Tralopyril (CAS# 122454-29-9) GreenScreen® for Safer Chemicals (GreenScreen®) Assessment

Prepared for:

State of Washington Department of Ecology

Prepared by:

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GreenScreen® Executive Summary for Tralopyril (CAS# 122454-29-9)

Tralopyril is used primarily as an antifoulant in paint for ships and other marine structures at use levels of 2.9% to 6%.

Tralopyril was assigned a **GreenScreen Benchmark™ Score of 2** ("Use but Search for Safer Substitutes"). This score is based on the following hazard score combinations:

- Benchmark 2c
  - High persistence (P) and Very High Ecotoxicity (acute aquatic (AA) and chronic aquatic (CA))
  - High persistence (P) and Very High Group II Human Toxicity (acute mammalian toxicity (AT))
  - High persistence (P) and High Group II* Human Toxicity (repeated dose systemic toxicity (STr*) and repeated dose neurotoxicity (Nr*))
  - High persistence (P) and Moderate Group II Human Toxicity (skin irritation (IrS) and eye irritation (IrE))
- Benchmark 2f
  - Very High Ecotoxicity (acute aquatic (AA) and chronic aquatic (CA)
  - Very High Group II Human Toxicity (acute mammalian toxicity (AT))
  - High Group II* Human Toxicity (repeated dose systemic toxicity (STr*) and repeated dose neurotoxicity (Nr*))

Data gaps (DG) exist for respiratory sensitization (SnR*). As outlined in CPA (2013) Section 12.2 (Step 8 – Conduct a Data Gap Analysis to assign a final Benchmark score), tralopyril meets requirements for a GreenScreen® Benchmark Score of 2 despite the hazard data gaps. In a worst-case scenario, if tralopyril was assigned a High score for the data gap SnR*, it would still be categorized as a Benchmark 2 Chemical.

GreenScreen® Benchmark Score for Relevant Route of Exposure:

As a standard approach for GreenScreen® evaluations, all exposure routes (oral, dermal and inhalation) were evaluated together, so the GreenScreen® Benchmark Score of 2 ("Use but Search for Safer Substitutes") is applicable for all routes of exposure.

GreenScreen® Hazard Ratings for Tralopyril

Group I Human					Group II and II* Human									Ecotox		Fate		Physical	
C	M	R	D	E	AT	ST		N		SnS*	SnR*	IrS	IrE	AA	CA	P	B	Rx	F
						single	repeated*	single	repeated*										
L	L	*L*	L	L	vH	DG	H	DG	H	L	DG	*M*	*M*	vH	vH	*H*	vL	L	L

Note: Hazard levels (Very High (vH), High (H), Moderate (M), Low (L), Very Low (vL)) in *italics* reflect estimated values, authoritative B lists, screening lists, weak analogues, and lower confidence. Hazard levels in **BOLD** font are used with good quality data, authoritative A lists, or strong analogues. Group II Human Health endpoints differ from Group II* Human Health endpoints in that they have four hazard scores (i.e., vH, H, M, and L) instead of three (i.e., H, M, and L), and are based on single exposures instead of repeated exposures. Please see Appendix A for a glossary of hazard acronyms. NA: Not assessed.

GreenScreen® Assessment for Tralopyril (CAS# 122454-29-9)

Method Version: GreenScreen® Version 1.2[1]
Assessment Type[2]: Certified

Chemical Name: Tralopyril

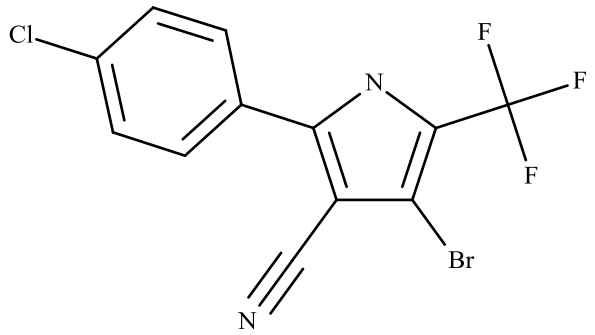
CAS Number: 122454-29-9

GreenScreen® Assessment Prepared By:
Name: Mouna Zachary, Ph.D.
Title: Toxicologist
Organization: ToxServices LLC
Date: December 06, 2014
Assessor Type: Licensed GreenScreen® Profiler

Quality Control Performed By:
Name: Bingxuan Wang, Ph.D.
Title: Toxicologist
Organization: ToxServices LLC
Date: January 4, 2015

Confirm application of the *de minimus* rule[3]: N/A

Chemical Structure(s):



Tralopyril (CAS# 122454-29-9)

Also called: 4-Bromo-2-(4-chlorophenyl)-5-(trifluoromethyl)pyrrole-3-carbonitrile; UNII-MEC6MCY7QB; 1H-Pyrrole-3-carbonitrile, 4-bromo-2-(4-chlorophenyl)-5-(trifluoromethyl)- (ChemIDplus 2014)

Chemical Structure(s) of Chemical Surrogates Used in the GreenScreen®:
No surrogates were used as available data were sufficient for the GreenScreen® assessment. However, for ecotoxicity endpoints (acute aquatic and chronic aquatic), data on the degradation products of tralopyril were used to support its GreenScreen® scoring for these endpoints. Tralopyril is expected to degrade rapidly via hydrolysis to CCL322,250 (parent compound minus fluorines and remaining

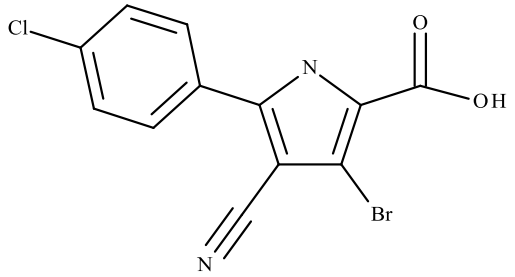
[1] Use GreenScreen® Assessment Procedure (Guidance) V1.2

[2] GreenScreen® reports are either "UNACCREDITED" (by unaccredited person), "AUTHORIZED" (by Authorized GreenScreen® Practitioner), "CERTIFIED" (by Licensed GreenScreen® Profiler or equivalent) or "CERTIFIED WITH VERIFICATION" (Certified or Authorized assessment that has passed GreenScreen® Verification Program)

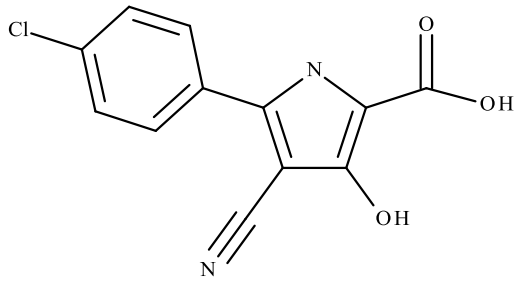
[3] Every chemical in a material or formulation should be assessed if it is:
1. intentionally added and/or
2. present at greater than or equal to 100 ppm

carbon hydrated, see below). CL322,250 further degrades by losing a bromine to debrominated CL322,250 forming CL322,248, only under anaerobic conditions or in sea water (U.S. EPA 2013a).

CL322,250 (No CAS number was identified)



CL322,248 (No CAS number was identified)



Identify Applications/Functional Uses:

1. As an antifoulant in paint at use levels of 2.9% to 6% (U.S. EPA 2013a, Pettit Paint 2014)

GreenScreen® Summary Rating for Tralopyril[4]: Tralopyril was assigned a **GreenScreen Benchmark™ Score of 2** ("Use but Search for Safer Substitutes"). This score is based on the following hazard score combinations:

- Benchmark 2c
  - High persistence (P) and Very High Ecotoxicity (acute aquatic (AA) and chronic aquatic (CA))
  - High persistence (P) and Very High Group II Human Toxicity (acute mammalian toxicity (AT))
  - High persistence (P) and High Group II* Human Toxicity (repeated dose systemic toxicity (STr*) and repeated dose neurotoxicity (Nr*))
  - High persistence (P) and Moderate Group II Human Toxicity (skin irritation (IrS) and eye irritation (IrE))
- Benchmark 2f
  - Very High Ecotoxicity (acute aquatic (AA) and chronic aquatic (CA))
  - Very High Group II Human Toxicity (acute mammalian toxicity (AT))

[4] For inorganic chemicals with low human and ecotoxicity across all hazard endpoints and low bioaccumulation potential, persistence alone will not be deemed problematic. Inorganic chemicals that are only persistent will be evaluated under the criteria for Benchmark 4.

- High Group II* Human Toxicity (repeated dose systemic toxicity (STr*) and repeated dose neurotoxicity (Nr*))

Data gaps (DG) exist for respiratory sensitization (SnR*). As outlined in CPA (2013) Section 12.2 (Step 8 – Conduct a Data Gap Analysis to assign a final Benchmark score), tralopyril meets requirements for a GreenScreen® Benchmark Score of 2 despite the hazard data gaps. In a worst-case scenario, if tralopyril were assigned a High score for the data gap SnR*, it would still be categorized as a Benchmark 2 Chemical.

Figure 1: GreenScreen®Hazard Ratings for Tralopyril

Group I Human					Group II and II* Human									Ecotox		Fate		Physical	
C	M	R	D	E	AT	ST		N		SnS*	SnR*	IrS	IrE	AA	CA	P	B	Rx	F
						single	repeated*	single	repeated*										
L	**L**	*L*	**L**	**L**	**vH**	DG	**H**	DG	**H**	**L**	DG	*M*	*M*	**vH**	**vH**	*H*	**vL**	**L**	**L**

Note: Hazard levels (Very High (vH), High (H), Moderate (M), Low (L), Very Low (vL)) in *italics* reflect estimated values, authoritative B lists, screening lists, weak analogues, and lower confidence. Hazard levels in **BOLD** font are used with good quality data, authoritative A lists, or strong analogues. Group II Human Health endpoints differ from Group II* Human Health endpoints in that they have four hazard scores (i.e., vH, H, M, and L) instead of three (i.e., H, M, and L), and are based on single exposures instead of repeated exposures. Please see Appendix A for a glossary of hazard acronyms. DG: Data Gap

Transformation Products and Ratings:

Identify feasible and relevant fate and transformation products (i.e., dissociation products, transformation products, valence states) **and/or moieties of concern**[5]

Hydrolysis of tralopyril is expected to be an important environmental fate process based on its hydrolysis half-lives of 14-175 days at pH 5 to 0.1-0.6 days in pH 9 and seawater. The major hydrolysis degradation product is CL 322,250, which is the defluorinated parent compound with a carboxylic acid group. Hydrogen fluoride is produced as a hydrolysis byproduct. CL322,250 further degrades to CL 322,248, which the debrominated compound of CL322,250, only under anaerobic conditions or in sea water. Hydrogen bromide is produced during this process. When heated to decomposition, tralopyril emits toxic fumes of carbon oxides, nitrogen oxides (NOx), hydrogen chloride gas, hydrogen bromide gas and hydrogen fluoride (U.S. EPA 2013a).

The Pharos listings for transformation products are tabulated below. No CAS numbers or data were identified for the transformation products CL322,250 and CL 322,248.

Hydrogen fluoride received a score of LT-P1. In order to determine if this transformation product is a Benchmark 1 and will affect the benchmark score of tralopyril, further assessment of hydrogen fluoride is required. Under current GreenScreen guidelines, separate evaluation of hydrogen fluoride is not required.

[5] A moiety is a discrete chemical entity that is a constituent part or component of a substance. A moiety of concern is often the parent substance itself for organic compounds. For inorganic compounds, the moiety of concern is typically a dissociated component of the substance or a transformation product.

Functional Use	Life Cycle Stage	Transformation Pathway	Transformation Products	CAS #	Feasible and Relevant?	GreenScreen® List Translator Score or Benchmark Score[6,7]
N/A	End of life	Heat decomposition, hydrolysis	Hydrogen bromide (HBr)	10035-10-6	Yes	LT-U
N/A	End of life	Heat decomposition, hydrolysis	Hydrogen fluoride (HF)	7664-39-3	Yes	LT-P1
N/A	End of Life	Heat decomposition	Nitrogen dioxide (NO_2)	10102-44-0	Yes	LT-U
N/A	End of Life	Heat decomposition	Nitrogen trioxide (N_2O_3)	10544-73-7	Yes	LT-U
N/A	End of Life	Heat decomposition	Nitrogen tetroxide (N_2O_4)	10544-72-6	Yes	LT-U
N/A	End of Life	Heat decomposition	Nitrogen oxide (NO_x)	11104-93-1	Yes	LT-U
N/A	End of Life	Heat decomposition	Hydrogen chloride (HCl)	7647-01-0	Yes	LT-U

Introduction

Tralopyril is an arylpyrrole biocide that acts by uncoupling mitochondrial oxidative phosphorylation. Its trade name is Econea™ and is used mainly as antifoulant in paint for ships and other marine structures at use levels of 2.9% to 6% (U.S. EPA 2013, Pettit Paint 2014).

ToxServices assessed tralopyril against GreenScreen® Version 1.2 (CPA 2013) following procedures outlined in ToxServices' SOP 1.69 (GreenScreen® Hazard Assessment) (ToxServices 2013).

GreenScreen® List Translator Screening Results

The GreenScreen® List Translator identifies specific authoritative or screening lists that should be searched to identify GreenScreen® benchmark 1 chemicals (CPA 2012b). Pharos (Pharos 2014) is an online list-searching tool that is used to screen chemicals against the List Translator electronically. It checks all of the lists in the List Translator with the exception of the U.S. Department of Transportation (U.S. DOT) lists (U.S. DOT 2008a,b) and these should be checked separately in conjunction with running the Pharos query. The output indicates benchmark or possible benchmark scores for each

[6] The GreenScreen® List Translator identifies specific authoritative or screening lists that should be searched to screen for GreenScreen® benchmark 1 chemicals (CPA 2012b). Pharos (Pharos 2014) is an online list-searching tool that is used to screen chemicals against the lists in the List Translator electronically.
[7] The way you conduct assessments for transformation products depends on the Benchmark Score of the parent chemical (See Guidance).

human health and environmental endpoint. The output for tralopyril can be found in Appendix C and a summary of the results can be found below:

- Mammalian
  - Korea NIER - GHS Classification - Acute toxicity (inhalation) - Category 3 [H331 - Toxic if inhaled]
  - Korea NIER - GHS Classification - Acute toxicity (oral) - Category 2 [H300 - Fatal if swallowed]
  - New Zealand HSNO/GHS - 6.1B (oral) - Acutely toxic
  - New Zealand HSNO/GHS - 6.1C (inhalation) - Acutely toxic
- Acute Aquatic
  - Korea NIER - GHS Classification - Hazardous to the aquatic environment (acute) - Category 1 [H400 - Very toxic to aquatic life]
  - New Zealand HSNO/GHS - 9.1A (fish) - Very ecotoxic in the aquatic environment
- Terrestrial
  - New Zealand HSNO/GHS - 9.3A - Very ecotoxic to terrestrial vertebrates
  - New Zealand HSNO/GHS - 9.4B - Ecotoxic to terrestrial invertebrates
- Chronic Aquatic
  - Korea NIER - GHS Classification - Hazardous to the aquatic environment (chronic) - Category 1 [H410 - Very toxic to aquatic life with long lasting effects]

Tralopyril is not listed by the DOT.

PhysicoChemical Properties of Tralopyril

Tralopyril is a white-off powder that is soluble in water at room temperature. Its experimental partition coefficient of 3.5 indicates that it is unlikely to have high bioaccumulation potential for animals and plants.

Table 1: Physical and Chemical Properties of Tralopyril (CAS #122454-29-9)

Property	Value	Reference
Molecular formula	$C_{12}H_5BrClF_3N_2$	ChemIDplus 2014
SMILES Notation	c1cc(ccc1c2c(c(c([nH]2)C(F)(F)F)Br)C#N)Cl	ChemIDplus 2014
Molecular weight	349.5365	ChemIDplus 2014
Physical state	Solid	U.S. EPA 2013a
Appearance	Off-white powder	U.S. EPA 2013a
Melting point	252.3 - 253.4^{o}C	U.S. EPA 2013a
Vapor pressure	3.45 x 10^{-10} mm Hg at 25^{o}C	U.S. EPA 2013a
Water solubility	Insoluble (0.16-0.17 mg/L)	U.S. EPA 2013a
Dissociation constant	pKa = 7.08 at 26^{o}C	U.S. EPA 2013a
Density/specific gravity	1.714 g/cm^3 at 20^{o}C	U.S. EPA 2013a
Partition coefficient	Log K_{ow} = 3.5	U.S. EPA 2013a

Hazard Classification Summary Section:

Group I Human Health Effects (Group I Human)

Carcinogenicity (C) Score (H, M, or L): *L*
Tralopyril was assigned a score of Low for carcinogenicity based on ECHA report that tralopyril is unlikely to be carcinogenic. Confidence level was reduced due to lack of experimental data. GreenScreen® criteria classify chemicals as a Low hazard for carcinogenicity when adequate data are available and negative, there are no structural alerts, and they are not classifiable under GHS (CPA 2012a).

- Authoritative and Screening Lists
  - *Authoritative:* not on any authoritative lists.
  - *Screening:* not on any screening lists.
- No data were identified for this endpoint.
- ECHA 2014
  - ECHA stated in its opinion document on the application for approval of the active substance tralopyril, that tralopyril is unlikely to be carcinogenic and is not classified to carcinogenicity endpoint according to the EU Regulation on Classification, Labelling and Packaging of Substances and Mixtures (CLP regulation).
- Toxtree 2014
  - No experimental data were identified for this endpoint. Modeling was performed using ToxTree program v2.6.6. Tralopyril contains at least one structural alert for non-genotoxic carcinogenicity (Halogenated benzene). However it is predicted negative for genotoxic carcinogenicity (see Appendix D).
- VEGA 2012
  - Tralopyril is predicted to be non-carcinogenic. However, the reliability of this prediction is reduced because the compound could be out of the model applicability domain. The program stated that one or more fragments possibly related to carcinogenic activity were found (Halogenated benzene). See Appendix E for justification
- U.S. EPA 2013b
  - Tralopyril is predicted to have low to moderate carcinogenic concern when evaluated as a halogenated aromatic hydrocarbon by OncoLogic. The software was unable to evaluate the moiety of the five-carbon ring with bromine and fluorine substitutions. See Appendix F for justification.
- Based on weight of evidence, a Low score was assigned. Although ToxTree modeling indicates that tralopyril contains a structural alert for non-genotoxic carcinogenicity; VEGA modeling predicts that tralopyril is not a carcinogen with acceptable reliability and OncoLogic's evaluation of the benzene moiety indicates low to moderate carcinogenicity concern. In addition, ECHA concluded in its PBT assessment of tralopyril that it does not meet the criteria for carcinogenicity endpoint. Therefore, based on weight of evidence, tralopyril is unlikely to be a carcinogen and a score of Low was assigned.

Mutagenicity/Genotoxicity (M) Score (H, M, or L): L
Tralopyril was assigned a score of Low for mutagenicity/genotoxicity based on negative results for mutagenicity/clastogenicity in *in vitro* and *in vivo* tests. GreenScreen® criteria classify chemicals as a Low hazard for mutagenicity/genotoxicity when negative data for mutagenicity and clastogenicity, no structural alerts, and no GHS classification are available (CPA 2012a).

- Authoritative and Screening Lists

  - *Authoritative:* not on any authoritative lists.
  - *Screening:* not on any screening lists.
- U.S. EPA 2013a
  - Negative results for mutagenicity were obtained in a mammalian cell mutation assay conducted similar to OECD 476. Chinese hamster ovary (CHO) cells were exposed to tralopyril (94%) in dimethylsulfoxide up to cytotoxic and insolubility concentrations, with and without metabolic activation. No increase in the mutation frequency was observed in the presence or absence of metabolic activation.
  - Tralopyril was negative in a mouse micronucleus assay conducted similar to OECD Guideline 474 in male mice (5/group). Animals were exposed to tralopyril (>94% pure) in olive oil at doses of 0, 3, 6, or 12 mg/kg via oral gavage and bone marrow cells were harvested at 24 hours after exposure. Treatment did not induce an increase in the number of polychromatic or normochromatic erythrocytes containing micronuclei.

Reproductive Toxicity (R) Score (H, M, or L): *L*
Tralopyril was assigned a score of Low for reproductive toxicity based on ECHA report that tralopyril is unlikely to be a reproductive toxicant. Confidence level was reduced due to lack of experimental data. GreenScreen®criteria classify as a Low hazard for reproductive toxicity when adequate data are available and are negative for reproductive toxicity, and when they are not classified under GHS (CPA 2012a).

- Authoritative and Screening Lists
  - *Authoritative:* Not listed on any authoritative lists for this endpoint.
  - *Screening:* Not listed on any screening lists for this endpoint.
- ECHA 2014
  - ECHA stated in its opinion document on the application for approval of the active substance tralopyril, that tralopyril is unlikely to be a reproductive toxicant and is not classified to reproductive toxicity endpoint according to CLP regulation.

Developmental Toxicity incl. Developmental Neurotoxicity (D) Score (H, M, or L): L
Tralopyril was assigned a score of Low for developmental toxicity based on experimental data. GreenScreen® criteria classify as a Low hazard for developmental toxicity when adequate data are available and are negative for developmental toxicity, and when they are not classified under GHS (CPA 2012a).

- Authoritative and Screening Lists
  - *Authoritative:* Not listed on any authoritative lists for this endpoint.
  - *Screening:* Not listed on any screening lists for this endpoint.
- U.S. EPA 2013a
  - In a developmental toxicity study similar to OECD 414, time-mated Sprague-Dawley female rats (25/dose) were administered oral doses of tralopyril (94.6%) at 0, 5, 10, or 20 mg/kg/day via gavage on gestation days 6 to 19. Treatment related maternal toxicity effects were seen in dams exposed to 20 mg/kg/day. These included increased mortality, frequent salivation, reduced body weight gain, decreased uterine weight, decreased corrected body weight gain, increased resorptions (total, early and late) and decreased placental weight. Frequent salivation was also seen in animal at 10 mg/kg/day dose group.. Pups in the high and mid dose group showed decrease in mean fetal weight and exhibited disturbances and delays of ossification (non/incomplete/dumbbell/biparte) of various skeletal structures (skull, sternebrae), as well as supernumerary (14th) and wavy ribs. The ossification delays, however, were considered to be secondary to maternal toxicity and correlated with decreased

fetal weight. The study authors identified a maternal and developmental toxicity NOAELs of 5 mg/kg/day and LOAELs of 10 mg/kg/day based on significant clinical signs of toxicity and decreased pup weights, respectively.

- The weight of evidence suggests that tralopyril is not a selective developmental toxicant and developmental toxicity was only observed at doses which are significantly toxic to the dam.

Endocrine Activity (E) Score (H, M, or L): L

Tralopyril was assigned a score of Low for endocrine activity based on ECHA report that tralopyril is unlikely to cause endocrine toxicity. GreenScreen®criteria classify as a Low hazard for endocrine activity when adequate data are available and are negative for endocrine activity, and when they are not classified under GHS (CPA 2012a).

- Authoritative and Screening Lists
  - *Authoritative:* Not listed on any authoritative lists for this endpoint.
  - *Screening:* Not listed on any screening lists for this endpoint.
- Not listed as a potential endocrine disruptor on the EU Priority List of Suspected Endocrine Disruptors.
- Not listed as a potential endocrine disruptor on the OSPAR List of Chemicals of Possible Concern.
- ECHA 2014
  - Tralopyril is not considered to have endocrine-disrupting properties as no effects on endocrine organs and/or reproduction observed in standard toxicity studies which would raise a concern for potential endocrine disruption.

Group II and II* Human Health Effects (Group II and II* Human)

Note: Group II and Group II* endpoints are distinguished in the v 1.2 Benchmark system. For Systemic Toxicity and Neurotoxicity, Group II and II* are considered sub-endpoints and test data for single or repeated exposures may be used. If data exist for single OR repeated exposures, then the endpoint is not considered a data gap. If data are available for both single and repeated exposures, then the more conservative value is used.

Acute Mammalian Toxicity (AT) Group II Score (vH, H, M, or L): vH

Tralopyril was assigned a score of Very High for acute toxicity based on its inhalation LC_{50} being < 0.5 mg/L and on being classified to GHS category 2 by GHS-country (screening list) for oral acute toxicity. GreenScreen® criteria classify chemicals as a Very High hazard for acute toxicity when oral LD_{50} values are ≤ 50mg/kg, inhalation LC_{50} values are < 0.5 mg/L for dusts and when they are classified to GHS category 1 or 2 (CPA 2012a).

- Authoritative and Screening Lists
  - *Authoritative:* Not listed on any authoritative lists for this endpoint.
  - *Screening:* Korea NIER - GHS Classification - Acute toxicity (inhalation) - Category 3 [H331 - Toxic if inhaled]
  - *Screening:* Korea NIER - GHS Classification - Acute toxicity (oral) - Category 2 [H300 - Fatal if swallowed]
  - *Screening:* New Zealand HSNO/GHS - 6.1B (oral) - Acutely toxic (GHS category 2, based on an oral LD_{50} of 27.0 mg/kg in rats)
  - *Screening:* New Zealand HSNO/GHS - 6.1C (inhalation) - Acutely toxic (GHS category 3, based on an inhalation LC_{50} of 0.77 mg/L in rats)

- U.S. EPA 2013a
    - Oral
        - No acute oral toxicity value was provided but U.S. EPA stated that tralopyril has been shown to have severe acute toxicity by the oral route and is classified to Toxicity Category 1.
    - Dermal
        - LD_{50} > 2,000 mg/kg (rabbits); Classified to toxicity category 3 by U.S. EPA
    - Inhalation
        - $LC_{50} \leq 510$ mg/m^3 (rats) equivalent to 0.5 mg/L; classified to toxicity category 2 by U.S. EPA.
- ECHA 2014
    - The proposed classifications for tralopyril according to CLP Regulation, by ECHA in its opinion document on the application for approval of the active substance tralopyril, included Acute Tox. 2 with hazard statement of H300 (Fatal if swallowed), Acute Tox. 3 with hazard statement of H311 (Toxic in contact with skin) and Acute Tox. 2 with hazard statement of H330 (Fatal if inhaled).

Systemic Toxicity/Organ Effects incl. Immunotoxicity (ST)
***Group II Score (single dose)* (vH, H, M, or L):** DG
Tralopyril was assigned a score of Data Gap for systemic toxicity (single dose) based on a lack of data for this endpoint.

- Authoritative and Screening Lists
    - *Authoritative:* Not listed on any authoritative lists for this endpoint.
    - *Screening:* Not listed on any screening lists for this endpoint.
- No data were identified for this endpoint.

***Group II* Score (repeated dose)* (H, M, or L): H**
Tralopyril was assigned a score of High for systemic toxicity (repeated dose) based on an inhalation LOAEC of 0.02 mg/L generated from a 90-day study in rats. GreenScreen® criteria classify chemicals as a High hazard for systemic toxicity (repeated dose) when the oral LOAEL is ≤ 10 mg/kg/day, dermal LOAEL is ≤ 20 mg/kg/day, inhalation LOAEC is ≤ 0.02 mg/L for dust or when they are classified to GHS category 1 (CPA 2012a).

- Authoritative and Screening Lists
    - *Authoritative:* Not listed on any authoritative lists for this endpoint.
    - *Screening:* Not listed on any screening lists for this endpoint.
- U.S. EPA 2013a
    - Oral
        - In a previously described developmental toxicity study similar to OECD 414, time-mated Sprague-Dawley female rats (25/dose) were administered oral doses of tralopyril (94.6%) at 0, 5, 10, or 20 mg/kg/day via gavage on gestation days 6 to 19. Treatment related maternal toxicity effects were seen in dams exposed to 20 mg/kg/day. These included increased mortality, frequent salivation, reduced body weight gain, decreased uterine weight, decreased corrected body weight gain, increased resorptions (total, early and late) and decreased placental weight. Frequented salivation was also seen in animal at 10 mg/kg/day dose group. Based on this, a NOAEL of 5 mg/kg/day for maternal toxicity was established.
        - In a 90-day repeated dose toxicity study conducted similar to OPPTS 870.3100, Sprague-Dawley rats (10/sex/dose group) were fed diet containing tralopyril (94.6%)

at concentrations of 0, 80, 250, or 750 ppm. These concentrations were equivalent to 5.2, 16.2, or 51.9 mg/kg/day, in males and to 6.3, 20.9, or 62.0 mg/kg/day, in females as calculated by the authors. An additional study was performed with 10 Sprague-Dawley rats/sex/dose at the same concentration as above with 5 rats/sex/dose undergoing terminal sacrifice by *in situ* perfusion after anesthesia after 3 months of exposure and 5 rats/sex/dose undergoing a 4-week recovery period. The animals were evaluated for clinical signs of toxicity, body weight, food consumption, hematology, clinical chemistry, gross pathology, and histopathology. No mortalities or clinical signs of toxicity were observed but several treatment-related effects were noted. A decrease in food consumption was seen in both males and females administered 750 ppm in the first week of treatment. The body weight gains were low throughout the study, likely due to low food consumption. Hematology analysis revealed only minor (<10% difference from controls) effects in high-dose animals, except for a 15% increase in platelets in the males with a corresponding increase in females that was not significant (9%). All hematological values were comparable to controls after the 4-week recovery period. There were statistically significant increases in alanine aminotransferase (ALT) and alkaline phosphatase (ALP) in males at the mid- and high-dose groups, which correlated to increased relative liver weights in the main group. Females displayed a lesser increase in ALP with only the high-dose group demonstrating a statistically significant change. A statistically significant increase in ALP was also seen in males at 80 ppm. The urinary excretion of the phosphorus, urea, and magnesium and serum total cholesterol were also statistically significantly increased in both males and females at high dose. Animals in this dose group have shown a statistically significant decrease in serum glucose. Recovery groups presented no significant clinical chemistry parameter abnormalities, indicating full recovery from all treatment related effects. Differences in creatinine and triglycerides were generally <10% compared to the main control group and were thus attributed to biological variation. Mean terminal body weight was statistically significantly decreased in mid- and high-dose males and in high-dose females, which affected the absolute and/or relative weights of several organs. These changes in organ weights included increases in relative brain weights (mid- and high-dose males; high-dose females) and liver weights (mid- and high-dose males; all treated females). However, the changes in ALP and ALT enzyme were reversible after 4 weeks and the increases in the liver were likely due to increased metabolism of the test article. Treatment-related microscopic findings in the main group included minimal to slight mucosal hyperplasia of the duodenum in the mid- and high-dose males and females that were reversible after 4 weeks. Neurotoxicity was also observed, which wass described in the neurotoxicity section below. Based on reduced body weight and body weight gain, reduced food consumption, hematology, clinical chemistry, and organ weights, the study authors identified a LOAEL of 16.2 mg/kg/day and a NOAEL of 5.2 mg/kg/day in male rats. A LOAEL of 6.3 mg/kg/day, the lowest dose tested, was established for female rats based on microscopic findings of the brain and spinal cord. ToxServices established the systemic effect LOAEL at 62.0 mg/kg/day and the NOAEL at 20.9 mg/kg/day for females based on reduced body weight, clinical chemistry and urinalysis.

- Dermal
  - In a 90-day repeated dose toxicity study conducted similar to OECD Guideline 414, tralopyril (94.6%) was applied dermally to the shaved skin of male and female rats

(10/sex/dose) at doses of 0, 100, 300, or 1,000 mg/kg/day, 6 hours/day for 5 days/week. No toxicologically relevant changes were reported. A dose dependent dermal irritation was observed in both males and females at the mid- and high-dose group. Consequently, the study authors identified a dermal NOAEL of 100 mg/kg/day and a NOAEL of >1,000 mg/kg/day for systemic toxicity, the highest dose tested.

- Inhalation

  In a two-phase, 90-day inhalation toxicity and neurotoxicity study in rats conducted according to the EPA OPPTS 870.3465, 870.6200 Guidelines, Sprague-Dawley rats (10/sex/dose in Phase I; 12/sex/dose in Phase II) were exposed (nose only) to tralopyril (94% pure) at air concentrations of 0, 20, 40, and 80 mg/m^3 for 6 hours/day, 7 days/week. The particle size was within the accepted range. Neurotoxicity effects are discussed in the neurotoxicity (repeat dose) section below. In the phase I study, signs of toxicity were seen in both males and females at all doses and these included brown staining on the neck, forelimb, abdominal area thoracic area, inguinal area, hindlimb, rump, anogenital area, urogenital area, and tail. Mean body weight was reduced in males at all doses and in females at mid- and high- dose groups. Tralopyril treatment caused effects on the nasal passages including inflammation, ulcerations, and exudate in males, and inflammation, epithelial hyperplasia, hyperkeratosis, and degeneration of the olfactory epithelium in females at 20 mg/m^3 (the lowest concentration tested). Based on this, a LOAEC of 20 mg/m^3 was established for male and female rats.

- Based on the above data, the inhalation LOAEC identified for tralopyril was 20 mg/m^3/6h/day in a 90-day study, which is equivalent to 0.02 mg/L/6h/day. The concentration is equal to the GHS category 1 cut-off value of 0.02 mg/L/6h/day for dust. On the other hand, the oral LOAEL (16.2 mg/kg/day for males) was above the cut-off value for GHS category 1 (10 mg/kg/day), but the NOAEL of 5.2 mg/kg/day was below the cut-off value. Therefore, tralopyril is classified to GHS category 1 for systemic toxicity via the inhalation route of exposure.

Neurotoxicity (N)

***Group II Score (single dose)* (vH, H, M, or L):** DG

Tralopyril was assigned a score of Data Gap for neurotoxicity (single dose) based on the lack of data identified for this endpoint.

- Authoritative and Screening Lists
  - *Authoritative:* not on any authoritative lists
  - *Screening:* not on any screening lists
- Not classified as a developmental neurotoxicant (Grandjean and Landrigan 2006, 2014).
- No data were identified for this endpoint

***Group II* Score (repeated dose)* (H, M, or L): H**

Tralopyril was assigned a score of High for neurotoxicity (repeated dose) based on being classified to GHS category 1 for neurotoxicity effects. GreenScreen® criteria classify chemicals as a High hazard for neurotoxicity (repeated dose) when adequate data are available and they are classified to GHS category 1 (CPA 2012a).

- Authoritative and Screening Lists
  - *Authoritative:* Not listed on any authoritative lists for this endpoint.
  - *Screening:* Not listed on any screening lists for this endpoint.

- Not classified as a developmental neurotoxicant (Grandjean and Landrigan 2006, 2014).
- U.S. EPA 2013a
    - Oral
        - In the previously described 90-day repeated dose toxicity study conducted similar to OPPTS 870.3100, Sprague-Dawley rats (10/sex/dose group) were fed diet containing tralopyril (94.6%) at concentrations of 0, 80, 250, or 750 ppm. These concentrations were equivalent to 5.2, 16.2, or 51.9 mg/kg/day, in males and to 6.3, 20.9, or 62.0 mg/kg/day, in females as calculated by the authors. Changes in organ weights included increases in relative brain weights (mid- and high-dose males; high-dose females). The relative brain weights in the high-dose animals remained elevated after the recovery period, though the values were only statistically significant in males. . However, frank effects were noted in the brain at necropsy in these recovery group animals. Microscopic findings of the brain and spinal cord were observed in the main group as follows: minimal to moderate multifocal vacuolation of the brain in the mid- and high-dose males and females as well as the low-dose females. These were spongiform myelinopathy in the central and the proximal peripheral nervous system. The most severe lesions were multifocal vacuolation of the white matter and intramyelinic vacuolation of the peripheral nerves in the cauda equina (lumbar cord and root fibers). No treatment related changes in behavior or parasympathetic movements (e.g. tremors, convulsions, impaired gait, posture, response to handling, activity, or arousal) were seen. After 4 weeks of no treatment, microscopic lesions were still observed in the brain and lumbar region. Minimal multifocal vacuolization of the brain in high-dose males and minimal to slight multifocal vacuolization of the brain in mid- and high-dose females were observed; minimal to slight multifocal vacuolization of the lumbar in mid- and high-dose males and minimal to moderate multifocal vacuolization of the lumbar in all treated females. The lesion severity increased with dose. Based on the microscopic findings of the brain and spinal cord, the study authors identified a LOAEL of 6.3 mg/kg/day for female rats, the lowest dose tested.
    - Inhalation
        - In a two-phase, 90-day inhalation toxicity and neurotoxicity study in rats conducted according to the EPA OPPTS 870.3465, 870.6200 Guidelines, Sprague-Dawley rats (10/sex/dose in Phase I; 12/sex/dose in Phase II) were exposed (nose only) to tralopyril (94% pure) at air concentrations of 0, 20, 40, and 80 mg/m^3 for 6 hours/day, 7 days/week. The particle size was within the accepted range. In the phase II study, animals underwent a full neurobehavioural examination (functional observational battery [FOB] and motor activity testing) the day before dosing, and at weeks 3, 7, and 12. Cholinesterase activity was not determined. At study termination, 6 animals/sex/dose in the controls and 80-mg/m^3 groups were perfused *in situ* for neuropathological examination of the central and peripheral nervous system tissues. An increase in mortality among animals receiving the 80 mg/m^3 was seen. Decreased motor activity was observed in males at 40 and 80 mg/m^3 and axonal degeneration in the peroneal nerve was observed in both males and females at 80 mg/m^3. Based on decreased motor activity, the neurotoxicity NOAEL of 20 mg/m^3 and LOAEC of 40 mg/m^3 were established for male rats. In females, the neurotoxicity NOAEC of 40 mg/m^3 and LOAEC of 80 mg/m^3 were established based on axonal degeneration observed in the peroneal nerve.

- Based on the above data, neurotoxicity effects were seen in animals exposed to tralopyril via oral and inhalation route of exposure. The inhalation neurotoxicity LOAEC identified for tralopyril was 40 mg/m^3/6h/day for male rats in a 90-day study, which is equivalent to 0.04 mg/L/6h/day, which is within the cut off for GHS category 2 (0.02-0.2 mg/L/6h/day) for dust. However, the oral neurotoxicity LOAEL (6.3 mg/kg/day) was below the cut-off value for GHS category 1 (10 mg/kg/day). Therefore, tralopyril is classified to GHS category 1 for neurotoxicity toxicity via the oral route of exposure.

Skin Sensitization (SnS) Group II* Score (H, M, or L): L
Tralopyril was assigned a score of Low for skin sensitization based on negative findings in an animal study. GreenScreen® criteria classify chemicals as a Low hazard for skin sensitization when there are adequate negative data available, no structural alerts, and they are not GHS classified (CPA 2012a).

- Authoritative and Screening Lists
  - *Authoritative:* not on any authoritative lists.
  - *Screening:* not on any screening lists.
- U.S. EPA 2013a
  - Tralopyril did not cause skin sensitizing in guinea pigs when tested according to EPA OPPTS 870.2600 Guideline (no further details were provided).

Respiratory Sensitization (SnR) Group II* Score (H, M, or L): DG
Tralopyril was assigned a score of Data Gap for respiratory sensitization based on the lack of data identified for this endpoint.

- Authoritative and Screening Lists
  - *Authoritative:* not on any authoritative lists.
  - *Screening:* not on any screening lists.
- No data were identified for this endpoint

Skin Irritation/Corrosivity (IrS) Group II Score (vH, H, M, or L): *M*
Tralopyril was assigned a score of Moderate for skin irritation/corrosivity based on mild irritation to the skin seenin an animal study. GreenScreen® criteria classify chemicals as a Low hazard for skin irritation/corrosivity when adequate data are available and, and when they are classified to GHS category 3 (Mild irritant) (CPA 2012a). The confidence level was reduced due to lack of study details for a definitive GHS classification.

- Authoritative and Screening Lists
  - *Authoritative:* Not listed on any authoritative lists for this endpoint.
  - *Screening:* Not listed on any screening lists for this endpoint.
- U.S. EPA 2013a
  - In an acute dermal toxicity study conducted according to EPA OPPTS 870.2500 Guideline, tralopyril was mildly irritating to the skin of rabbits based on very slight erythema, but no edema at 72 hours (no further details were provided).

Eye Irritation/Corrosivity (IrE) Group II Score (vH, H, M, or L): *M*
Tralopyril was assigned a score of Low for eye irritation/corrosivity based on mild irritation to the eye seen in an animal study GreenScreen® criteria classify chemicals as a Low hazard for eye irritation/ corrosivity when adequate data are available and they are classified to GHS category 2B (mild irritating) (CPA 2012a). The confidence level was reduced due to lack of study details for a definitive GHS classification.

- Authoritative and Screening Lists
  - *Authoritative:* Not listed on any authoritative lists for this endpoint.
  - *Screening:* Not listed on any screening lists for this endpoint.
- U.S. EPA 2013a
  - Tralopyril was mildly irritating to rabbit eyes in an ocular irritation study conducted according to EPA OPPTS 870.2400 Guideline (no further details were provided).

Ecotoxicity (Ecotox)

Acute Aquatic Toxicity (AA) Score (vH, H, M, or L): vH
Tralopyril was assigned a score of Very High for acute aquatic toxicity based on acute aquatic toxicity data for tralopyril and its degradation product being less than 1.0 mg/L. GreenScreen® criteria classify chemicals as a Very High hazard for acute aquatic toxicity when acute aquatic toxicity values are <1.0 mg/L, when they are classified to GHS category 1, or associated with EU hazard statement of H400 (CPA 2012a).

- Authoritative and Screening Lists
  - *Authoritative:* Not present on any authoritative lists
  - *Screening:* Korea NIER - GHS Classification - Hazardous to the aquatic environment (acute) - Category 1 [H400 - Very toxic to aquatic life]
  - *Screening:* New Zealand HSNO/GHS - 9.1A (fish) - Very ecotoxic in the aquatic environment (GHS category 1; based on LC_{50} in fish of 0.26 mg/l (96h))
- U.S. EPA 2013a
  - *Tralopyril technical grade:* LC_{50} in *Oncorhynchus mykiss* of 1.300 ppb (fish, 96h)
  - *Tralopyril technical grade*: LC_{50} in *Lepomis macrochirus* of 3.2 ppb (fish, 96h)
  - *Tralopyril technical grade*: EC_{50} in *Daphnia magna* of 1.5 ppb (invertebrate, 48h)
  - *CL 322,250:* LC_{50} in *Oncorhynchus mykiss* of 520 ppb (fish, 96h)
  - *CL 322,250:* LC_{50} in *Lepomis macrochirus* of 1,200 ppb (fish, 96h)
  - *CL 322,250:* EC_{50} in *Daphnia magna* of 510 – 1,630 ppb (invertebrate, 48h)
- ECHA 2014
  - The proposed classifications for tralopyril according to CLP Regulation by ECHA in its opinion document on the application for approval of the active substance tralopyril, included Acute Aquatic 1 with a hazard statement of H400.

Chronic Aquatic Toxicity (CA) Score (vH, H, M, or L): vH
Tralopyril was assigned a score of Very High for chronic aquatic toxicity based on chronic aquatic toxicity data for tralopyril and its degradation product being less than 0.1 mg/L. GreenScreen® criteria classify chemicals as a Very High hazard for chronic aquatic toxicity when chronic aquatic toxicity values are < 0.1 mg/L (CPA 2012a).

- Authoritative and Screening Lists
  - *Authoritative:* Not present on any authoritative lists
  - *Screening:* Not present on any screening lists
  - *Others:* Korea NIER - GHS Classification - Hazardous to the aquatic environment (chronic) - Category 1 [H410 - Very toxic to aquatic life with long lasting effects]
- U.S. EPA 2013a
  - *Tralopyril technical grade:* LOEC and NOAEC in *Danio rerio* of 0.37 and 0.17 ppb, respectively (fish, 33d)
  - *Tralopyril technical grade:* LOAEC and NOAEC in *Daphnia magna* of 0.57 and 0.20 ppb, respectively (invertebrate, 21d)

- *CL 322,250:* LOEC and NOAEC in *Danio rerio* of 140 and 69 ppb, respectively (fish, 35d)
- *CL 322,250:* LOAEC and NOAEC in *Daphnia magna* of 540 and 300 ppb, respectively (invertebrate, 21d)

Environmental Fate (Fate)

Persistence (P) Score (vH, H, M, L, or vL): *H*

Tralopyril was assigned a score of High for persistence based on modeled data for its degradation product. Confidence level was reduced due to lack of experimental data. GreenScreen® criteria classify chemicals as a High hazard for persistence when data indicate a half-life of > 60-180 days in soil or sediment (CPA 2012a).

- Authoritative and Screening Lists
  - *Authoritative:* Not present on any authoritative lists
  - *Screening:* Domestic Substances List - DSL substances that are Persistent
- U.S. EPA 2013a
  - Tralopryl has a measured hydrolysis half-life of 0.4 day at pH of 7 in fresh water and 0.1 day at pH of 8 in seawater. The majority was converted to CL 322, 250, which did not undergo further hydrolysis (9.4 – 96.2% in distilled water and 95.2 – 96.3% in seawater after 30 days) (OPPTS 835.2120).
  - Tralopyril could also undergo aqueous photolysis at 25ºC with a half-life of approximately 1 day. The major degradation product CL 322,250 had a photolysis half-life of 0.025 day (OPPTS 835.2240). It was not clear what the photolysis products were. It should be noted that this study has not been formally reviewed by EPA and will be during the registration review.
  - Under aerobic conditions, the biodegradation half-life of tralopryl was 12 days in freshwater and 0.6 days in seawater at 21ºC. The major degradation product CL 322, 250 has a biodegradation half-life of 25 days in freshwater and 23 days in seawater (OPPTS 835.4400).
  - Under anaerobic conditions, the biodegradation half-life of tralopryl was 29 days in freshwater and 0.7 days in seawater at 21ºC. The major degradation product CL 322, 250 has a biodegradation half-life of 31 days in freshwater and 22 days in seawater (OPPTS 835.4300).
  - The degradation products of tralopryl have lower molecular weight, higher water solubility and log partition coefficient, suggesting that they will be present in water with a higher water:sediment ratio than the parent compound. In addition, neither tralopyril nor the degradation products are expected to enter the air from water based on MW, vapor pressures, Henry's Law constants and study results. Instead, sorption to sediment is a significant route of dissipation for tralopyril and its degradation products.
- U.S. EPA 2012
  - As tralopyril degrades rapidly by both abiotic and biotic processes to form CL 322,250, modeling was performed on its degradation product, CL322,250. The BIOWIN modeling Ready Biodegradable Predictor indicates that CL322,250 is not expected to be readily biodegradable.
  - Using a fugacity model, CL322,250 is predicted to appear mainly in the soil compartment (89.5%), with 10.1% in water and less than 1% in air and sediment.. The predicted half-life in soil and water are 120 days and 60 days, respectively (See Appendix G for modeling output).

- Based on the above data, no ready biodegradability studies were available for tralopyril or its degradation products. Therefore, a Very Low score was not applicable which is specifically reserved for chemicals that meet the 10-day biodegradation window in ready biodegradability studies. Measured data showed that tralopyril is hydrolyzed/photolyzed rapidly with the half-life of less than 1 day in water. The major degradation product CL 322, 250 is not hydrolysable, but had a very short photolysis half-life (0.025 day) in water (no photolysis products were identified). Aquatic metabolism studies on tralopyril and CL 322, 250 indicated that the aquatic degradation half-lives were between 12 – 31 days. Therefore, tralopyril does not meet the GHS Rapid degradability criteria (reaching 60 – 70% biodegradation in 28 days in water). Therefore, EPISuite modeling was performed to determine the predominant compartment of tralopyril's major degradation product. CL322,250 has a predicted a half-life of 120 days in its major compartment soil. ToxServices did not consider degradation half-lives data in the water (biotic and abiotic) relevant for this evaluation. The predicted half-life of 120 days in soil corresponds to a score of High. Confidence level was reduced due to the use of modeled data.

Bioaccumulation (B) Score (vH, H, M, L, or vL): vL
Tralopyril was assigned a score of Very Low for bioaccumulation based on both measured BCF and log K_{ow}. GreenScreen® criteria classify chemicals as a Very Low hazard for bioaccumulation when the BCF is less than 100 and log K_{ow} is < 4 (CPA 2012a).

- Authoritative and Screening Lists
  - *Authoritative:* Not present on any authoritative lists
  - *Screening:* Not present on any screening lists
- U.S. EPA 2013a
  - Tralopyril has a measured log K_{ow} value of 3.5 according to EPA OPPTS 830.7550 Guideline. A bioconcentration factor (BCF) of 3.2-32X in fish was also measured for tralopyril in carp at acidic conditions, which are below the pKa of 7.08. According to GHS criteria, this BCF suggests a low potential for bioconcentration in aquatic organisms.

Physical Hazards (Physical)

Reactivity (Rx) Score (vH, H, M, or L): L
Tralopyril was assigned a score of Low for reactivity based on not being explosive. GreenScreen® criteria classify chemicals as a Low hazard for reactivity when it is not explosive, unless there are data showing reactivity otherwise (CPA 2012a).

- Authoritative and Screening Lists
  - *Authoritative:* Not listed on any authoritative lists for this endpoint.
  - *Screening:* Not listed on any screening lists for this endpoint.
- U.S. EPA 2013a
  - Tralopyril is expected to be stable under normal conditions of use and not reactive when tested according to EPA OPPTS 830.6313 Guideline.

Flammability (F) Score (vH, H, M, or L): L
Tralopyril was assigned a score of Low for flammability based on not being flammable when tested. GreenScreen® criteria classify chemicals as a Low hazard for flammability when they are not classified as flammable solids to GHS (CPA 2012a).

- Authoritative and Screening Lists
  - *Authoritative:* not on any authoritative lists.
  - *Screening:* not on any screening lists.

- U.S. EPA 2013a
    - Tralopyril was not combustible/flammable when tested according to EPA OPPTS 830.6315 Guideline.

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APPENDIX A: Hazard Benchmark Acronyms
(in alphabetical order)

(AA) Acute Aquatic Toxicity

(AT) Acute Mammalian Toxicity

(B) Bioaccumulation

(C) Carcinogenicity

(CA) Chronic Aquatic Toxicity

(D) Developmental Toxicity

(E) Endocrine Activity

(F) Flammability

(IrE) Eye Irritation/Corrosivity

(IrS) Skin Irritation/Corrosivity

(M) Mutagenicity and Genotoxicity

(N) Neurotoxicity

(P) Persistence

(R) Reproductive Toxicity

(Rx) Reactivity

(SnS) Sensitization- Skin

(SnR) Sensitization- Respiratory

(ST) Systemic/Organ Toxicity

APPENDIX B: Results of Automated GreenScreen® Score Calculation for Tralopyril (CAS# 122454-29-9)

TOXSERVICES TOXICOLOGY RISK ASSESSMENT CONSULTING

GREEN SCREEN FOR SAFER CHEMICALS

GreenScreen® Score Inspector

Table 1: Hazard Table

			Group I Human					Group II and II* Human									Ecotox		Fate		Physical	
			Carcinogenicity	Mutagenicity/Genotoxicity	Reproductive Toxicity	Developmental Toxicity	Endocrine Activity	Acute Toxicity	Systemic Toxicity		Neurotoxicity		Skin Sensitization*	Respiratory Sensitization*	Skin Irritation	Eye Irritation	Acute Aquatic Toxicity	Chronic Aquatic Toxicity	Persistence	Bioaccumulation	Reactivity	Flammability
Table 2: Chemical Details									S	R *	S	R *	*	*								
Inorganic Chemical?	**Chemical Name**	**CAS#**	**C**	**M**	**R**	**D**	**E**	**AT**	**STs**	**STr**	**Ns**	**Nr**	**SNS***	**SNR***	**IrS**	**IrE**	**AA**	**CA**	**P**	**B**	**Rx**	**F**
No	Tralopyril	122454-29-9	*L*	L	*L*	L	L	vH	DG	H	DG	H	L	DG	*M*	*M*	vH	vH	*H*	vL	L	L

Table 3: Hazard Summary Table

Benchmark	a	b	c	d	e	f	g
1	No	No	No	No	No		
2	No	No	Yes	No	No	Yes	No
3	STOP						
4	STOP						

Table 4

Chemical Name	Preliminary GreenScreen® Benchmark Score
Tralopyril	2

Note: Chemical has not undergone a data gap assessment. Not a Final GreenScreen™ Score

Table 6

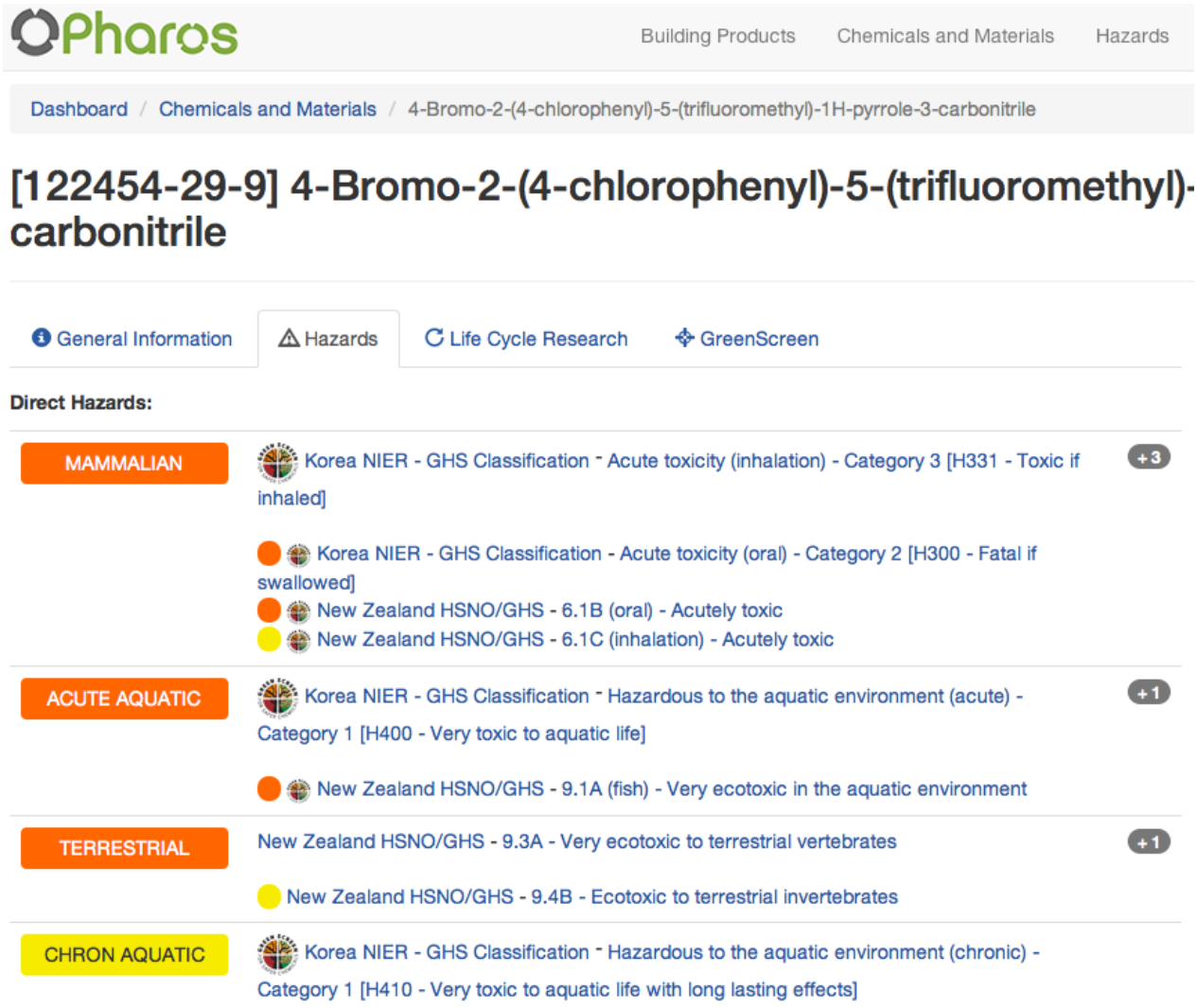
Chemical Name	Final GreenScreen® Benchmark Score
Tralopyril	2

After Data gap Assessment
Note: No Data gap Assessment Done if Preliminary GS Benchmark Score is 1.

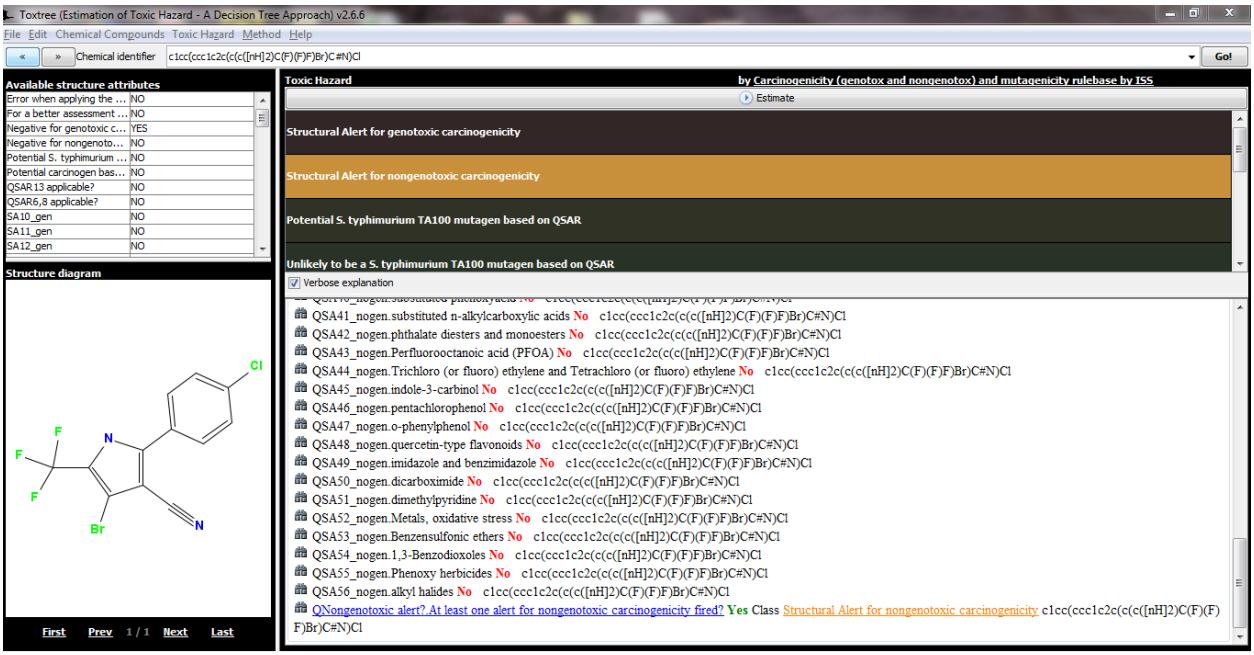
Table 5: Data Gap Assessment Table

Datagap Criteria	a	b	c	d	e	f	g	h	i	j	bm4	End Result
1												
2	Yes	Yes	Yes	Yes	Yes							2
3												
4												

APPENDIX C: Pharos Output for Tralopyril (CAS# 122454-29-9)



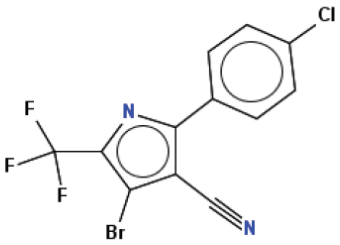
APPENDIX D: Toxtree Carcinogenicity Results for Tralopyril (CAS# 122454-29-9)



APPENDIX E: VEGA Carcinogenicity Results for Tralopyril (CAS# 122454-29-9)

1. Prediction Summary

Prediction for compound 1 (Molecule 1)

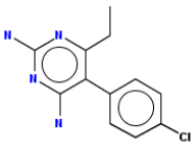


Prediction: Reliability:

Model assessment: Prediction is NON-Carcinogen, but the result may be not reliable. Careful check of the information given in the following section should be done, paying particular attention to the following issues:
- only moderately similar compounds with known experimental value in the training set have been found
- some similar molecules found in the training set have experimental values that disagree with the predicted value
- some atom centered fragments of the compound have not been found in the compounds of the training set or are rare fragments
- predicted substance falls into a neuron that is populated by no compounds of the training set
One or more fragments possibly related to carcinogenic activity were found: Halogenated benzene

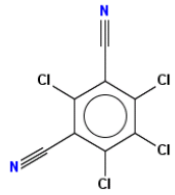
Compound: 1
Compound SMILES: N#Cc1c([nH]c(c1Br)C(F)(F)F)c2ccc(cc2)Cl
Experimental value: -
Prediction: NON-Carcinogen
Carcinogen: 0.35
NON-Carcinogen: 0.65
Structural Alerts: Halogenated benzene
Reliability: Compound is out of model Applicability Domain
Remarks for the prediction:
none

3.1 Applicability Domain:
Similar Compounds, with Predicted and Experimental Values



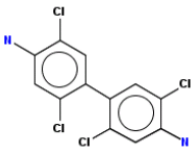
CAS: 58-14-0
Dataset id: 684 (test set)
SMILES: n1c(nc(c(c1N)c2ccc(cc2)Cl)CC)N
Similarity: 0.645

Experimental value: NON-Carcinogen
Predicted value: NON-Carcinogen



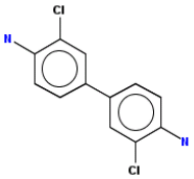
CAS: 1897-45-6
Dataset id: 167 (training set)
SMILES: N#Cc1c(c(C#N)c(c(c1Cl)Cl)Cl)Cl
Similarity: 0.608

Experimental value: Carcinogen
Predicted value: Carcinogen



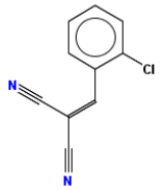
CAS: 15721-02-5
Dataset id: 723 (training set)
SMILES: Nc1cc(c(cc1Cl)c2cc(c(N)cc2Cl)Cl)Cl
Similarity: 0.569

Experimental value: NON-Carcinogen
Predicted value: NON-Carcinogen



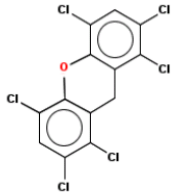
CAS: 91-94-1
Dataset id: 223 (training set)
SMILES: Nc1ccc(cc1Cl)c2ccc(N)c(c2)Cl
Similarity: 0.556

Experimental value: Carcinogen
Predicted value: Carcinogen



CAS: 2698-41-1
Dataset id: 154 (training set)
SMILES: N#CC(C#N)=Cc1ccccc1Cl
Similarity: 0.542

Experimental value: NON-Carcinogen
Predicted value: NON-Carcinogen



CAS: 70-30-4
Dataset id: 363 (training set)
SMILES: Oc1c(cc(c(c1Cc2c(O)c(cc(c2Cl)Cl)Cl)Cl)Cl)Cl
Similarity: 0.534

Experimental value: NON-Carcinogen
Predicted value: NON-Carcinogen

3.2 Applicability Domain:
Measured Applicability Domain Scores

✖	**Global AD Index** AD Index = 0.287 Explanation: predicted substance is outside the Applicability Domain of the model.
⚠	**Similar molecules with known experimental value** Similarity index = 0.626 Explanation: only moderately similar compounds with known experimental value in the training set have been found.
⚠	**Concordance for similar molecules** Concordance index = 0.528 Explanation: some similar molecules found in the training set have experimental values that disagree with the predicted value.
✔	**Accuracy of prediction for similar molecules** Accuracy index = 1 Explanation: accuracy of prediction for similar molecules found in the training set is good.
⚠	**Atom Centered Fragments similarity check** ACF matching index = 0.85 Explanation: some atom centered fragments of the compound have not been found in the compounds of the training set or are rare fragments.
✔	**Model assignment reliability** Pos/Non-Pos difference = 0.309 Explanation: model class assignment is well defined.
✖	**Neural map neurons concordance** NN concordance = 0.5 Explanation: predicted substance falls into a neuron that is populated by no compounds of the training set.
✔	**Model descriptors range check** Descriptors range check = true Explanation: descriptors for this compound have values inside the descriptor range of the compounds of the training set.

Symbols explanation:

✔ The feature has a good assessment, model is reliable regarding this aspect.

⚠ The feature has a non optimal assessment, this aspect should be reviewed by an expert.

✖ The feature has a bad assessment, model is not reliable regarding this aspect.

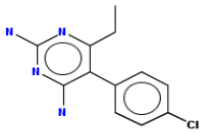
4.1 Reasoning:
Relevant Chemical Fragments and Moieties

(Mol. 1) Reasoning on fragments/structural alerts:

Fragment found: Halogenated benzene

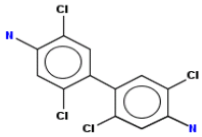
Halogenated benzene (Benigni/Bossa structural alert no. 31).

Following, the most similar compounds from the model's dataset having the same fragment.



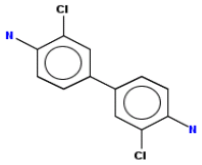
CAS: 58-14-0
Dataset id: 684 (test set)
SMILES: n1c(nc(c(c1N)c2ccc(cc2)Cl)CC)N
Similarity: 0.645

Experimental value: NON-Carcinogen
Predicted value: NON-Carcinogen



CAS: 15721-02-5
Dataset id: 723 (training set)
SMILES: Nc1cc(c(cc1Cl)c2cc(c(N)cc2Cl)Cl)Cl
Similarity: 0.569

Experimental value: NON-Carcinogen
Predicted value: NON-Carcinogen

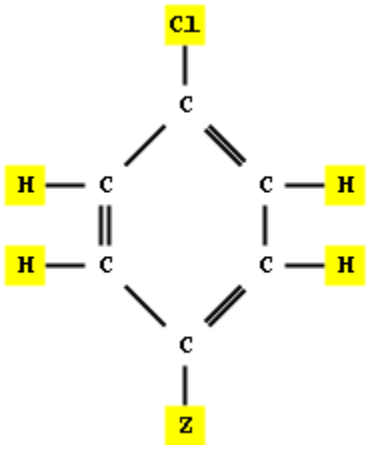


CAS: 91-94-1
Dataset id: 223 (training set)
SMILES: Nc1ccc(cc1Cl)c2ccc(N)c(c2)Cl
Similarity: 0.556

Experimental value: Carcinogen
Predicted value: Carcinogen

APPENDIX F: OncoLogic Output for Tralopyril (CAS #122454-29-9)

OncoLogic Justification Report



SUMMARY :

CODE NUMBER : tralopyril

SUBSTANCE ID :

The level of concern for this compound, disregarding any substituent denoted as 'Z', is LOW-MODERATE.

The effect of any substituent denoted by 'Z' is uncertain.

JUSTIFICATION

Halogenated aromatics
include the following type of halogenated compounds: benzenes, naphthalenes, biphenyls, terphenyls, diphenyl ethers, diphenyl sulfides, dibenzo-p-dioxins, dibenzofurans, dibenzothiophenes, and diphenyl alkanes and alkenes. Although a number of halogenated aromatics have been shown to be carcinogenic in experimental animals, the mechanism of their carcinogenic action is not clearly understood. However, there is a prevalent view that these chemicals may be carcinogenic through epigenetic mechanisms rather than by direct action on DNA. For instance, there is considerable evidence showing that the initial event involved in 2,3,7,8-tetrachlorodibenzo-p-dioxin (TCDD) carcinogenesis is binding to the cytosolic Ah receptor. The subsequent translocation of the TCDD-receptor complex into the nucleus leads to a modulation of gene expression which is believed to be responsible for the various biochemical (e.g. induction of the cytochrome P-450 1A family) and toxicological effects (including tumorigenesis) of the compound. Since the key requirement for the binding of TCDD to the cytosolic Ah receptor is a planar molecule with the halogens at the lateral position (i.e., 2,3,7,8-position of TCDD), it has been suggested that other halogenated aromatics with a molecular shape isosteric with TCDD may act by a mechanism similar to that of TCDD. Indeed, like TCDD, a number of halogenated biphenyls and naphthalenes with halogens at the lateral positions are also inducers of the cytochrome

P-450 1A family.

Other halogenated biphenyls, naphthalenes and
benzenes, which induce the cytochrome P-450 2B family, on the other hand, have been postulated to act via inhibition of "intercellular communication" (also called "metabolic cooperation"). Other epigenetic mechanisms that have been linked to carcinogenesis of halogenated aromatics include (i) hormone imbalance (e.g. estrogen mimics), (ii) immunosuppresion, and (iii) cytotoxicity.

Halogenation
of the aromatics renders them more lipid-soluble, more slowly metabolized, and therefore more persistent in animal tissues. In general, the rate of oxidative metabolism decreases as the degree of halogenation increases because of steric hindrance by the halogen atoms. Moreover, the position of halogenation plays an important role in determining the rate of oxidative metabolism. For instance, it has been shown that chlorinated and brominated benzenes having two adjacent unsubstituted carbon atoms are more rapidly metabolized than those without adjacent unsubstituted carbon atoms, despite a similar degree of halogenation. Hence, in addition to the type of halogens, the degree and position of halogenation are important factors in evaluating the carcinogenicity potential of halogenated aromatics. The carcinogenicity concern levels of these compounds are determined based on structure-activity relationship analysis as well as metabolism and mechanism considerations.

The halogenated benzene with one chloro, bromo, or iodo has a level of concern of LOW-MODERATE.

As a result of the combined substituent modifications, the level of concern remains LOW-MODERATE.

The effect of any other substituent (denoted by 'Z') is uncertain. Therefore, the level of concern remains LOW-MODERATE.

The final level of concern for this compound is LOW-MODERATE.

APPENDIX G: EPISuite Output for Tralopyril Degradation Product, CL322,250

SMILES : c1cc(ccc1c2c(c(c(n2)C(=O)O)Br)C(#N))CL
CHEM :
MOL FOR: C12 H6 Br1 CL1 N2 O2
MOL WT : 325.55
---------- EPI SUMMARY (v4.11) ----------
Physical Property Inputs:
Log Kow (octanol-water): ------
Boiling Point (deg C) : ------
Melting Point (deg C) : ------
Vapor Pressure (mm Hg) : ------
Water Solubility (mg/L): ------
Henry LC (atm-m3/mole) : ------

Log Octanol-Water Partition Coef (SRC):
Log Kow (KOWWIN v1.68 estimate) = 3.61

Boiling Pt, Melting Pt, Vapor Pressure Estimations (MPBPVP v1.43):
Boiling Pt (deg C): 475.24 (Adapted Stein & Brown method)
Melting Pt (deg C): 201.03 (Mean or Weighted MP)
VP(mm Hg,25 deg C): 1.6E-009 (Modified Grain method)
VP (Pa, 25 deg C) : 2.13E-007 (Modified Grain method)
Subcooled liquid VP: 1.15E-007 mm Hg (25 deg C, Mod-Grain method)
: 1.54E-005 Pa (25 deg C, Mod-Grain method)

Water Solubility Estimate from Log Kow (WSKOW v1.42):
Water Solubility at 25 deg C (mg/L): 3.93
log Kow used: 3.61 (estimated)
no-melting pt equation used

Water Sol Estimate from Fragments:
Wat Sol (v1.01 est) = 2.4129 mg/L

ECOSAR Class Program (ECOSAR v1.11):
Class(es) found: Pyrazoles/Pyrroles -acid

Henrys Law Constant (25 deg C) [HENRYWIN v3.20]:
Bond Method : 4.00E-014 atm-m3/mole (4.05E-009 Pa-m3/mole)
Group Method: Incomplete
For Henry LC Comparison Purposes:
User-Entered Henry LC: not entered
Henrys LC [via VP/WSol estimate using User-Entered or Estimated values]:
HLC: 1.744E-010 atm-m3/mole (1.767E-005 Pa-m3/mole)
VP: 1.6E-009 mm Hg (source: MPBPVP)
WS: 3.93 mg/L (source: WSKOWWIN)

Log Octanol-Air Partition Coefficient (25 deg C) [KOAWIN v1.10]:
Log Kow used: 3.61 (KowWin est)

Log Kaw used: -11.786 (HenryWin est)
Log Koa (KOAWIN v1.10 estimate): 15.396
Log Koa (experimental database): None

Probability of Rapid Biodegradation (BIOWIN v4.10):
Biowin1 (Linear Model) : 0.7837
Biowin2 (Non-Linear Model) : 0.8531
Expert Survey Biodegradation Results:
Biowin3 (Ultimate Survey Model): 2.1426 (months)
Biowin4 (Primary Survey Model) : 3.0017 (weeks)
MITI Biodegradation Probability:
Biowin5 (MITI Linear Model) : 0.3981
Biowin6 (MITI Non-Linear Model): 0.0809
Anaerobic Biodegradation Probability:
Biowin7 (Anaerobic Linear Model): 0.3177
Ready Biodegradability Prediction: NO

Hydrocarbon Biodegradation (BioHCwin v1.01):
Structure incompatible with current estimation method!

Sorption to aerosols (25 Dec C)[AEROWIN v1.00]:
Vapor pressure (liquid/subcooled): 1.53E-005 Pa (1.15E-007 mm Hg)
Log Koa (Koawin est): 15.396
Kp (particle/gas partition coef. (m3/ug)):
Mackay model : 0.196
Octanol/air (Koa) model: 611
Fraction sorbed to airborne particulates (phi):
Junge-Pankow model : 0.876
Mackay model : 0.94
Octanol/air (Koa) model: 1

Atmospheric Oxidation (25 deg C) [AopWin v1.92]:
Hydroxyl Radicals Reaction:
OVERALL OH Rate Constant = 3.0420 E-12 cm3/molecule-sec
Half-Life = 3.516 Days (12-hr day; 1.5E6 OH/cm3)
Half-Life = 42.193 Hrs
Ozone Reaction:
No Ozone Reaction Estimation
Reaction With Nitrate Radicals May Be Important!
Fraction sorbed to airborne particulates (phi):
0.908 (Junge-Pankow, Mackay avg)
1 (Koa method)
Note: the sorbed fraction may be resistant to atmospheric oxidation

Soil Adsorption Coefficient (KOCWIN v2.00):
Koc : 574.9 L/kg (MCI method)
Log Koc: 2.760 (MCI method)
Koc : 350.5 L/kg (Kow method)
Log Koc: 2.545 (Kow method)

Aqueous Base/Acid-Catalyzed Hydrolysis (25 deg C) [HYDROWIN v2.00]:
Rate constants can NOT be estimated for this structure!

Bioaccumulation Estimates (BCFBAF v3.01):
Log BCF from regression-based method = 0.500 (BCF = 3.162 L/kg wet-wt)
Log Biotransformation Half-life (HL) = 0.3025 days (HL = 2.007 days)
Log BCF Arnot-Gobas method (upper trophic) = 2.452 (BCF = 283)
Log BAF Arnot-Gobas method (upper trophic) = 2.452 (BAF = 283.3)
log Kow used: 3.61 (estimated)

Volatilization from Water:
Henry LC: 4E-014 atm-m3/mole (estimated by Bond SAR Method)
Half-Life from Model River: 2.641E+010 hours (1.1E+009 days)
Half-Life from Model Lake : 2.881E+011 hours (1.2E+010 days)

Removal In Wastewater Treatment:
Total removal: 15.79 percent
Total biodegradation: 0.21 percent
Total sludge adsorption: 15.58 percent
Total to Air: 0.00 percent
(using 10000 hr Bio P,A,S)

Level III Fugacity Model:

	Mass Amount (percent)	Half-Life (hr)	Emissions (kg/hr)
Air	2.24e-006	84.4	1000
Water	10.1	1.44e+003	1000
Soil	89.5	2.88e+003	1000
Sediment	0.36	1.3e+004	0

Persistence Time: 2.74e+003 hr

Sources to Check for GreenScreen® Hazard Assessment

Note: For a GreenScreen® Hazard Assessment, data queries should be initially limited to the following references. If data gaps exist after these references have been checked, additional references may be utilized.

U.S. EPA High Production Volume Information System (HPVIS):
http://www.epa.gov/hpvis/index.html

UNEP OECD Screening Information Datasets (SIDS):
http://www.chem.unep.ch/irptc/sids/OECDSIDS/sidspub.html

OECD Existing Chemicals Database: http://webnet.oecd.org/hpv/ui/SponsoredChemicals.aspx

European Chemical Substances Information System IUCLID Chemical Data Sheets:
http://esis.jrc.ec.europa.eu/index.php?PGM=dat

National Toxicology Program: http://ntp.niehs.nih.gov/

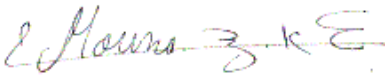
International Agency for the Research on Cancer:
http://monographs.iarc.fr/ENG/Classification/index.php

Human and Environmental Risk Assessment (HERA) on ingredients of household cleaning products:
http://www.heraproject.com/RiskAssessment.cfm

European Chemicals Agency (ECHA) REACH Dossiers: http://echa.europa.eu/

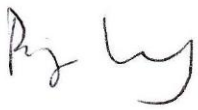
Licensed GreenScreen® Profilers

Tralopyril GreenScreen® Evaluation Prepared By:



Mouna Zachary, Ph.D.
Toxicologist
ToxServices LLC

Tralopyril GreenScreen® Evaluation QC'd By:



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