm-1: COO- (carboxylate and acetate) 1468 cm-1: CH_2 (deformation) 721 cm-1: CH_2 (chain) (Purity 99%)

NMR Spectral data	The active substance is composed of 24 chemical species. The 1H and 13C NMR spectra are representative of the structure of these species. (Purity 99%)
MS Spectral data	The active substance is composed of 24 chemical species. The Mass spectra are representative of the structures of these species. (Purity 99%)
Photostability (DT_{50}) (aqueous, sunlight, state pH)	Half-life, reaction with OH-radicals = 0.125 d (This is based on a 24 hr time period and a hydroxyl radical concentration of 5 x 10^5 molec/cm^3) See Environmental Section of the CAR for further details
Quantum yield of direct phototransformation in water at Σ > 290 nm	Not applicable
Flammability	Not flammable (Purity 99%). Non-flammable conclusion is also supported by theoretical consideration – the technical material as manufactured contains *ca.* 80% water.
Explosive properties	Not explosive based on theoretical considerations – the functional groups are not considered to be explosive.
Oxidising properties	Not oxidising based on theoretical considerations – the functional groups are not considered to be oxidising.

Classification and Proposed Labelling

With regard to physical/chemical data	Not applicable
With regard to toxicological data	*Reg. 1272/2008* H302: Harmful if swallowed H314: Causes severe skin burns and eye damage H361f Cat 2: Suspected of damaging fertility H373: *STOT RE Cat 1* May cause damage to organs (eyes, mesenteric lymph nodes, male/female genital systems) through prolonged or repeated exposure
With regard to fate and behaviour data	Not applicable
With regard to ecotoxicological data	*Reg. 1272/2008* *Aquatic chronic Cat. 1* H410: Very toxic to aquatic life with long lasting effects EUH401: To avoid risks to human health and the environment, comply with the instructions for use. M-Factors: M = 10 (acute), M = 1 (chronic)

CHAPTER 2: METHODS OF ANALYSIS

Analytical Methods for the Active Substance

Technical active substance (principle of method)	HPLC-CAD method. HPLC-MS/MS used as a confirmatory method. Outstanding issues regarding reference standards and the number of active ingredient components used for method validation.
Impurities in technical active substance (principle of method)	Ampholyt is a UVCB substance and therefore does not contain "impurities" as such.

Analytical Methods for Residues

Soil (principle of method and LOQ)	HPLC-MS/MS method. Residue definition – The residue definition for monitoring includes three lead components:: [redacted] The LOQ = 0.05 mg/kg for each lead component. (critical NO(A)EC in soil = 363 mg/kg soil) The applicant has only used a single ion transition for method validation. The applicant should validate the lead components for an additional ion transition.
Air (principle of method and LOQ)	Not required. Ampholyt exposure in air is not considered to be significant with regards to the method of application and intended use pattern. However, if other methods of application and intended uses are requested in the future, the requirement for a method of analysis for air will have to be revisited.
Water (principle of method and LOQ)	<u>Surface water and sediment:</u> A validated method of analysis is required for surface water and sediment matrices. (the relevant NOEC in surface water = 2.3 µg/L). <u>Drinking water:</u> HPLC-MS/MS method. Residue definition – The residue definition for monitoring includes three lead components: [redacted]

	[redacted]
	The LOQ = 0.1 µg/L for each lead component. The applicant has only used a single ion transition for method validation. The applicant should validate the lead components for an additional ion transition.
Body fluids and tissues (principle of method and LOQ)	*Reg. 1272/2008* The active is classified as Danger (H372). The applicant will need to provide a validated method of analysis for monitoring in body fluids and tissues with an LOQ which allows determination at the critical NOAEL (21.012 mg/kg).
Food/feed of plant origin (principle of method and LOQ for methods for monitoring purposes)	Residue transfer into food and feed of plant origin is unlikely due to risk mitigation measures and the intrinsic properties of the substance, however, the accuracy of this statement cannot be concluded due to the absence of relevant guidance or data to quantitatively confirm this assertion. The applicant has provided a validated HPLC-MS/MS method of analysis for red wine, white wine, beer matrices: The LOQ = 0.01 mg/kg for each lead component. The RMS notes that we do not know the % of the three lead components or their metabolites in food of plant origin after Ampholyt application because the applicant has not provided metabolism studies for food of plant origin. The applicant needs to elaborate further regarding their choice of lead components for monitoring. The applicant will also need to provide an ILV study for the method if it is decided that MRLs are required for food of animal origin.
Food/feed of animal origin (principle of method and LOQ for methods for monitoring purposes)	Residue transfer into food and feed of animal origin is unlikely due to risk mitigation measures and the intrinsic properties of the substance, however, the accuracy of this statement cannot be concluded due to the absence of relevant guidance or data to quantitatively confirm this assertion. The applicant has provided a validated HPLC-MS/MS method of analysis for meat, fat and milk matrices.

	The method has been validated for four Ampholyt components: [redacted]
	The LOQ = 0.01 mg/kg for each component. The applicant has only used a single ion transition for method validation. The applicant should validate the lead components for an additional ion transition if a method of analysis is required. The RMS notes that we do not know the % of the four components or their metabolites in food of animal origin after Ampholyt application because the applicant has not provided metabolism studies for food of animal origin. The applicant needs to elaborate further regarding their choice of lead components for monitoring. The applicant will also need to provide an ILV study for the method if it is decided that MRLs are required for food of animal origin.

CHAPTER 3: IMPACT ON HUMAN HEALTH

Absorption, Distribution, Metabolism and Excretion in Mammals

Rate and extent of oral absorption:	34 % systemic bioavailability after oral administration t_{max}: 6 h
Rate and extent of dermal absorption:	Potentially absorbable dose: From concentrated product: 2 % From in-use dilution: 15 % Values derived from the *in vitro* Human Skin Model Test: Ampholyt, the *in vitro* percutaneous absorption of radiolabelled Ampholyt in the concentrate and a single in-use dilution through human skin).
Distribution:	Widely distributed, highest residues in the residual carcass
Potential for accumulation:	No accumulation Mean terminal elimination half-life ($t_{1/2}$) is in the range of 71 – 77 hours
Rate and extent of excretion:	Rapid, predominately excreted via faeces (60%), expired air (18%), urine (9%)
Metabolism in animals	Oxidation (hydroxylation) of the parent and some other metabolites like dehydrogenated and acetylated compounds in urine and faeces
Toxicologically significant metabolite(s)	None

Acute Toxicity

Rat LD_{50} oral	> 660 mg a.i./kg bw (males), 864 mg a.i./kg bw (females) > 3300 mg/kg bw (males), 4318 mg/kg bw (females) manufactured 20 % aqueous solution
Rat LD_{50} dermal	> 400 mg a.s./kg bw (male and female) > 2000 mg/kg bw (male and female) manufactured 20 % aqueous solution
Rat LC_{50} inhalation	Not required
Skin irritation	Causes severe skin burns (active substance as manufactured, i.e. 20 % aqueous solution)
Eye irritation	Causes serious eye damage (active substance as manufactured, i.e. 20 % aqueous solution)
Skin sensitization (test method used and result)	Not sensitising

Repeated Dose Toxicity

Species/ target / critical effect	<u>90 day Rat</u> Target organs identified at necropsy were liver, kidney, heart, thymus, pancreas, spleen, and uterus. Atrophy of the male and female genital system (cervix, epididymis, ovary, prostrate, uterus and vagina from 15 mg/kg bw/day and atrophy of the germinal epithelium in the testes at 45 mg/kg bw/day). <u>2 year Rat</u>

	Target organs were the mesenteric lymph nodes. Histopathological examinations revealed treatment-related changes in the mesenteric lymph nodes of high dose animals only (12.1 mg/kg bw/d for males and 15.5 mg/kg bw/d for females). These findings comprised aggregates of macrophages often showing signs of degeneration and necrosis, in a few rats accompanied by disturbance of lymph node architecture.
Lowest relevant oral NOAEL / LOAEL	*90-d study, rat:* LOAEL: 5 mg a.i./kg bw/d NOAEL: 2.5 mg a.i./kg bw/d *2-year study with Ampholyt 15 DL, rat:* LOAEL: 3.27 mg a.i./kg bw/d NOAEL: 1.03 mg a.i./kg bw/d
Lowest relevant dermal NOAEL / LOAEL	Not required
Lowest relevant inhalation NOAEL / LOAEL	Not required

Genotoxicity	Not genotoxic *in vitro*

Carcinogenicity

Species/type of tumour	Rat and mouse/no tumours observed. However, it is not clear if carcinogenicity was adequately tested because of the high level of mortality in the mouse study.
Lowest dose with tumours	None

Reproductive Toxicity

Species/ Reproduction target / critical effect	Rat / toxicity to fertility / slightly reduced no. of implantation sites and no. of living pups on day 1
Lowest relevant reproductive NOAEL / LOAEL	NOAEL parental: 6 mg a.i./kg bw/d NOAEL reproductive: 6 mg a.i./kg bw/d
Species/Developmental target / critical effect	Rabbit / teratogenicity / no teratogenic or embryotoxic effects
Developmental toxicity	≥ 30 mg a.i./kg bw/day
Lowest relevant developmental NOAEL / LOAEL	Rat / teratogenicity Based on treatment-related effects observed at the 100- and 250 mg/kg bw/day doses. There were treatment related increases in the incidence of unilateral hydroureter compared with the concurrent controls in conjunction with interparietal and supraoccipital delayed ossification. There was also a slight increase in unilateral renal pelvic cavitation at these two higher doses. These observations occurred in the absence of maternal toxicity in rats.
	NOAEL maternal (rat): 20 mg a.i./kg bw/day NOAEL developmental (rat): 8 mg a.i./kg bw/day NOAEL maternal and developmental (rabbit):10 mg a.i./kg bw/day

Neurotoxicity / Delayed Neurotoxicity

Species/ target/critical effect	Not an organophosphorus compound, which is why acute delayed neurotoxicity studies were not conducted.

Lowest relevant developmental NOAEL / LOAEL	No evidence for neurotoxic potential from other studies

Other Toxicological Studies

Not investigated.

Medical Data

No evidence of any negative impact on the staff handling amphotensides.

Summary

	Value	Study	Safety factor
Non-professional user			
AEL $_{\text{long term}}$	0.0035 mg/kg bw/day	2-years rat (feeding) (NOAEL 1 mg/kg). systemic, considering 34 % oral absorption	100
AEL $_{\text{medium term}}$	0.0085 mg/kg bw/day	90-day oral toxicity study in rats (NOAEL 2.5 mg/kg) systemic, considering 34% oral absorption	100
AEL $_{\text{short term}}$	0.027 mg/kg bw/day	Rat developmental toxicity study (NOAEL 8 mg/kg). systemic, considering 34 % oral absorption	100
ADI (if residues in food or feed)	0.01 mg/kg bw/day	2-years rat (feeding) (NOAEL 1 mg/kg).	100
ARfD (acute reference dose)	0.08 mg/kg bw/day	Rat developmental toxicity study (NOAEL 8 mg/kg).	100
Professional user			
Reference value for inhalation (proposed OEL)	Assumed to be 100%		
Reference value for dermal absorption	2% (concentrate) 15% (dilution) Values derived from *in vitro* Human Skin Model Test: Ampholyt, the *in vitro* percutaneous absorption of radiolabelled Ampholyt in the concentrate and a single in-use dilut on through human skin).		

Product Type 3

Acceptable Exposure Scenarios (including method of calculation)

Professional users	**Low-pressure spraying:** TNsG Spraying Model 1 was used to determine professional operator exposure for mixing and loading, application and post-application of TEGO 2000. Systemic exposure while wearing the prescribed PPE (coveralls and gloves) resulted in an exposure dose of 0.0128 mg/kg bw/day (MOS 66.34) when compared to the $AEL_{\text{medium-term}}$ of 0.0085 mg/kg bw/day. **Footbath:** Foot bath exposure (EUROPOEM II database) resulted in a systemic exposure dose of 0.006

	mg/kg bw/day wearing the prescribed PPE (coveralls and gloves) and a MOS value of 141.8 when compared to the $AEL_{medium\text{-}term}$ of 0.0085 mg/kg bw/day. **Ready-to-use (0.2% a.s) trigger spray:** Consumer Spraying and Dusting Model 2 – hand held trigger spray (TNsG 2002) Tier 2 assessment (coveralls and gloves) returned a total exposure value of 0.00069 mg/kg bw/day (MOS 1237).
Production of active substance	Not evaluated
Formulation of biocidal product	Not evaluated
Intended uses	For use by professional operators as a sprayed or wiped surface treatment disinfectant used in areas in which animals are housed.
Secondary exposure/Indirect exposure	Exposure scenarios have been assessed for indirect oral and dermal exposure to children. Infants crawling on treated floors were expected to receive a systemic exposure of 6.42×10^{-3} mg/kg bw/day (MOS 132)
Non-professional users	The product is for professional use only.

CHAPTER 4: FATE AND BEHAVIOUR IN THE ENVIRONMENT

Route and Rate of Degradation in Water

Hydrolysis of active substance and relevant metabolites (DT_{50}) (state pH and temperature)	Hydrolytically stable (pH 4, 7, 9 at 50 °C)
Photolytic / photo-oxidative degradation of active substance and resulting relevant metabolites	Half-life, reaction with OH-radicals = 0.125 d (This is based on a 24 hr time period and a hydroxyl radical concentration of 5 x 10^5 molec/cm^3)
Readily biodegradable (yes/no)	Yes (failing the 10 d window)
Biodegradation in seawater	Not required
Non-extractable residues	Not required
Distribution in water / sediment systems (active substance)	Not required
Distribution in water / sediment systems (metabolites)	Not required

Route and Rate of Degradation in Soil

Mineralisation (aerobic)	Not applicable
Laboratory studies (range or median, with number of measurements, with regression coefficient)	Not required
Field studies (state location, range or median with number of measurements)	Not required
Anaerobic degradation	Not applicable
Soil photolysis	Not applicable
Non-extractable residues	Not applicable
Relevant metabolites - name and/or code, % of applied a.i. (range and maximum)	Not applicable
Soil accumulation and plateau concentration	Not applicable

Adsorption / Desorption

Ka , Kd

Ka_{oc} , Kd_{oc}

K_d oc (cm^3/g)	K_a	K_{aOC}	K_d	K_{dOC}	1/n (ads)
	□$g^{1-1/n}(cm^3)^{1/n}\ g^{-1}$				
31,645.0	1,407.34	62,031.6	2130.2	97,685	1.1303

Average values

Following discussions at the ECHA WG III meeting 2014 it was agreed to perform the environmental exposure assessment with the lowest measured Koc (15,431.8 cm^3/g) to cover the lower range of Kocs and the highest permitted Koc in EUSES (1

	x 10^6 cm^3/kg) to cover the higher range of Kocs exhibited by some components of Ampholyt .
pH dependence (yes / no) (if yes type of dependence)	Individual components of Ampholyt are expected to exhibit pH dependent adsorption

Fate and Behaviour in Air

Direct photolysis in air	Not applicable
Quantum yield of direct photolysis	Not applicable
Photo-oxidative degradation in air	$t_{½}$ (•OH) = 0.125 hr (model calculation)
Volatilisation	

Monitoring Data, if available

Soil (indicate location and type of study)	Not available
Surface Water (indicate location and type of study)	Not available
Groundwater (indicate location and type of study)	Not available
Air (indicate location and type of study)	Not available

CHAPTER 5: EFFECTS ON NON-TARGET SPECIES

Toxicity Data for Aquatic Species (most sensitive species of each group)

Species	Time-scale	Endpoint	Toxicity
Fish			
Oncorhynchus mykiss	96 h	LC_{50}	0.2074 mg a.s./L (measured)
Oncorhynchus mykiss	28d	NOEC	0.052 mg a.s./L (measured)
Invertebrates			
Daphnia magna	48 h semi-static	LC_{50}	0.0333 mg a.s./L (measured)
Daphnia magna	21d	NOEC	0.0023 mg a.s./L (measured)
Algae			
Pseudokirchneriella subcapitata	72 h	NOE_rC E_rC_{50}	0.00955 mg a.s/L (measured) 0.0237 mg a.s./L (measured)
Micro-organisms			
Activated sludge from municipal STP	3 h	LC_{50}	22 mg a.s/L (nominal)

Effects on Earthworms or other Soil Non-target Organisms

Acute toxicity to…	Earthworms 14 day $LC_{50} \geq 1000$ mg a.s./L (nominal)
Reproductive toxicity to…	Not determined

Effects on Soil Micro-organisms

Nitrogen mineralisation	NOEC 64 mg a.s/kg dwt (nominal)
Carbon mineralisation	NOEC 160-400 mg a.s/kg dwt (nominal)

Effects on Terrestrial Vertebrates

Acute toxicity to mammals	Not determined
Acute toxicity to birds	Not determined
Dietary toxicity to birds	Not determined
Reproductive toxicity to birds	Not determined

Effects on Honeybees

Acute oral toxicity	Not determined
Acute contact toxicity	Not determined

Effects on other Beneficial Arthropods

Acute oral toxicity	Not determined

Acute contact toxicity	Not determined
Acute toxicity to…	Not determined

Effects on Terrestrial Plants

Acute toxicity to:*Triticum aestivum, Sinapis alba, Raphanus sativus, Phaseoulus aureus, Lactuca sativa, Avena sativa*	EC_{50} 363 mg a.s./kg (*Lactuca sativa*) All other species EC_{50} > 1000 mg a.s./kg

Bioconcentration

Bioconcentration factor (BCF)	
Depuration time (DT50) (DT90)	Not determined
Level of metabolites (%) in organisms accounting for > 10 % of residues	Not determined

CHAPTER 6: OTHER ENDPOINTS

Not applicable

APPENDIX II: LIST OF INTENDED USES

Product-type: PT3

Claim of the participant:

Ampholyt is used as a disinfectant, antimicrobial cleaner and detergent sanitizer to prevent the spread of various micro-organisms. The spectrum of antimicrobial activity is focused on the destruction of gram-positive and gram-negative bacteria, yeasts as well as a limited viruicide activity against enveloped viruses and against the non-enveloped adenovirus.

The products, organisms or objects to be protected are mainly humans and animals. Objects to be treated to achieve the protection claims are surfaces, walls, and floors in various areas in veterinary areas. The intended effect is inhibition of growth and viability of pathogens, thus to prevent disease to humans and animals, spread of communicable diseases, to prevent waste of valuable resources by extending the lifetime of articles and products, to maintain the efficiency of industrial processes, to protect consumers from microbial contamination of food and other articles, and to maintain the structural integrity of materials.

Target organisms:

Intended target organisms are:

- Bacteria (gram-positive and gram-negative)
- Yeasts
- Viruses, limited virucide (effective against enveloped viruses) and active against Adenovirus

Concentration:

The concentration of the active substance in the product TEGO 2000 is: Ampholyt at 20% w/w (range 17-21% w/w). Physical state of preparation: Liquid. Formulation type: Aqueous solution

In use concentrations are:

PT 3	0.1–0.2 % active matter as aqueous solution, i.e. 0.5–1 % TEGO 2000 (1:200–1:100) or RTU (0.2% a.s.).

Categories of users:

Professionals

Type of application:

PT 3	Low pressure spraying
	Mopping
	Wiping
	Footbath
	Hand-held RTU trigger spray

APPENDIX III: LIST OF STUDIES

Data protection is claimed by the applicant in accordance with Article 12.1(c) (i) and (ii) of Council Directive 98/8/EC for all study reports marked "Y" in the "Data Protection Claimed" column of the table below. For studies marked Yes(i) data protection is claimed under Article 12.1(c) (i), for studies marked Yes(ii) data protection is claimed under Article 12.1(c) (ii). These claims are based on information from the applicant. It is assumed that the relevant studies are not already protected in any other Member State of the European Union under existing national rules relating to biocidal products. It was however not possible to confirm the accuracy of this information.

Section No / Reference No	Author(s)	Year	Title. Source, Report No. GLP / (Un)Published	Data Protection Claimed (Yes/No)	Owner
A2/01	[redacted]	2006	Processing Ampholyt 20 (Alkylaminopropyl Glycine) Goldschmidt GmbH, Essen, Germany, Not GLP / Unpublished	Y (New/First)	Evonik Industries AG
A2/02	[redacted]	2007	Characterisation of Ampholyt 20 by HPLC-CAD Goldschmidt GmbH, Essen, Germany, Not GLP / Unpublished	Y (New/First)	Evonik Industries AG
A2/03	[redacted]	2006	Amphoteric surfactants as microbicidal substances Goldschmidt GmbH, Essen, Germany, Not GLP / Unpublished	Y (New/First)	Evonik Industries AG
A2/04	[redacted]	2003	Ampholyt 51/27 DL Goldschmidt GmbH, Essen, Germany, Not GLP / Unpublished	Y (New/First)	Evonik Industries AG
A2/05	[redacted]	2006	Processing Ampholyt 51 DL (Alkyl Di (Aminoethyl) Glycine / Alkylaminopropyl Glycine) Goldschmidt GmbH, Essen, Germany, Not GLP / Unpublished	Y (New/First)	Evonik Industries AG
A2/06	[redacted]	2006	Structure Determination of Ampholyt 20 und Ampholyt 51/27 DL Goldschmidt GmbH, Essen, Germany, Not GLP / Unpublished	Y (New/First)	Evonik Industries AG
A3.1.1/01	[redacted]	2005	Ampholyt 20/100 ES64A25143, Melting point A.1 (OECD 102), Boiling point (OECD 103) Siemens AG, Frankfurt, Germany, Report No.: 20050230.01 GLP / Unpublished	Y (New/First)	Evonik Industries AG
A3.1.3/01	[redacted]	2005	Ampholyt 20/100 ES64A25143, Relantive density A.3 (OECD 109) Siemens AG, Frankfurt, Germany, Report No.: 20050230.02 GLP / Unpublished	Y (New/First)	Evonik Industries AG

Section No / Reference No	Author(s)	Year	Title. Source, Report No. GLP / (Un)Published	Data Protection Claimed (Yes/No)	Owner
A3.2/01	[redacted]	2005	Ampholyt 20/100 ES64A25143, Vapour pressunre A.4 (OECD104) Siemens AG, Frankfurt, Germany, Report No.: 20050230.03 GLP / Unpublished	Y (New/First)	Evonik Industries AG
A3.2.1/01	[redacted]	2007	Calculation of Henry's law constant of Ampholyt 20 EBRC Consulting GmbH, Hannover, Germany, Report No.: GOL-070401-01 Not GLP / Unpublished	Y (New/First)	Evonik Industries AG
A3.2.1/02	[redacted]	2007	Model calculation of Henry's law constant of Ampholyt 20 EBRC Consulting GmbH, Hannover, Germany, Report No.: GOL-070606-01 Not GLP / Unpublished	Y (New/First)	Evonik Industries AG
A3.3.1/01	[redacted]	2006	Determination of the appearance of Ampholyt 20/100 EBRC Consulting GmbH, Hannover, Germany, Report No.: DEG-20060922-01 Not GLP / Unpublished	Y (New/First)	Evonik Industries AG
A3.4.1/01	[redacted]	2006	Determination of spectral properties of Ampholyt 20/100 Aqura GmbH, Marl, Germany, Report No.: AN-ASB 0332 GLP / Unpublished	Y (New/First)	Evonik Industries AG
A3.4.4/01	[redacted]	2007	Test report according to DIN EN ISO/IEC 17025 Aqura GmbH, Hanau, Germany, Report No.: A070012787 Not GLP / Unpublished	Y (New/First)	Evonik Industries AG
A3.5/01	[redacted]	2002	Determination of physio-chemical properties of TEGO 2000 Infracor GmbH, Marl, Germany, Report No.: AN-ASB 0198 GLP / Unpublished	Y (New/First)	Evonik Industries AG
A3.7/01	[redacted]	2007	Ampholyt 20/100 – solubility in organic solvents Dr.U.Noack-Laboratorien, Sarstedt, Germany, Report No.: CLS117241 GLP / Unpublished	Y (New/First)	Evonik Industries AG
A3.9/01	[redacted]	1994	The partition coefficient n-octanol/water of TEGO 2000 TNO Prins Maurits Laboratory, Rijswijk, The Netherland, Report No.: 213194726a GLP / Unpublished	Y (New/First)	Evonik Industries AG
A3.9/02	[redacted]	2007	Calculation of partition coefficient of Ampholyt 20 EBRC Consulting GmbH, Hannover, Germany, Report No.: GOL-070524-01 Not GLP / Unpublished	Y (New/First)	Evonik Industries AG

Section No / Reference No	Author(s)	Year	Title. Source, Report No. GLP / (Un)Published	Data Protection Claimed (Yes/No)	Owner
A3.11/01	[redacted]	2005	Ampholyt 20/100 ES64A25143, Flammability (solids) A.10. Auto-flammability A.16 (solids-determination of relative self-ignition temperature) Siemens AG, Frankfurt, Germany, Report No.: 20050230.04 GLP / Unpublished	Y (New/First)	Evonik Industries AG
A3.13/01	[redacted]	2000	Determination of the surface tension of an aqueous solution of TEGO 2000 NOTOX B.V., 's-Hertogenbosch, The Netherlands, Report No.: 283444 GLP / Unpublished	Y (New/First)	Evonik Industries AG
A3.15/01	[redacted]	2007	Explosive properties of Ampholyt 20/100, Expert Statement EBRC Consulting GmbH, Hannover, Germany, Report No.: GOL-070713/01 Not GLP / Unpublished	Y (New/First)	Evonik Industries AG
A3.16/01	[redacted]	2007	Oxidising properties of Ampholyt 20/100, Expert Statement EBRC Consulting GmbH, Hannover, Germany, Report No.: GOL-070713/02 Not GLP / Unpublished	Y (New/First)	Evonik Industries AG
A4.1/01	[redacted]	2000	Determination of the content of microbicidal amphotheric in Tego 2000 NOTOX B.V., 's-Hertogenbosch, The Netherlands, Report No.: 266525 GLP / Unpublished	Y (New/First)	Evonik Industries AG
A4.1/02	[redacted]	2007	Analytical procedure – short description, analytical method for the analysis of Ampholyt 20 by HPLC – CAD Goldschmidt GmbH, Essen, Germany, Not GLP / Unpublished	Y (New/First)	Evonik Industries AG
A4.1/03	[redacted]	2007	Determination of acetic acid in Ampholyt 20 by HPLC-UV Goldschmidt GmbH, Essen, Germany, Not GLP / Unpublished	Y (New/First)	Evonik Industries AG
A4.2/01	[redacted]	2007	Validation for the substance-specific analysis of Ampholyt 20/100 in soil Fraunhofer (IME), Schmallenberg, Germany, Report No.: EBR-013/7-24 GLP / Unpublished	Y (New/First)	Evonik Industries AG
A4.2/02	[redacted]	2007	Validation for the substance-specific analysis of Ampholyt 20/100 in water Fraunhofer (IME), Schmallenberg, Germany, Report No.: EBR-013/7-26 GLP / Unpublished	Y (New/First)	Evonik Industries AG

Section No / Reference No	Author(s)	Year	Title. Source, Report No. GLP / (Un)Published	Data Protection Claimed (Yes/No)	Owner
A4.3/01	[redacted]	1978	Über den Stellenwert amphoterer Desinfektionsmittelspuren in Speisegelatine - Ergebnisse entsprechender rückstandsanalytischer, mikrobiologischer und toxikologischer Untersuchungen Arch. Lebensmittel 29, 62-69, Not GLP / Published	N	
A4.3/02	[redacted]	1993	Adsorptionsvermögen von mikrobiziden Amphotensiden auf harten Oberflächen (with english translation) Goldschmidt GmbH, Essen, Germany, Report No.: THG-AL/022-02-B Not GLP / Unpublished	Y (New/First)	Evonik Industries AG
A4.3/03	[redacted]	1991	Photometric micro-method for the determination of TEGOL 2000 residues in food substrates Goldschmidt GmbH, Essen, Germany, Not GLP / Unpublished	Y (New/First)	Evonik Industries AG
A4.3/04	[redacted]	2010	Ampholyt 20 Components – Analytical Method for the Determination of Residues in Food of Animal Origin, Wine, and Beer Dr. U. Noack-Laboratorien, Sarstedt, Germany Report No.: CRA13322 GLP / Unpublished	Y (New/First)	Evonik Industries AG
A5.1/01	[redacted]	2000	Ampholyt 20 Function (Benefit / Usefullness) Goldschmidt AG, Essen, Germany, Not GLP / Unpublished	Y (New/First)	Evonik Industries AG
A5.4/01	[redacted]		Mechanism of killing microorganisms unspecified, Not GLP / Unpublished	Y (New/First)	Evonik Industries AG
A6.1.1/01	[redacted]	1984	Ni 4648 L (TEGOL 2000) acute oral toxicity study in the rat (LD50) Hazleton Laboratories, Münster, Germany, Report No.: 493-348/66 Not GLP / Unpublished	Y (New/First)	Evonik Industries AG
A6.1.1/02	[redacted]	2003	Acute oral toxicity study with Ampholyt 20/100 in rats TNO, Zeist, The Netherlands, Report No.: V 4410/24 GLP / Unpublished	Y (New/First)	Evonik Industries AG
A6.1.2/01	[redacted]	1988	Test to evaluate the acute toxicity following a single cutaneous application (limit test), in the rat Hazleton Laboratories, Münster, Germany, Report No.: 810350 Not GLP / Unpublished	Y (New/First)	Evonik Industries AG

Section No / Reference No	Author(s)	Year	Title. Source, Report No. GLP / (Un)Published	Data Protection Claimed (Yes/No)	Owner
A6.1.4/01	[redacted]	1990	Acute dermal irritation/corrosion study with TEGOL 2000 in albino rabbits TNO-CIVO Institutes, Zeist, The Netherlands, Report No.: V 90.386/2000061 GLP / Unpublished	Y (New/First)	Evonik Industries AG
A6.1.4/02	[redacted]	2005	Ampholyt 20/100: Primary skin irritation study in rabbits (4-hour semi-occlusive application) RCC Ltd., Füllinsdorf, Switzerland, Report No.: 857588 GLP / Unpublished	Y (New/First)	Evonik Industries AG
A6.1.4/03	[redacted]	2004	In vitro skin corrosion: human skin model test with Ampholyt 20/100 RCC, Rossdorf, Germany, Report No.: 849900 GLP / Unpublished	Y (New/First)	Evonik Industries AG
A6.1.4/04	[redacted]	1988	Test for eye irritation of TEGOL 2000 (conc.) in rabbits IBR, Hannover, Germany, Report No.: 1-3-81-88 Not GLP / Unpublished	Y (New/First)	Evonik Industries AG
A6.1.5/01	[redacted]	2004	Maximisation sensitisation test according to Magnusson & Kligman of "Ampholyt 20/100" in the guinea pig Harlan Bioservice, Walsrode, Germany, Report No.: 10-5-0120-04 GLP / Unpublished	Y (New/First)	Evonik Industries AG
A6.2/01	[redacted]	2007	Qualitative assessment of the toxicokinetic behaviour of Ampholyt 20 EBRC Consulting GmbH, Hannover, Germany, Report No.: DEG-070705-01 Not GLP / Unpublished	Y (New/First)	Evonik Industries AG
A6.2/02	[redacted]	2012	Kinetic and metabolism of ^{14}C-labelled Ampholyt 20 after single repeated intravenous and oral administration to CD® rats. LPT, Hamburg, Germany, Report No.: 23199 GLP / Unpublished	Y (New/First)	Evonik Industries AG
A6.2/03	[redacted]	2008	Ampholyt 20: the in vitro percutaneous absorption of radiolabelled Ampholyt 20 in the concentrate and a single in-use dilution through human skin Charles River Laboratories, Edinburgh, UK, Report No.: 28710 GLP / Unpublished	Y (New/First)	Evonik Industries AG
A6.4.1/01	[redacted]	1988	TEGO 2000/TEGOL 2000 90 day oral (gavage) subchronic toxicity study in the rat Hazleton Laboratories, Münster, Germany, Report No.: 797-348/152 Not GLP / Unpublished	Y (New/First)	Evonik Industries AG

Section No / Reference No	Author(s)	Year	Title. Source, Report No. GLP / (Un)Published	Data Protection Claimed (Yes/No)	Owner
A6.4.1/02	[redacted]	1989	TEGO 2000/TEGOL 2000 90 day oral (gavage) subchronic toxicity study in the rat Hazleton Laboratories, Münster, Germany, Report No.: 868-348-153 Not GLP / Unpublished	Y (New/First)	Evonik Industries AG
A6.4.1/03	[redacted]	2008	Repeated dose 90-day oral toxicity study of Ampholyt 20 in beagle dogs LPT, Hamburg, Germany, Report No.: 21006 GLP / Unpublished	Y (New/First)	Evonik Industries AG
A6.5/01	[redacted]	1977	Chronic (2-year) oral toxicity study with Ampholyt 15DL in rats TNO, Zeist, The Netherlands, Report No.: R 5364 Not GLP / Unpublished	Y (New/First)	Evonik Industries AG
A6.5/02	[redacted]	2011	Combined chronic toxicity and carcinogenicity study of Ampholyt 20 by dietary administration to CD-1 mice LPT, Hamburg, Germany Report No.: 21008 GLP / Unpublished	Y (New/First)	Evonik Industries AG
A6.6.1/01	[redacted]	2003	Bacterial reverse mutation test with Ampholyt 20/100 TNO, Zeist, The Netherlands, Report No.: V 4405/37 GLP / Unpublished	Y (New/First)	Evonik Industries AG
A6.6.1/02	[redacted]	1988	Mutagenicity evaluation of TESOL 2000 in the Ames Salmonella/microsome plate incorporation test L+S AG, Bad Bocklet, Germany, Report No.: 75140 Not GLP / Unpublished	Y (New/First)	Evonik Industries AG
A6.6.2/01	[redacted]	1989	Chromosome analysis of the cultured human lymphocytes following in vitro treatment with TEGOL 2000 TNO-CIVO Institutes, Zeist, The Netherlands, Report No.: V 89.360 GLP / Unpublished	Y (New/First)	Evonik Industries AG
A6.6.3/01	[redacted]	1989	In vitro assay for the induction of point mutations in the HGPRT-locus of Chinese hamster ovary cells by TEGOL 2000 TNO-CIVO Institutes, Zeist, The Netherlands, Report No.: V 89.478 GLP / Unpublished	Y (New/First)	Evonik Industries AG
A6.8.1/01	[redacted]	2001	Embryotoxicity and teratogenicity study with TEGO 2000 administered by oral gavage in female albino NZW rabbits NOTOX B.V., 's-Hertogenbosch, The Netherlands, Report No.: 285716 GLP / Unpublished	Y (New/First)	Evonik Industries AG

Section No / Reference No	Author(s)	Year	Title. Source, Report No. GLP / (Un)Published	Data Protection Claimed (Yes/No)	Owner
A6.8.1/02	[redacted]	2001	Embryotoxicity and teratogenicity study with TEGO 2000 administered by oral gavage in female wistar rats NOTOX B.V., 's-Hertogenbosch, The Netherlands, Report No.: 285872 GLP / Unpublished	Y (New/First)	Evonik Industries AG
A6.8.2/01	[redacted]	2002	Two-generation reproduction toxicity study with TEGO 2000 administered by oral gavage in wistar rats NOTOX B.V., 's-Hertogenbosch, The Netherlands, Report No.: 285637 GLP / Unpublished	Y (New/First)	Evonik Industries AG
A6.12.1/01	[redacted]	2007	Arbeitsmedizinische Betreuung Goldschmidt AG, Essen, Germany, Not GLP / Unpublished	Y (New/First)	Evonik Industries AG
A6.12.7/01	[redacted]	2007	Detoxication of Microbicidal Amphoterics Goldschmidt GmbH, Essen, Germany, Not GLP / Unpublished	Y (New/First)	Evonik Industries AG
A6.15/01	[redacted]	1982	Determination and toxicological significance of adhesive amphotensid residues on disinfected surfaces in the food industry Fleischwirtsch. 62, 880-882, Not GLP / Published	N	
A6.16/01	[redacted]	1997	Rückstände des Desinfektionsmittels Tego 2000 auf ausgewählten Werkstoffoberflächen (with english translation) Fleischwirts. 77, 534-536, Not GLP / Published	N	
A6.16/02	[redacted]	1993	Adsorptionsvermögen von Tego 2000 auf harten Oberflächen (with english translation) Goldschmidt GmbH, Essen, Germany, Not GLP / Unpublished	N	Evonik Industries AG
A6.16/03	[redacted]	1990	Adsorptionsvermögen von Tego 2000 auf harten Oberflächen (with english translation) Goldschmidt GmbH, Essen, Germany, Not GLP / Unpublished	N	Evonik Industries AG
A7.1.1.1.1/01	[redacted]	2001	Statement on the determination of the hydrolysis of Tego 2000 as a function of pH NOTOX B.V., 's-Hertogenbosch, The Netherlands, Report No.: 314652 Not GLP / Unpublished	Y (New/First)	Evonik Industries AG

Section No / Reference No	Author(s)	Year	Title. Source, Report No. GLP / (Un)Published	Data Protection Claimed (Yes/No)	Owner
A7.1.1.1.1/02	[redacted]	2008	Hydrolysis of Ampholyt 20/100 in water according to the OECD-Guideline 111 "Hydrolysis as a function of pH" Fraunhofer (IME), Schmallenberg, Germany, Report No.: EBR-013/7-28 GLP / Unpublished	Y (New/First)	Evonik Industries AG
A7.1.1.1.2/01	[redacted]	2008	Direct phototransformation of Ampholyt 20/100 in water according to the draft OECD-Guideline 111 "Phototransformation of chemicals in water - direct and indirect photolysis", and SETAC procedures Fraunhofer (IME), Schmallenberg, Germany, Report No.: EBR-013/7-05 GLP / Unpublished	Y (New/First)	Evonik Industries AG
A7.1.1.2.1/01	[redacted]	2002	Ampholyt 20/100 – Determination of the biodegradability in the DOC DIE-AWAY test Infracor GmbH, Marl, Germany, Report No.: DDA-179/02 GLP / Unpublished	Y (New/First)	Evonik Industries AG
A7.1.1.2.1/02	[redacted]	2002	TEGO 2000 – Determination of the biodegradability in the DOC DIE-AWAY Test Infracor GmbH, Marl, Germany, Report No.: DDA-163/01 GLP / Unpublished	Y (New/First)	Evonik Industries AG
A7.1.1.2.1/03	[redacted]	1991	The biodegradability of the product TEGO 2000/TEGOL 2000 in a closed bottle test TNO, Delft, The Netherlands, Report No.: R91/221 GLP / Unpublished	Y (New/First)	Evonik Industries AG
A7.1.2.1.1/01	[redacted]	1993	The elimination of TEGO 2000/TEGOL 2000 in a continuous activated sludge system ("coupled units test") TNO, Delft, The Netherlands, Report No.: IMW-R92/327 GLP / Unpublished	Y (New/First)	Evonik Industries AG
A7.1.2.1.1/02	[redacted]	1992	Biologische Abbaubarkeit von TEGOL 2000/TEGO 2000 (with english translation) Bayerische Landesanstalt für Wasserforschung, München, Germany, Not GLP / Unpublished	Y (New/First)	Evonik Industries AG
A7.1.2.1.2/01	[redacted]	2007	Anaerobic biodegrability test, ultimate anaerobic biodegrability of Ampholyt 20 by digested sludge Fraunhofer (IME), Schmallenberg, Germany, Report No.: EBR-013/3-30 GLP / Unpublished	Y (New/First)	Evonik Industries AG

Section No / Reference No	Author(s)	Year	Title. Source, Report No. GLP / (Un)Published	Data Protection Claimed (Yes/No)	Owner
A7.2.3.1/01	[redacted]	2008	Determination of the adsorption/desorption of 14C Ampholyt 20/100 Fraunhofer (IME), Schmallenberg, Germany, Report No.: EBR-013/7-13 GLP / Unpublished	Y (New/First)	Evonik Industries AG
A7.3.1/01	[redacted]	2007	Estimation of the photochemical oxidative degradation rate in the atmosphere of Ampholyt 20 EBRC Consulting GmbH, Hannover, Germany, Report No.: GOL-070713-03 Not GLP / Unpublished	Y (New/First)	Evonik Industries AG
A7.4.1.1/01	[redacted]	2002	Ampholyt 20/100 – determination of the acute toxicity for the fish Cyprinus carpio (Acute Fish Toxicity Test) Infracor GmbH, Marl, Germany, Report No.: FK 1444 GLP / Unpublished	Y (New/First)	Evonik Industries AG
A7.4.1.1/02	[redacted]	1995	Semi-static acute toxicity test with TEGO 2000 and Brachydanio rerio TNO, Delft, The Netherlands, Report No.: IMW-94-0039-02 GLP / Unpublished	Y (New/First)	Evonik Industries AG
A7.4.1.1/03	[redacted]	2008	Oncorhynchus mykiss, acute toxicity test (OECD 203) flow-through exposure - effect of Ampholyt 20 on the acute toxicity to rainbow trout Fraunhofer (IME), Schmallenberg, Germany, Report No.: EBR-013/4-13 GLP / Unpublished	Y (New/First)	Evonik Industries AG
A7.4.1.2/01	[redacted]	2002	Ampholyt 20/100 – determination of the immobilisation of Daphnia magna STRAUS (Acute Immobilisation Test) Infracor GmbH, Marl, Germany, Report No.: DK 795 GLP / Unpublished	Y (New/First)	Evonik Industries AG
A7.4.1.2/02	[redacted]	1995	Static acute toxicity test with TEGO 2000 and Daphnia magna TNO, Delft, The Netherlands, Report No.: IMW-94-0039-01 GLP / Unpublished	Y (New/First)	Evonik Industries AG
A7.4.1.2/03	[redacted]	2007	Daphnia magna, Acute Immobilisation Test (OECD 202) Semi-static exposure. Effect of Ampholyt 20 on the immobilization of Daphnia magna Fraunhofer (IME), Schmallenberg, Germany, Report No.: EBR-013/4-20 GLP / Unpublished	Y (New/First)	Evonik Industries AG

Section No / Reference No	Author(s)	Year	Title. Source, Report No. GLP / (Un)Published	Data Protection Claimed (Yes/No)	Owner
A7.4.1.2/04	[redacted]	2008	Amendment no. 1 to study report Daphnia magna, acute immobilisation test (OECD 202) semi-static exposure. Effect of Ampholyt 20 on the immobilization of Daphnia magna. Recalculation of effect values based on analytically verified test concentrations Fraunhofer (IME), Schmallenberg, Germany, Report No.: EBR-013/4-20 GLP / Unpublished	Y (New/First)	Evonik Industries AG
A7.4.1.3/01	[redacted]	2002	Ampholyt 20/100 – determination of the growth inhibition of the green algae Desmodesmus subspicatus (Alga, growth inhibition test) Infracor GmbH, Marl, Germany, Report No.: AW 488 GLP / Unpublished	Y (New/First)	Evonik Industries AG
A7.4.1.3/02	[redacted]	1995	Effect of TEGO 2000 on the growth of the green alga Selenastrum capricornutum TNO, Delft, The Netherlands, Report No.: IMW-94-0039-03 GLP / Unpublished	Y (New/First)	Evonik Industries AG
A7.4.1.3/03	[redacted]	2007	Alga, Growth inhibition test - Effect of Ampholyt 20 on the growth of Pseudokirchneriella subcapitata, static conditions Fraunhofer (IME), Schmallenberg, Germany, Report No.: EBR-013/4-30 GLP / Unpublished	Y (New/First)	Evonik Industries AG
A7.4.1.3/04	[redacted]	2008	Amendment to study report alga, growth inhibition test - Effect of Ampholyt 20 on the growth of Pseudokirchneriella subcapitata, static conditions Fraunhofer (IME), Schmallenberg, Germany, Report No.: EBR-013/4-30 GLP / Unpublished	Y (New/First)	Evonik Industries AG
A7.4.1.3/05	[redacted]	2008	Alga, growth inhibition test. Effect of Ampholyt 20 on the growth of Pseudokirchneriella subcapitata, static conditions Fraunhofer (IME), Schmallenberg, Germany, Report No.: EBR-013/4-30/1 GLP / Unpublished	Y (New/First)	Evonik Industries AG
A7.4.1.4/01	[redacted]	2002	Ampholyt 20/100 – determination of the inhibition of activated sludge respiration Infracor GmbH, Marl, Germany, Report No.: BH-02/05 GLP / Unpublished	Y (New/First)	Evonik Industries AG

Section No / Reference No	Author(s)	Year	Title. Source, Report No. GLP / (Un)Published	Data Protection Claimed (Yes/No)	Owner
A7.4.1.4/02	[redacted]	2000	Activated sludge respiration inhibition test with TEGO 2000 (contact time: 3 hours) NOTOX B.V., 's-Hertogenbosch, The Netherlands, Report No.: 291342 GLP / Unpublished	Y (New/First)	Evonik Industries AG
A7.4.1.4/03	[redacted]	1992	Abwasser- und Peptonabbauhemmungsuntersuchungen im Sapromat und modifizierter OECD-Bestätigungstest mit TEGOL 2000 (with english translation) Bayerische Landesanstalt für Wasserforschung, München, Germany, Not GLP / Unpublished	Y (New/First)	Evonik Industries AG
A7.4.2/01	[redacted]	2007	Estimation of the bioconcentration factor (BCF-fish) of Ampholyt 20 EBRC Consulting GmbH, Hannover, Germany, Report No.: DEG-20070628-01 Not GLP / Unpublished	Y (New/First)	Evonik Industries AG
A7.4.3.2/01	[redacted]	2008	Oncorhynchus mykiss, juvenile growth test (OECD 215) flow-through exposure. Effect of Ampholyt 20 on the growth of juvenile rainbow trout Fraunhofer (IME), Schmallenberg, Germany, Report No.: EBR-013/4-63 GLP / Unpublished	Y (New/First)	Evonik Industries AG
A7.4.3.4/01	[redacted]	2007	Daphnia magna, Reproduction test (OECD 211) Semi-static exposure. Effect of Ampholyt 20 on the reproduction of Daphnia magna Fraunhofer (IME), Schmallenberg, Germany, Report No.: EBR-013/4-21 GLP / Unpublished	Y (New/First)	Evonik Industries AG
A7.4.3.4/02	[redacted]	2008	Amendment no. 1 to study report Daphnia magna, reproduction test (OECD 211) semi-static exposure. Effect of Ampholyt 20 on the reproduction of Daphnia magna. Recalculation of effect values based on analytically verified test concentrations Fraunhofer (IME), Schmallenberg, Germany, Report No.: EBR-013/4-21 GLP / Unpublished	Y (New/First)	Evonik Industries AG
A7.5.1.1/01	[redacted]	2007	Soil microorganisms – effects of Ampholyt 20 on nitrogen and carbon transformation Fraunhofer (IME), Schmallenberg, Germany, Report No.: EBR-013/3-35 GLP / Unpublished	Y (New/First)	Evonik Industries AG

Section No / Reference No	Author(s)	Year	Title. Source, Report No. GLP / (Un)Published	Data Protection Claimed (Yes/No)	Owner
A7.5.1.2/01	[redacted]	2007	Earthworm acute toxicity test – acute toxicity of Ampholyt 20 on Eisenia fetida Fraunhofer (IME), Schmallenberg, Germany, Report No.: EBR-013/3-08 GLP / Unpublished	Y (New/First)	Evonik Industries AG
A7.5.1.3/01	[redacted]	2007	Terrestrial plants, growth test: Effect of Ampholyt 20 on the seedling emergence and growth of Avena sativa, Lactuca sativa, Phaseolus aureus, Raphanus sativus, Sinapis alba, and Triticum aestivum Fraunhofer (IME), Schmallenberg, Germany, Report No.: EBR-013/3-40 GLP / Unpublished	Y (New/First)	Evonik Industries AG
A7.5.5.1/01	[redacted]	2007	Estimation of terrestrial bioconcentration factor (BCF-earthworm) of Ampholyt 20 EBRC Consulting GmbH, Hannover, Germany, Report No.: DEG-20070704-01 Not GLP / Unpublished	Y (New/First)	Evonik Industries AG
A8.1/01	[redacted]	2007	Safety data sheet according to 93/112/EC – version 3.2. Ampholyt 20 Goldschmidt AG, Essen, Germany, Not GLP / Unpublished	Y (New/First)	Evonik Industries AG

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B3.5/01	[redacted]	2000	Determination of the pH of an aqueous dispersion of Tego 2000 NOTOX B.V., 's-Hertogenbosch, The Netherlands, Report No.: 283455 GLP / Unpublished	Y (New/First)	Evonik Industries AG
B3.6/01	[redacted]	2000	Determination of the density (liquid) of Tego 2000 NOTOX B.V., 's-Hertogenbosch, The Netherlands, Report No.: 283422 GLP / Unpublished	Y (New/First)	Evonik Industries AG
B3.7/01	[redacted]	2011	Ampholyt 20: Real time storage stability testing (2 years). Dr. U. Noack-Laboratorien, Sarstedt, Germany Report no. CLR13263. GLP / Unpublished	Y (New/First)	Evonik Industries AG
B3.7/02	[redacted]	2007	Ampholyt 20 – low temperature stability of liquid formulations Dr.U.Noack-Laboratorien, Sarstedt, Germany, Report No.: CLN117231 GLP / Unpublished	Y (New/First)	Evonik Industries AG
B3.7/03	[redacted]	2009	Ampholyt 20: Accelerated storage procedure Dr. U. Noack-Laboratorien, Sarstedt, Germany Report no. CPL12678. GLP / Unpublished	Y (New/First)	Evonik Industries AG
B3.8/01	[redacted]	2007	Ampholyt 20 – persistent foaming Dr.U.Noack-Laboratorien, Sarstedt, Germany, Report No.: CFO117231 GLP / Unpublished	Y (New/First)	Evonik Industries AG
B3.10.1/01	[redacted]	2000	Determination of the surface tension of an aqueous solution of TEGO 2000 NOTOX B.V., 's-Hertogenbosch, The Netherlands, Report No.: 283444 GLP / Unpublished	Y (New/First)	Evonik Industries AG
B3.10.2/01	[redacted]	2007	Ampholyt 20, Batch no.: ES67345616, Kinematic viscosity Siemens AG, Frankfurt, Germany, Report No.: 20070644.01 GLP / Unpublished	Y (New/First)	Evonik Industries AG
B5/01	[redacted]		TEGO 2000: Labels of the product as presently marketed Goldschmidt GmbH, Essen, Germany, Not GLP / Published	N	
B5.10.2/01	[redacted]	2000	Experts report on the bactericidal activity of the chemical disinfectant TEGO 2000 produced by Goldschmidt AG for the use in food areas Goldschmidt AG, Essen, Germany, Not GLP / Unpublished	Y (New/First)	Evonik Industries AG

Section No / Reference No	Author(s)	Year	Title. Source, Report No. GLP / (Un)Published	Data Protection Claimed (Yes/No)	Owner
B5.10.2/02	[redacted]	2002	TEGO 2000, EN 1276 (August 2001): Quantitative suspension test for the evaluation of bactericidal activity (phase 2 / step 1) HygCen, Schwerin, Germany, Report No.: SN 2553.2 Not GLP / Unpublished	Y (New/First)	Evonik Industries AG
B5.10.2/03	[redacted]	2002	TEGO 2000 (Charge ES62705464): Quantitative Suspension test according to DIN EN 1276 (Membrane filtration) L+S AG, Bad Bocklet-Großenbrach, Germany, Report No.: 08607362 Not GLP / Unpublished	Y (New/First)	Evonik Industries AG
B5.10.2/04	[redacted]	2002	TEGO 2000 (Charge ES62705464): Quantitative Suspension test according to DIN EN 1650 (Membrane filtration) L+S AG, Bad Bocklet-Großenbrach, Germany, Report No.: 08607362 Not GLP / Unpublished	Y (New/First)	Evonik Industries AG
B5.10.2/05	[redacted]	2003	Expert report on virucidal activity of TEGO 2000 against vaccina virus strain Elstree Mikrolab GmbH, Bremen, Germany, Not GLP / Unpublished	Y (New/First)	Evonik Industries AG
B5.10.2/06	[redacted]	2003	Expert report on virucidal activity of TEGO 2000 against bovine viral diarrhea virus (surrogate of HCV) Mikrolab GmbH, Bremen, Germany, Not GLP / Unpublished	Y (New/First)	Evonik Industries AG
B5.10.2/07	[redacted]	2004	Expert report on virucidal activity of TEGO 2000 against herpes simplex virus type 1 Mikrolab GmbH, Bremen, Germany, Not GLP / Unpublished	Y (New/First)	Evonik Industries AG
B5.10.2/08	[redacted]	2004	Evaluation of the Effectiveness of TEGO 2000 against Bovine Coronavirus Mikrolab GmbH, Bremen, Germany, Not GLP / Unpublished	Y (New/First)	Evonik Industries AG
B5.10.2/09	[redacted]	1990	Evaluation of the effect on hepatitis B virus of TEGOL 2000 = TEGO 2000 Max v. Pettenkofer-Inst. Hyg., Med. Mikrobiol., München, Germany, Not GLP / Unpublished	Y (New/First)	Evonik Industries AG
B5.10.2/10	[redacted]	1986	External appraisal, TEGOL 2000 Inst. Hygiene und Technologie, München, Germany, Not GLP / Unpublished	Y (New/First)	Evonik Industries AG
B5.10.2/11	[redacted]	1986	Prüfung des Desinfektionsmittels TEGOL 2000 Inst. Hygiene und Technologie, München, Germany, Not GLP / Unpublished	Y (New/First)	Evonik Industries AG

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B5.10.2/12	[redacted]	1987	Prüfung des Desinfektionsmittels TEGOL 2000 bei 10°C; Ergänzung zu unserem Gutachten vom 04.11.86 Inst. Hygiene und Technologie, München, Germany, Not GLP / Unpublished	Y (New/First)	Evonik Industries AG
B5.10.2/13	[redacted]	1988	Prüfung des Desinfektionsmittels TEGOL 2000 bei 20°C; Ergänzung zu unserem Gutachten vom 04.11.86 Inst. Hygiene und Technologie, München, Germany, Not GLP / Unpublished	Y (New/First)	Evonik Industries AG
B5.10.2/14	[redacted]	1988	Prüfung des Desinfektionsmittels TEGOL 2000 bei 10°C; Ergänzung zu unserem Gutachten vom 04.11.86 and 13.05.87 and 18.04.88 Inst. Hygiene und Technologie, München, Germany, Not GLP / Unpublished	Y (New/First)	Evonik Industries AG
B5.10.2/15	[redacted]	1990	Prüfung auf mikrobiozide Wirksamkeit Goldschmidt AG, Essen, Germany, Not GLP / Unpublished	Y (New/First)	Evonik Industries AG
B5.10.2/16	[redacted]	2003	Optimization of disinfection procedures in food supply facilities of the German armed forces in view to the B. cereus prevalence of surfaces and food Zentr. Inst. Sanit. BW Koblenz, Mainz, Germany, Not GLP / Unpublished	Y (New/First)	Evonik Industries AG
B5.10.2/17	[redacted]	1992	Virucidal efficacy of TEGOL 2000 against the human rotavirus, strain Wa Hygiene-Institut, Bremen, Germany, Not GLP / Unpublished	Y (New/First)	Evonik Industries AG
B5.10.2/18	[redacted]	2006	Evaluation of the effectiveness of Ampholyt 20 against avian influenza virus A Mikrolab GmbH, Bremen, Germany, Not GLP / Unpublished	N	Evonik Industries AG
B5.10.2/19	[redacted]	1978	Virucidal activity of some disinfectants containing amphoteric, cationic and cationogenic surface compounds (TEGO*) Archiv Lebensmittelhyg. 29, 81-120, Not GLP / Published	N	
B5.10.2/20	[redacted]	2007	Evaluation of disinfectant effectiveness BIOLAB SPA, Vimodrone, Italy, Report No.: AM1590 Not GLP / Unpublished	Y (New/First)	Evonik Industries AG

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B6.2/01	[redacted]	1988	Test for eye irritation of TEGOL 2000 (conc.) in rabbits IBR, Hannover, Germany, Report No.: 1-3-81-88 Not GLP / Unpublished	Y (New/First)	Evonik Industries AG
B6.6/01	[redacted]	2007	Estimation of human exposure to Ampholyt 20 from application of TEGO 2000 as a "private area and public health area disinfectant" EBRC Consulting GmbH, Hannover, Germany, Report No.: DEG-20070720-01 Not GLP / Unpublished	Y (New/First)	Evonik Industries AG
B6.6/02	[redacted]	2007	Estimation of human exposure to Ampholyt 20 from application of TEGO 2000 as a "veterinary hygiene biocidal product" EBRC Consulting GmbH, Hannover, Germany, Report No.: DEG-20070720-02 Not GLP / Unpublished	Y (New/First)	Evonik Industries AG
B6.6/03	[redacted]	2007	Estimation of human exposure to Ampholyt 20 from application of TEGO 2000 as a "food and feed area disinfectant" EBRC Consulting GmbH, Hannover, Germany, Report No.: DEG-20070720-03 Not GLP / Unpublished	Y (New/First)	Evonik Industries AG
B6.6/04	[redacted]	1998	Abschlussbericht zur Ermittlung der Tropfengrößenverteilung von TEGO 51 bei gegebener Düse , Not GLP / Unpublished	Y (New/First)	Evonik Industries AG
B6.6/05	[redacted]	2008	Estimation of human exposure to Ampholyt 20 from application of TEGO 2000 as a "private area and public health area disinfectant" EBRC Consulting GmbH, Hannover, Germany, Report No.: DEG-20080626-01 Not GLP / Unpublished	Y (New/First)	Evonik Industries AG
B6.6/06	[redacted]	2008	Estimation of human exposure to Ampholyt 20 from application of TEGO 2000 as a "veterinary hygiene biocidal product" EBRC Consulting GmbH, Hannover, Germany, Report No.: DEG-20080722-01 Not GLP / Unpublished	Y (New/First)	Evonik Industries AG
B6.6/07	[redacted]	2008	Estimation of human exposure to Ampholyt 20 from application of TEGO 2000 as a "food and feed area disinfectant" EBRC Consulting GmbH, Hannover, Germany, Report No.: DEG-20080626-03 Not GLP / Unpublished	Y (New/First)	Evonik Industries AG

Section No / Reference No	Author(s)	Year	Title. Source, Report No. GLP / (Un)Published	Data Protection Claimed (Yes/No)	Owner
B6.6/08	[redacted]	2012	Estimation of human exposure to Ampholyt 20 from application of TEGO 2000 as a "private area and public health area disinfectant". EBRC Consulting GmbH, Hannover, Germany, Report No.: DEG-20120405-01 Not GLP / Unpublished	Y (New/First)	Evonik Industries AG
B6.6/09	[redacted]	2012	Estimation of human exposure to Ampholyt 20 from application of TEGO 2000 as a "veterinary hygiene biocidal product". EBRC Consulting GmbH, Hannover, Germany, Report No.: DEG-20120406-01 Not GLP / Unpublished	Y (New/First)	Evonik Industries AG
B7.1/01	[redacted]	2007	Estimation of predicted environmental concentrations of Ampholyt 20 from application of TEGO 2000 as a disinfectant (Product types 2, 3, and 4) EBRC Consulting GmbH, Hannover, Germany, Report No.: DEG-20070722-01 Not GLP / Unpublished	N	Evonik Industries AG
B7.1/02	[redacted]	2009	Estimation of predicted environmental concentrations of Ampholyt 20 from application of TEGO 2000 as a disinfectant (Product types 2, 3, and 4) EBRC Consulting GmbH, Hannover, Germany, Report No.: DEG-20090114-01 Not GLP / Unpublished	Y (New/First)	Evonik Industries AG
B7.1/03	[redacted]	2009	Estimation of predicted environmental concentrations of Ampholyt 20 in groundwater (PECgw) using FOCUS PELMO 3.3.2 following application of TEGO 2000 as a disinfectant (Product types 2, 3, and 4) EBRC Consulting GmbH, Hannover, Germany, Report No.: DEG-20090203-01 Not GLP / Unpublished	Y (New/First)	Evonik Industries AG
B8.1/01	[redacted]	2007	Safety data sheet according to 93/112/EC – version 3.2. Ampholyt 20 Goldschmidt AG, Essen, Germany, Not GLP / Unpublished	Y (New/First)	Evonik Industries AG