ERUCAMIDE

(CAS #112-84-5)

GREENSCREEN® FOR SAFER CHEMICALS (GREENSCREEN®) ASSESSMENT

Prepared by:

ToxServices LLC

Assessment Date: November 12, 2021

Expiration Date: November 12, 2026

TOXSERVICES
TOXICOLOGY RISK ASSESSMENT CONSULTING
1367 Connecticut Ave., N.W., Suite 300
Washington, D.C. 20036

TABLE OF CONTENTS

GreenScreen® Executive Summary for Erucamide (CAS #112-84-5) i
Chemical Name 1
GreenScreen® Summary Rating for Erucamide: 2
Environmental Transformation Products 3
Introduction 3
U.S. EPA Safer Choice Program's Safer Chemical Ingredients List 3
GreenScreen® List Translator Screening Results 3
Hazard Statement and Occupational Control 4
Physicochemical Properties of Erucamide 4
Toxicokinetics 5
Hazard Classification Summary 5
Group I Human Health Effects (Group I Human) 5
Carcinogenicity (C) Score 5
Mutagenicity/Genotoxicity (M) Score 6
Reproductive Toxicity (R) Score 7
Developmental Toxicity incl. Developmental Neurotoxicity (D) Score 7
Endocrine Activity (E) Score 8
Group II and II* Human Health Effects (Group II and II* Human) 8
Acute Mammalian Toxicity (AT) (Group II) Score 8
Systemic Toxicity/Organ Effects incl. Immunotoxicity (ST-single) (Group II) Score 9
Systemic Toxicity/Organ Effects incl. Immunotoxicity (ST-repeat) (Group II*) Score 10
Neurotoxicity (single dose, N-single) (Group II) Score 11
Neurotoxicity (repeated dose, N-repeated) (Group II*) Score 12
Skin Sensitization (SnS) (Group II*) Score 12
Respiratory Sensitization (SnR) (Group II*) Score 13
Skin Irritation/Corrosivity (IrS) (Group II) Score 13
Eye Irritation/Corrosivity (IrE) (Group II) Score 13
Ecotoxicity (Ecotox) 14
Acute Aquatic Toxicity (AA) Score 14
Chronic Aquatic Toxicity (CA) Score 14
Environmental Fate (Fate) 15
Persistence (P) Score 15
Bioaccumulation (B) Score 16
Physical Hazards (Physical) 16
Reactivity (Rx) Score 16
Flammability (F) Score 16

Use of New Approach Methodologies (NAMs) in the Assessment, Including Uncertainty Analyses of Input and Output 18
References 21
APPENDIX A: Hazard Classification Acronyms 23
APPENDIX B: Results of Automated GreenScreen® Score Calculation for Erucamide (CAS #112-84-5) 24
APPENDIX C: Pharos Output for Erucamide (CAS #112-84-5) 25
APPENDIX D: Toxtree Carcinogenicity Results for Erucamide (CAS #112-84-5) 26
APPENDIX E: VEGA Carcinogenicity Modeling Results for Erucamide (CAS #112-84-5) 27
APPENDIX F: Danish QSAR Predictions Output for Erucamide (CAS #112-84-5) 46
APPENDIX G: OECD Toolbox Respiratory Sensitization Results for Erucamide (CAS #112-84-5) 49
APPENDIX H: EPI Suite™ Modeling Results for Erucamide (CAS #112-84-5) 50
APPENDIX I: Known Structural Alerts for Reactivity 54
APPENDIX J: Change in Benchmark Score 58
Licensed GreenScreen® Profilers 59

TABLE OF FIGURES

Figure 1: GreenScreen® Hazard Summary Table for Erucamide 3

TABLE OF TABLES

Table 1: Occupational Exposure Limits and Recommended Personal Protective Equipment for Erucamide (CAS #112-84-5) 4
Table 2: Physical and Chemical Properties of Erucamide (CAS #112-84-5) 4
Table 3: Summary of NAMs Used in the GreenScreen® Assessment, Including Uncertainty Analyses 18
Table 4: Change in GreenScreen® Benchmark™ for Erucamide 58

GreenScreen® Executive Summary for Erucamide (CAS #112-84-5)

Erucamide is a C22 unsaturated fatty acid amide with a double bond at C13. It functions as a foam stabilizer, a solvent for waxes and resin, an emulsifier, an antiblock agent for polyethylene, and an adherent. Erucamide is a solid at room temperature. Its low vapor pressure indicates that it is unlikely to volatilize. It has very low water solubility, is hydrophobic, and is not reactive or flammable.

Erucamide was assigned a **GreenScreen Benchmark™ Score of 2_{DG}** ("Use but Search for Safer Substituents" due to data gaps). Prior to data gap analysis, it was assigned a preliminary benchmark score of 4 ("Prefer – Safer Chemical"). This score is based on the following hazard score combinations:

- Benchmark 4 (lowered to Benchmark 2_{DG} because of data gaps)
    - Low Group I Human Toxicity (carcinogenicity-C, mutagenicity-M and developmental toxicity-D)
    - Low Group II Human Toxicity (acute toxicity-AT, single dose systemic toxicity-STs, single dose neurotoxicity-Ns, skin irritation-IrS and eye irritation-IrE)
    - Low Group II* Human Toxicity (repeated dose systemic toxicity-STr*, repeated dose neurotoxicity-Nr*, skin sensitization-SnS*, and respiratory sensitization-SnR*)
    - Low Ecotoxicity (acute aquatic-AA and chronic aquatic-CA)
    - Low Fate Concerns (persistence-P and bioaccumulation-B)
    - Low Physical Hazards (reactivity-R and flammability-F)

Data gaps (DG) exist for reproductive toxicity-R and endocrine activity-E. As outlined in GreenScreen® Guidance Section 11.6.2.1 and Annex 5 (Conduct a Data Gap Analysis), erucamide does not meet the requirement of a Benchmark 4 or 3, however it meets requirements for a GreenScreen Benchmark™ Score of 2 due to hazard data gaps. In a worst-case scenario, if erucamide were assigned a High score for the data gaps R or E, it would be categorized as a Benchmark 1 Chemical.

New Approach Methodologies (NAMs) used in this GreenScreen® include *in silico* modeling for carcinogenicity, endocrine activity, respiratory sensitization, and bioaccumulation, and *in vitro* testing for genotoxicity. The quality, utility, and accuracy of NAM predictions are greatly influenced by two primary types of uncertainties:

- Type I: Uncertainties related to the input data used
- Type II: Uncertainties related to extrapolations made

Type I (input data) uncertainties in erucamide's NAMs dataset include lack of data on carcinogenicity, endocrine activity, respiratory sensitization, and bioaccumulation. Erucamide's Type II (extrapolation output) uncertainties include limitations of modeling software Toxtree and OECD Toolbox in identifying structural alerts without defining applicability domains, the limitations of *in vitro* genotoxicity assays in mimicking *in vivo* metabolic systems and the uncertain *in vivo* relevance of *in silico* modeling of endocrine receptor binding. Some of erucamide's type II uncertainties were alleviated by the use of *in vitro* test batteries and/or in combination of *in vivo* data.

GreenScreen® Hazard Summary Table for Erucamide

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**
						s	r*	s	r*	*	*								
L	**L**	DG	**L**	DG	**L**	**L**	**L**	*L*	**L**	**L**	*L*	**L**	**L**	*L*	**L**	**L**	*L*	*L*	**L**

Note: Hazard levels (Very High (vH), High (H), Moderate (M), Low (L), Very Low (vL)) in *italics* reflect lower confidence in the hazard classification while hazard levels in **BOLD** font reflect higher confidence in the hazard classification. 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. Group II* Human Health endpoints are indicated by an * after the name of the hazard endpoint or after "repeat" for repeated exposure sub-endpoints. Please see Appendix A for a glossary of hazard acronyms.

GreenScreen® Chemical Assessment for Erucamide (CAS #112-84-5)

Method Version: GreenScreen® Version 1.4
Assessment Type[1]: Certified
Assessor Type: Licensed GreenScreen® Profiler

GreenScreen® Assessment (v.1.1) Prepared By:
Name: Kristen Schaefer, M.F.S.
Title: Associate Toxicologist
Organization: ToxServices LLC
Date: April 28, 2011

Quality Control Performed By:
Name: Dr. Margaret H. Whittaker, Ph.D., M.P.H., CBiol., F.R.S.B., E.R.T., D.A.B.T.
Title: Managing Director and Chief Toxicologist
Organization: ToxServices LLC
Date: April 28, 2011

GreenScreen® Assessment (v.1.2) Updated By:
Name: Sara M. Ciotti, Ph.D.
Title: Toxicologist
Organization: ToxServices LLC
Date: February 19, 2016

Quality Control Performed By:
Name: Bingxuan Wang, Ph.D., D.A.B.T.
Title: Toxicologist
Organization: ToxServices LLC
Date: February 19, 2016

GreenScreen® Assessment (v.1.4) Updated By:
Name: Megan B. Boylan, M.S.
Title: Toxicologist
Organization: ToxServices LLC
Date: August 11, 2021

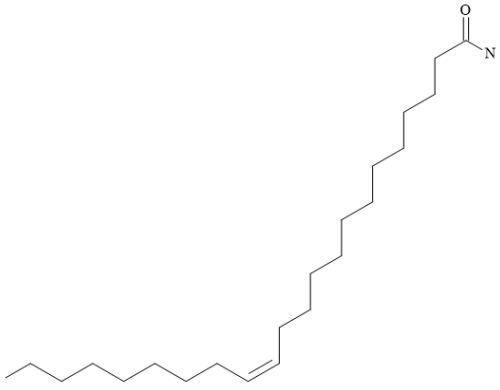
Quality Control Performed By:
Name: Bingxuan Wang, Ph.D., D.A.B.T.
Title: Senior Toxicologist
Organization: ToxServices LLC
Date: August 12, 2021, November 12, 2021

Expiration Date: November 12, 2026[2]

Chemical Name: Erucamide

CAS Number: 112-84-5

Chemical Structure(s):



[1] GreenScreen® reports are either "UNACCREDITED" (by unaccredited person), "AUTHORIZED" (by Authorized GreenScreen® Practitioner), or "CERTIFIED" (by Licensed GreenScreen® Profiler or equivalent).
[2] Assessments expire five years from the date of completion starting from January 1, 2019. An assessment expires three years from the date of completion if completed before January 1, 2019 (CPA 2018a).

Also called: 13-Docosenamide; Erucic acid amide; Erucyl amide; 13-Docosenamide, (Z)-; Erucylamide; (Z)-Docos-13-enamide; 13-Docosenamide, (13Z)- (ChemIDplus 2021).

Suitable surrogates or moieties of chemicals used in this assessment (CAS #'s):
No surrogates were identified for the data gaps for erucamide. Therefore, no chemical surrogates were used in this GreenScreen®.

Identify Applications/Functional Uses:
1. Foam stabilizer
2. Solvent for waxes and resins
3. Emulsifier
4. Antiblock agent for polyethylene
5. Opacifying agent
6. Adherent
7. Viscosity controlling agent
(Pharos 2021)

Known Impurities[3]:
No information is available. The screen is performed on the theoretical pure substance.

GreenScreen® Summary Rating for Erucamide:[4,5 6,7] Erucamide was assigned a **GreenScreen Benchmark™ Score of 2_{DG}** ("Use but Search for Safer Substituents" due to data gaps). Prior to data gap analysis, it was assigned a preliminary benchmark score of 4 ("Prefer – Safer Chemical") (CPA 2018b). This score is based on the following hazard score combinations:

- Benchmark 4 (lowered to Benchmark 2_{DG} because of data gaps)
    - Low Group I Human Toxicity (carcinogenicity-C, mutagenicity-M and developmental toxicity-D)
    - Low Group II Human Toxicity (acute toxicity-AT, single dose systemic toxicity-STs, single dose neurotoxicity-Ns, skin irritation-IrS and eye irritation-IrE)
    - Low Group II* Human Toxicity (repeated dose systemic toxicity-STr*, repeated dose neurotoxicity-Nr*, skin sensitization-SnS*, and respiratory sensitization-SnR*)
    - Low Ecotoxicity (acute aquatic-AA and chronic aquatic-CA)
    - Low Fate Concerns (persistence-P and bioaccumulation-B)
    - Low Physical Hazards (reactivity-R and flammability-F)

Data gaps (DG) exist for reproductive toxicity-R and endocrine activity-E. As outlined in GreenScreen® Guidance Section 11.6.2.1 and Annex 5 (Conduct a Data Gap Analysis), erucamide does not meet the requirement of a Benchmark 4 or 3, however it meets requirements for a GreenScreen Benchmark™ Score of 2 due to hazard data gaps. In a worst-case scenario, if erucamide were assigned a High score for the data gaps R or E, it would be categorized as a Benchmark 1 Chemical.

[3] Impurities of the chemical will be assessed at the product level instead of in this GreenScreen®.
[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.
[5] See Appendix A for a glossary of hazard endpoint acronyms.
[6] For inorganic chemicals only, see GreenScreen® Guidance v1.4 Section 12 (Inorganic Chemical Assessment Procedure).
[7] For Systemic Toxicity and Neurotoxicity, repeated exposure data are preferred. Lack of single exposure data is not a Data Gap when repeated exposure data are available. In that case, lack of single exposure data may be represented as NA instead of DG. See GreenScreen® Guidance v1.4 Annex 2.

Figure 1: GreenScreen® Hazard Summary Table for Erucamide

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
						s	r*	s	r*	*	*								
L	**L**	DG	**L**	DG	**L**	**L**	**L**	*L*	**L**	**L**	*L*	**L**	**L**	*L*	**L**	**L**	*L*	*L*	**L**

Note: Hazard levels (Very High (vH), High (H), Moderate (M), Low (L), Very Low (vL)) in *italics* reflect lower confidence in the hazard classification while hazard levels in **BOLD** font reflect higher confidence in the hazard classification. 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. Group II* Human Health endpoints are indicated by an * after the name of the hazard endpoint or after "repeat" for repeated exposure sub-endpoints. Please see Appendix A for a glossary of hazard acronyms.

Environmental Transformation Products

Per GreenScreen® guidance (CPA 2018b), chemicals that degrade rapidly and completely (i.e., meet criteria for a Very Low or Low for persistence) are not likely to form persistent biodegradation intermediates because the degradation intermediates will not persist long enough to be encountered after use or release of the parent chemical (i.e., relevant). As erucamide is readily biodegradable, it is not expected to have relevant transformation products.

Introduction

Erucamide is a C22 unsaturated fatty acid amide with a double bond at C13. It functions as a foam stabilizer, a solvent for waxes and resin, an emulsifier, an antiblock agent for polyethylene, and an adherent (Pharos 2021). It is produced by the reaction of erucic acid with anhydrous ammonia (PubChem 2021).

ToxServices assessed erucamide against GreenScreen® Version 1.4 (CPA 2018b) following procedures outlined in ToxServices' SOPs (GreenScreen® Hazard Assessment) (ToxServices 2020).

U.S. EPA Safer Choice Program's Safer Chemical Ingredients List

The SCIL is a list of chemicals that meet the Safer Choice standard (U.S. EPA 2021a). It can be accessed at: http://www2.epa.gov/saferchoice/safer-ingredients. Chemicals on the SCIL have been assessed for compliance with the Safer Choice Standard and Criteria for Safer Chemical Ingredients (U.S. EPA 2015).

Erucamide is not listed on the SCP SCIL.

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 2018b). Pharos (Pharos 2021) is an online list-searching tool that is used to screen chemicals against all of the lists in the List Translator electronically. ToxServices also checks the U.S. Department of Transportation (U.S. DOT) lists (U.S. DOT 2008a,b),[8] which are not considered GreenScreen® Specified Lists but are additional information sources, in conjunction with the Pharos query. The output indicates benchmark or possible benchmark scores for each human health and environmental endpoint. The output for erucamide can be found in Appendix C.

[8] DOT lists are not required lists for GreenScreen® List Translator v1.4. They are reference lists only.

- Erucamide is an LT-UNK chemical when screened using Pharos, and therefore a full GreenScreen® is required.
- Erucamide is not listed on the U.S. DOT list.
- Erucamide is on the following lists for multiple endpoints:
  - German FEA Class 1 – Low Hazard to Waters, and
  - EC – CEPA DSL Inherently Toxic in Humans (iTH).
- Specified lists for single endpoints are reported in individual hazard endpoints in the hazard assessment section below.

Hazard Statement and Occupational Control

There are no EU harmonized Globally Harmonized System of Classification and Labelling of Chemicals (GHS) hazard statements for erucamide, and the majority of EU notifiers and its REACH dossier authors did not self-classify it with any hazards (ECHA 2021a,b). General personal protective equipment (PPE) recommendations are presented in Table 1, below. No occupational exposure limits (OELs) were identified.

Table 1: Occupational Exposure Limits and Recommended Personal Protective Equipment for Erucamide (CAS #112-84-5)

Personal Protective Equipment (PPE)	Reference	Occupational Exposure Limits (OEL)	Reference
Respiratory: use dust mask	ECHA 2021b	None identified	
Hand: protective gloves of neoprene or synthetic rubber			
Eye: wear safety goggles			
Skin: wear suitable protective clothing			

Physicochemical Properties of Erucamide

Erucamide is a solid at room temperature. Its low vapor pressure indicates that it is unlikely to volatilize. It has very low water solubility and its log K_{ow} of 8 indicates that it is hydrophobic.

Table 2: Physical and Chemical Properties of Erucamide (CAS #112-84-5)

Property	Value	Reference
Molecular formula	$C_{22}H_{43}NO$	ChemIDplus 2021
SMILES Notation	CCCCCCCC\C=C/CCCCCCCCCCCC(=O)N	ChemIDplus 2021
Molecular weight	337.588 g/mol	ChemIDplus 2021
Physical state	Solid	ECHA 2021b
Appearance	Off-white organic powder with a fatty odor	ECHA 2021b
Melting point	64-83°C	ECHA 2021b
Boiling point	461.05°C (estimated)	U.S. EPA 2017
Vapor pressure	8.28×10^{-8} mm Hg @ 25°C (estimated)	U.S. EPA 2017
Water solubility	<0.738 µg/L @ 20°C	ECHA 2021b
Dissociation constant	N/A due to a lack of ionizable groups	ECHA 2021b
Density/specific gravity	0.908 @ 20°C	ECHA 2021b
Partition coefficient	Log K_{ow} = 8	ECHA 2021b

Toxicokinetics

Erucamide was tested in a 4-week digestibility study in Sprague-Dawley rats, where 10% erucamide was administered in a semi-synthetic diet without any added fat. Weekly fecal samples were weighed and analyzed for fat content; the analysis showed that 52.8-72.9% erucamide was absorbed from the gastrointestinal tract. Its absorption could be limited as it is expected to undergo hydrolysis to erucic acid and ammonia in the gastrointestinal tract. Erucamide was found to be efficiently hydrolyzed by rat liver homogenate, with 37.6% hydrolyzed in four hours (Klimisch 2, reliable with restrictions) (Health Canada 2019, ECHA 2021b).

Due to the low vapor pressure, inhalation is not expected to be a significant route of exposure (Health Canada 2019).

No additional toxicokinetics data are identified for erucamide.

Hazard Classification Summary

Group I Human Health Effects (Group I Human)

Carcinogenicity (C) Score (H, M, or L): *L*

Erucamide was assigned a score of Low for carcinogenicity based on the lack of structural alerts in Toxtree, a weight of evidence from reliable predictions of three VEGA models, and negative and in domain results from all models in the Danish QSAR database. ToxServices also attempted to use OncoLogic to evaluate erucamide, but the program is not capable of evaluating its structure. GreenScreen® criteria classify chemicals as a Low hazard for carcinogenicity when adequate data are available and they are not classified under GHS (CPA 2018b). The confidence in the score is reduced as it is based on modeled data.

- Authoritative and Screening Lists
  - *Authoritative:* Not present on any authoritative lists for this endpoint.
  - *Screening:* Not present on any screening lists for this endpoint.
- Toxtree 2018
  - Erucamide contains no structural alerts for genotoxic and non-genotoxic carcinogenicity. See Appendix D for modeling results.
- VEGA 2021
  - CAESAR Carcinogenicity Model (v.2.1.9) predicts that the compound is a non-carcinogen with moderate reliability. An applicability domain (AD) index of 0.761 is calculated (Appendix E), indicating that the prediction is reliable.
  - ISS Carcinogenicity Model (v.1.0.2) predicts that the compound is a non-carcinogen with low reliability. An AD index of 0.623 is calculated (Appendix E), indicating that the prediction is not reliable.
  - IRFMN/Antares Carcinogenicity Model (v.1.0.0) predicts that the compound is a non-carcinogen with strong reliability. An AD index of 0.935 is calculated (Appendix E), indicating that the prediction is reliable.
  - IRFMN/ISSCAN-CGX Carcinogenicity Model (v.1.0.0) predicts that the compound is a carcinogen with moderate reliability. An AD index of 0.716 is calculated (Appendix E), indicating that the prediction is reliable.
    - *ToxServices notes that none of the read-across chemicals are amides, and some of them contain structural alerts for carcinogenicity that are not present in erucamide. Therefore, the reliability of this prediction is questionable.*

    - IRFMN oral classification model (v.1.0.0) predicts that the compound is a carcinogen with moderate reliability. An AD index of 0.60 is calculated (Appendix E), indicating that the prediction is unreliable.
    - IRFMN inhalation classification model (v.1.0.0) predicts that the compound is a carcinogen with low reliability. An AD index of 0.00 is calculated (Appendix E), indicating that the prediction is unreliable.
- DTU 2021
    - All seven FDA RCA cancer models within E Ultra (i.e., male rat, female rat, rat, male mouse, female mouse, mouse, and rodent) predicts erucamide to be negative, and all predictions are in domain. Similarly, all seven FDA RCA cancer models within Leadscope (i.e., male rat, female rat, rat, male mouse, female mouse, mouse, and rodent) predicts erucamide to be negative, and all predictions are in domain (Appendix F).
    - The liver specific cancer in rat or mouse model battery predicts erucamide to be negative, based on negative and in domain results by Case Ultra and SciQSAR (Appendix F).
- The rule-based models Toxtree and IRFMN/Antares, and the statistically based models CAESAR and Danish QSAR all predict erucamide to be negative for carcinogenicity. The predictions from the remaining models are not reliable. Therefore, the overall weight of evidence suggests erucamide is not carcinogenic.

Mutagenicity/Genotoxicity (M) Score (H, M, or L): L

Erucamide was assigned a score of Low for mutagenicity/genotoxicity based on negative results for mutagenicity and clastogenicity in *in vitro* studies. GreenScreen® criteria classify chemicals as a Low hazard for mutagenicity/genotoxicity when negative data are available for both gene mutations and chromosome aberrations, and they are not GHS classified (CPA 2018b). The confidence in the score is high as it is based on reliable experimental data.

- Authoritative and Screening Lists
    - *Authoritative:* Not present on any authoritative lists for this endpoint.
    - *Screening:* Not present on any screening lists for this endpoint.
- ECHA 2021b
    - *In vitro:* Negative results for mutagenicity were obtained in a GLP-compliant bacterial reverse mutation assay according to OECD Guideline 471. *Salmonella typhimurium* test strains TA98, TA100, TA1535, TA1537, and TA1538 were exposed to erucamide (97% purity in acetone) at concentrations up to 5,000 µg/plate both in the presence and absence of metabolic activation. No increases in the mutation frequency were observed in the presence and absence of metabolic activation (Klimisch 1, reliable without restriction).
    - *In vitro:* Negative results for mutagenicity were obtained in a GLP-compliant mouse lymphoma forward mutation assay conducted according to OECD Guideline 476. Mouse lymphoma L5178Y ($TK^{+/-}$) cells were exposed to erucamide (99.2% purity in acetone) at concentrations up to 1,200 µg/mL both in the presence and absence of metabolic activation. No increases in mutation frequency were observed in the presence and absence of metabolic activation (Klimisch 1, reliable without restriction).
    - *In vitro:* Negative results for clastogenicity were obtained in a GLP-compliant chromosomal aberration assay conducted according to OECD Guideline 473. Chinese hamster lung fibroblasts (V79) were exposed to erucamide (99.2% in acetone) at concentrations up to 1,250 µg/mL both in the presence and absence of metabolic activation. There was no increase in structural chromosomal aberrations seen at all dose levels in the presence and absence of metabolic activation (Klimisch 1, reliable without restriction).

Reproductive Toxicity (R) Score (H, M, or L): DG
Erucamide was assigned a score of Data Gap for reproductive toxicity based on a lack of reproductive toxicity studies. The absence of adverse effects on reproductive parameters in a developmental toxicity study in rats (see developmental toxicity section below) and no treatment-related effects on estrous cycle, sperm parameters, or reproductive organ weights in a 90-day oral toxicity study in rats were insufficient to warrant a Low score, as reproduction function/performance was not tested under exposure conditions.

- Authoritative and Screening Lists
  - *Authoritative:* Not present on any authoritative lists for this endpoint.
  - *Screening:* Not present on any screening lists for this endpoint.
- ECHA 2021b
  - *Oral:* In a GLP-compliant repeated dose toxicity study conducted according to OECD Guideline 408, male and female Wistar rats (10/sex/dose) received 100, 300, and 1,000 mg/kg/day erucamide (purity not reported) in corn oil via gavage for 90 days. The estrous cycle of females was evaluated over a period of 8 days on days 4, 8, and 12. Sperm parameters (sperm motility, testicular sperm count, and sperm morphology) were evaluated at necropsy. Additionally, reproductive organ weights (testes, epididymides, ovaries, and uterus with cervix) were recorded following sacrifice. No treatment-related effects were found. Authors identified a NOAEL of 1,000 mg/kg/day, which was the highest dose tested (Klimisch 1, reliable without restriction).

Developmental Toxicity incl. Developmental Neurotoxicity (D) Score (H, M, or L): L
Erucamide was assigned a score of Low for developmental toxicity based on the absence of developmental toxicity effects in an OECD Guideline 414 study in rats. GreenScreen® criteria classify chemicals as a Low hazard for developmental toxicity when adequate negative data are available and they are not GHS classified (CPA 2018b). The confidence in the score is high as it is based on reliable experimental data.

- Authoritative and Screening Lists
  - *Authoritative:* Not present on any authoritative lists for this endpoint.
  - *Screening:* Not present on any screening lists for this endpoint.
- ECHA 2021b
  - A GLP-compliant OECD Guideline 414 study to evaluate the developmental toxicity of erucamide was conducted and is described in the Reproductive Toxicity section above. One untreated male was cohoused with every two females; gestational day (GD) 0 was defined as the date on which sperm could be observed in a vaginal smear. Female Wistar Crl:WI(Han) rats (20-22/dose) were administered test erucamide (100% active) dissolved in corn oil via oral gavage at doses 100, 300, or 1,000 mg/kg body weight daily on GD 5-19. Animals were evaluated for general indications of intoxication twice per day, and received a more in-depth evaluation once daily. The latter evaluation included observations for activity, behavior, response to handling, body position, convulsions or tremors, breathing abnormalities, vocalization, diarrhea, and abnormalities in skin, eyes, fur, or mucous membranes. Sperm-positive females were weighed and assessed for food consumption relative to baseline at GD 5, 8, 11, 14, 17, and 20. On GD 20, females were euthanized and subjected to necropsy. Investigators closely examined the organs and body cavities to identify abnormalities, and uterine contents were evaluated. Non-pregnant females' uteri were removed and stained to check for evidence of resorption. Fetuses were euthanized *in utero*, excised, and examined for signs of toxicity. Parameters evaluated included numbers of live and dead fetuses, sex ratio, litter weights, male/female litter weights, external

morphology, soft tissue characteristics, skeletal morphology, and brain/cranium characteristics. Minor effects were observed in a non-dose responsive pattern, including discoloration of the left lobe of the liver in multiple offspring in the middle dosage group and incomplete ossification of the interparietal bone in the lowest dosage group. The investigators did not consider these effects test material-related and reported a NOAEL of 1,000 mg/kg/day for maternal and developmental toxicity (Klimisch 1, reliable without restriction).

Endocrine Activity (E) Score (H, M, or L): DG
Erucamide was assigned a score of Data Gap for endocrine activity based on a lack of sufficient data for this endpoint. While *in silico* modeling does not suggest a concern for interaction with the estrogen, androgen and thyroid pathways, no *in vivo* data are available that measured endocrine levels.

- Authoritative and Screening Lists
  - *Authoritative:* Not present on any authoritative lists for this endpoint.
  - *Screening:* Not present on any screening lists for this endpoint.
- U.S. EPA 2021b
  - Erucamide is predicted to be inactive for androgen agonism, antagonism and binding by the ToxCast COMPARA (consensus) model.
- DTU 2021 (only results that are in domain are described below)
  - Erucamide and its predicted metabolites have no structural alerts for estrogen receptor binding (Appendix F).
  - Erucamide is predicted to be negative for androgen receptor inhibition by the model battery consisting of negative and in domain predictions by Leadscope and SciQSAR (Appendix F).
  - Erucamide is predicted to be negative for androgen receptor activation, CoMPARA data (*in vitro*) by the Leadscope model (Appendix F).
  - Erucamide is predicted to be negative for thyroperoxidase (TPO) inhibition QSAR 1 and QSAR 2 (rat *in vitro*) models in Leadscope (Appendix F).

Group II and II* Human Health Effects (Group II and II* Human)
Note: Group II and Group II* endpoints are distinguished in the v 1.4 Benchmark system (the asterisk indicates repeated exposure). For Systemic Toxicity and Neurotoxicity, Group II and II* are considered sub-endpoints. See GreenScreen® Guidance v1.4, Annex 2 for more details.

Acute Mammalian Toxicity (AT) (Group II) Score (vH, H, M, or L): L
Erucamide was assigned a score of Low for acute toxicity based on oral and dermal LD_{50} values >2,000 mg/kg. GreenScreen® criteria classify chemicals as a Low hazard for acute toxicity when oral and dermal LD_{50} values are >2,000 (CPA 2018b). The confidence in the score is high as it is based on reliable experimental data.

- Authoritative and Screening Lists
  - *Authoritative:* Not present on any authoritative lists for this endpoint.
  - *Screening:* Not present on any screening lists for this endpoint.
- ECHA 2021b
  - *Oral:* LD_{50} >2,500 mg/kg, GLP-compliant, OECD Guideline 423, Sprague-Dawley rat, male and female (Klimisch 1, reliable without restriction)
  - *Oral:* LD_{50} = 5,000 mg/kg, GLP-compliant, EU Method B.1, Wistar rats, male and female (Klimisch 2, reliable with restrictions)
  - *Oral:* LD_{50} >2,000 mg/kg, GLP-compliant, GLP-compliant, OECD Guideline 401, Sprague-Dawley rats, male and female (Klimisch 2, reliable with restrictions)

- *Dermal:* LD_{50} >2,000 mg/kg, GLP-compliant, OECD Guideline 402, RccHan:WIST rats, male and female (Klimisch 1, reliable without restriction)
- *Inhalation (dust):* Nose only LC_{50} (4-hr) >2.8 mg/L, GLP-compliant, OECD Guideline 436, RccHan:WIST rats, male and female (Klimisch 1, reliable without restriction)

Systemic Toxicity/Organ Effects incl. Immunotoxicity (ST-single) (Group II) Score (vH, H, M, or L): L

Erucamide was assigned a score of Low for systemic toxicity (single dose) based on the absence of adverse systemic toxic effects following acute oral, dermal, and inhalation exposures. GreenScreen® criteria classify chemicals as a Low hazard for systemic toxicity (single dose) when adequate data are available, studies are negative, and it is not GHS classified (CPA 2018b). The confidence in the score is high as it is based on reliable experimental data.

- Authoritative and Screening Lists
  - *Authoritative:* Not present on any authoritative lists for this endpoint.
  - *Screening:* Not present on any screening lists for this endpoint.
- ECHA 2021b
  - *Oral:* In a GLP-compliant study conducted according to OECD Guideline 423, investigators exposed groups of male and female Sprague-Dawley rats (3/sex) to erucamide (purity not reported) via gavage. Test material was dissolved in peanut oil such that all animals received 2,000 mg/kg-bw in 10 mL/kg body weight. Investigators evaluated animals for overt signs of toxicity at post-exposure hours 0.5, 1, 2, and 4; animals were examined again daily for the course of the 14-day study. After two weeks had elapsed, all animals were euthanized and subjected to a gross pathological examination. No deaths were observed, nor were any gross pathological changes found in the exposed animals. The authors established an LD_{50} >2,500 mg/kg, as no deaths were observed at 2,000 mg/kg (Klimisch 1, reliable without restriction).
    - *ToxServices notes that only a dose of 2,000 mg/kg was administered, and it is not clear why the LD_{50} was determined to be > 2,500 mg/kg.*
  - *Oral:* A GLP-compliant study conducted according to OECD Guideline 401 found no deaths or adverse effects on clinical signs, body weight development, and gross pathology examinations when male and female Sprague-Dawley rats (5/sex) received erucamide in corn oil via gavage at 2,000 mg/kg. The authors established an LD_{50} >2,000 mg/kg (Klimisch 2, reliable with restrictions).
  - *Oral:* A GLP-compliant study conducted according to EU Method B.1 was carried out to assess oral toxicity of erucamide. Male and female Wistar rats (5/sex) received two doses of 2,500 mg/kg erucamide (purity not reported) within 24 hours. Animals were observed for 14 days, and then euthanized and subjected to macroscopic evaluation. No deaths, adverse effects during the observation period, or macroscopic abnormalities were noted. The authors established an LD_{50} >5,000 mg /kg (Klimisch 2, reliable with restrictions).
  - *Dermal:* In a GLP-compliant study conducted according to OECD Guideline 402, investigators assessed the acute dermal toxicity of erucamide in male and female Wistar rats. Investigators clipped the hair from the backs of the test animals (5/sex) the night before the experiment was slated to begin. Animals were checked to have fully intact, non-irritated skin before test material was applied on study day 1. Test material, in the form of a powder, was applied to moistened gauze pads which were placed onto the backs of the experimental animals and held in place with semi-occlusive dressings. All animals received 2,000 mg erucamide/kg. The dressings and test material were removed 24 hours after application, and then animals were washed gently with warm water and dried with paper towels. Animals

were evaluated for signs of intoxication and viability at post-exposure hours 0.5, 1, 2, 3, 5, and twice daily starting at post-exposure hour 24. No deaths were observed, and body weights stayed in the normal range for animals of the relevant age and strain. There were no clinical signs of toxicity or effects on gross pathology. No skin damage was observed except for minor peeling at days 7 and 8 in one male animal. Authors established an LD_{50} >2,000 mg/kg (Klimisch 1, reliable without restriction).

- *Inhalation:* In a GLP-compliant study conducted according to OECD Guideline 436, male and female Wistar rats were exposed to erucamide (99.51% purity) via inhalation of a dust. Animals (3/sex) were fitted with nose-only, flow-past exposure systems, and received 2.8 mg test material/L air continuously for four hours. This concentration was deemed the highest feasible concentration that would generate respirable dusts without agglomerating, and no deaths or adverse effects on clinical signs, body weight, or gross pathology were noted over the 14-day observation period or at necropsy. Authors established an LC_{50} >2.8 mg/L/4 hr (Klimisch 1, reliable without restriction).

Systemic Toxicity/Organ Effects incl. Immunotoxicity (ST-repeat) (Group II*) Score (H, M, or L): L

Erucamide was assigned a score of Low for systemic toxicity (repeated dose) based on the absence of adverse effects in 28- and 90-day oral studies in rats. GreenScreen® criteria classify chemicals as a Low hazard for systemic toxicity (repeated dose) when there are no adverse effects seen following oral exposures >100 mg/kg/day in subchronic studies (CPA 2018b). The confidence in the score is high as it is based on reliable experimental data.

- Authoritative and Screening Lists
  - *Authoritative:* Not present on any authoritative lists for this endpoint.
  - *Screening:* Not present on any screening lists for this endpoint.
- ECHA 2021b
  - *Oral:* In a GLP-compliant repeated dose toxicity study conducted according to OECD Guideline 408, male and female Wistar rats (10/sex/dose) received 100, 300, and 1,000 mg/kg/day erucamide (purity not reported) in corn oil via oral gavage for 90 days. Animals were evaluated weekly for changes in clinical observations, body weight, and food consumption. Animals were subjected to an ophthalmoscopic examination during the last week of treatment. Blood and urine samples were collected prior to sacrifice for hematology, clinical chemistry, and urinalysis. At completion of the study animals were euthanized and subjected to a gross pathological and histopathological examination. No toxicologically significant treatment-related changes were found. Based on the lack of systemic toxicity effects seen, authors identified a NOAEL of 1,000 mg/kg/day (Klimisch 1, reliable without restriction).
  - *Oral:* A non-GLP study was conducted similarly to OECD Guideline 407 to evaluate repeated-dose toxicity of erucamide. Male weanling Sprague-Dawley rats were fed a non-standard diet containing erucamide (98.5% purity) in place of the 10% sucrose that would normally be present. Based on mean body weight and food consumption for weanling Sprague-Dawley rats, the animals received 14,700 mg erucamide/kg/day[9] in this study. Rats had access to test diet and water *ad libitum*, and the study lasted 28 days. At the end of the exposure period, the animals were euthanized and subjected to necropsy to identify any test material-related gross abnormalities. Hematological and clinical chemistry evaluations were also conducted. No deaths or abnormalities other than decreased terminal body weight were

[9] Dose was calculated using the male Sprague-Dawley rat food factor value for weanling studies (TERA Undated).
10% = 100,000 mg/kg food * 0.147 kg food/kg BW/day = 14,700 mg/kg body weight/day

observed in the experimental group, and so the study was reported with a NOAEL of 14,700 mg erucamide/kg/day. Because the caloric content of the test diet was slightly different than standard diet, the effects on body weight are not likely a result of toxicity but merely energy imbalance (Klimisch 2, reliable with restrictions).

- *ToxServices compared the NOAEL to tripled guidance values due to the 28-day duration of the study, as GHS guidance values are based on 90-day studies.*

Neurotoxicity (single dose, N-single) (Group II) Score (vH, H, M, or L): *L*

Erucamide was assigned a score of Low for neurotoxicity (single dose) based on a lack of effects on clinical signs and gross pathology suggestive of neurotoxicity in acute toxicity studies. GreenScreen® criteria classify chemicals as a Low hazard for neurotoxicity (single dose) when adequate data are available and they are not classified under GHS (CPA 2018b). Confidence in the score is low as specific neurotoxicity examinations are not carried out in standard acute toxicity studies.

- Authoritative and Screening Lists
  - *Authoritative:* Not present on any authoritative lists for this endpoint.
  - *Screening:* Not present on any screening lists for this endpoint.
- ECHA 2021b
  - *Oral:* In a GLP-compliant study conducted according to OECD Guideline 423, investigators exposed groups of male and female Sprague-Dawley rats (3/sex) to erucamide (purity not reported) via gavage. Test material was dissolved in peanut oil such that all animals received 2,000 mg/kg-bw in 10 mL/kg body weight. Investigators evaluated animals for overt signs of toxicity at post-exposure hours 0.5, 1, 2, and 4; animals were examined again daily for the course of the 14-day study. After two weeks had elapsed, all animals were euthanized and subjected to a gross pathological examination. No deaths were observed, nor were any gross pathological changes found in the exposed animals. The authors established an LD_{50} >2,500 mg/kg, as no deaths were observed at 2,000 mg/kg (Klimisch 1, reliable without restriction).
    - *ToxServices notes that only a dose of 2,000 mg/kg was administered, and it is not clear why the LD_{50} was determined to be > 2,500 mg/kg.*
  - *Oral:* A GLP-compliant study conducted according to OECD Guideline 401 found no deaths or adverse effects on clinical signs, body weight development, and gross pathology examinations when male and female Sprague-Dawley rats (5/sex) received erucamide in corn oil via gavage at 2,000 mg/kg. The authors established an LD_{50} >2,000 mg/kg (Klimisch 2, reliable with restrictions).
  - *Oral:* A GLP-compliant study conducted according to EU Method B.1 was carried out to assess oral toxicity of erucamide. Male and female Wistar rats (5/sex) received two doses of 2,500 mg/kg erucamide (purity not reported) within 24 hours. Animals were observed for 14 days, and then euthanized and subjected to macroscopic evaluation. No deaths, adverse effects during the observation period, or macroscopic abnormalities were noted. The authors established an LD_{50} >5,000 mg /kg (Klimisch 2, reliable with restrictions).
  - *Dermal:* In a GLP-compliant study conducted according to OECD Guideline 402, investigators assessed the acute dermal toxicity of erucamide in male and female Wistar rats. Investigators clipped the hair from the backs of the test animals (5/sex) the night before the experiment was slated to begin. Animals were checked to have fully intact, non-irritated skin before test material was applied on study day 1. Test material, in the form of a powder, was applied to moistened gauze pads which were placed onto the backs of the experimental animals and held in place with semi-occlusive dressings. All animals received 2,000 mg erucamide/kg. The dressings and test material were removed 24 hours after application, and then animals were washed gently with warm water and dried with paper towels. Animals

were evaluated for signs of intoxication and viability at post-exposure hours 0.5, 1, 2, 3, 5, and twice daily starting at post-exposure hour 24. No deaths were observed, and body weights stayed in the normal range for animals of the relevant age and strain. There were no clinical signs of toxicity or effects on gross pathology. No skin damage was observed except for minor peeling at days 7 and 8 in one male animal. Authors established an LD_{50} >2,000 mg/kg (Klimisch 1, reliable without restriction).

- *Inhalation:* In a GLP-compliant study conducted according to OECD Guideline 436, male and female Wistar rats were exposed to erucamide (99.51% purity) via inhalation of a dust. Animals (3/sex) were fitted with nose-only, flow-past exposure systems, and received 2.8 mg test material/L air continuously for four hours. This concentration was deemed the highest feasible concentration that would generate respirable dusts without agglomerating, and no deaths or adverse effects on clinical signs, body weight, or gross pathology were noted over the 14-day observation period or at necropsy. Authors established an LC_{50} >2.8 mg/L/4 hr (Klimisch 1, reliable without restriction).

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

Erucamide was assigned a score of Low for neurotoxicity (repeated dose) based on the lack of treatment-related effects on neurobehavioral parameters in an oral 90-day toxicity study in rats. GreenScreen® criteria classify chemicals as a Low hazard for neurotoxicity (repeated dose) when adequate data are available, studies are negative, and it is not GHS classified (CPA 2018b). The confidence in the score is high as it is based on reliable experimental data.

- Authoritative and Screening Lists
  - *Authoritative:* Not present on any authoritative lists for this endpoint.
  - *Screening:* Not present on any screening lists for this endpoint.
- ECHA 2021b
  - *Oral:* In the previously described GLP-compliant repeated dose toxicity study conducted according to OECD Guideline 408, male and female Wistar rats (10/sex/dose) received 100, 300, and 1,000 mg/kg/day erucamide (purity not reported) in corn oil via gavage for 90 days. A neurobehavioral examination was performed before the first exposure and once during the last treatment week. Investigators evaluated a battery of functions including sensory activity, grip strength, motor activity, body temperature, rearing, urination, and defecation. No treatment-related effects were found. ToxServices established a neurobehavioral NOAEL of 1,000 mg/kg/day (Klimisch 1, reliable without restriction).

Skin Sensitization (SnS) (Group II*) Score (H, M, or L): L

Erucamide was assigned a score of Low for skin sensitization based on no sensitization seen in a local lymph node assay in mice. GreenScreen® criteria classify chemicals as a Low hazard for skin sensitization when adequate data are available, studies are negative, and is not GHS classified (CPA 2018b). The confidence in the score is high as it is based on reliable experimental data.

- Authoritative and Screening Lists
  - *Authoritative:* Not present on any authoritative lists for this endpoint.
  - *Screening:* Not present on any screening lists for this endpoint.
- ECHA 2021b
  - In a GLP-compliant local lymph node assay (LLNA) conducted according to OECD Guideline 429, erucamide (99.2% purity) was applied to CBA female mice (5/dose) at concentrations of 5, 10, and 25% w/v in tetrahydrofuran. The highest tested concentration (25%) was determined based on its solubility as well as results of a preliminary test showing no signs of skin irritation. Erucamide was not found to be a skin sensitizer under the

conditions of this study, with stimulation indices of 0.81, 0.84, and 0.70 at the 5, 10, and 25% concentrations, respectively (Klimisch 1, reliable without restriction).

Respiratory Sensitization (SnR) (Group II*) Score (H, M, or L): *L*

Erucamide was assigned a score of Low for respiratory sensitization based on ECHA's guidance on respiratory sensitization evaluation. GreenScreen® criteria classify chemicals as a Low hazard for respiratory sensitization when adequate and negative data and no GHS classification are available (CPA 2018b). The confidence in the score is low as this evaluation does not include non-immunologic mechanisms of respiratory sensitization, and no specific data are available for respiratory sensitization.

- Authoritative and Screening Lists
  - *Authoritative:* Not present on any authoritative lists for this endpoint.
  - *Screening:* Not present on any screening lists for this endpoint.
- OECD 2020a
  - Erucamide does not contain any structural alerts for respiratory sensitization (Appendix G).
- Based on the weight of evidence and guidance from ECHA regarding assessment of respiratory sensitization potential, a score of Low was assigned. The guidance from ECHA states that the mechanisms leading to respiratory sensitization are essentially similar to those leading to skin sensitization (ECHA 2017). ECHA recommended that if a chemical is not a dermal sensitizer based on high quality data, it is unlikely to be a respiratory sensitizer. ECHA also noted that this rationale does not cover respiratory hypersensitivity caused by non-immunological mechanisms, for which human experience is the main evidence of activity (ECHA 2017). As erucamide was not sensitizing to the skin (see skin sensitization section above), and a literature search did not find any human evidence of respiratory sensitization by erucamide, and as erucamide does not contain any structural alerts for respiratory sensitization (OECD 2020a), erucamide is not expected to be a respiratory sensitizer.

Skin Irritation/Corrosivity (IrS) (Group II) Score (vH, H, M, or L): L

Erucamide was assigned a score of Low for skin irritation/corrosivity based on the absence of skin irritation seen in rabbits. GreenScreen® criteria classify chemicals as a Low hazard for skin irritation/corrosivity when adequate data are available, studies are negative, and is not GHS classified (CPA 2018b). The confidence in the score is high as it is based on reliable experimental data.

- Authoritative and Screening Lists
  - *Authoritative:* Not present on any authoritative lists for this endpoint.
  - *Screening:* Not present on any screening lists for this endpoint.
- ECHA 2021b
  - In a GLP-compliant skin irritation study conducted according to EU Method B.4, three female New Zealand white rabbits were administered erucamide (99% purity) at a dose of 0.5 g on clipped skin for four hours on the left flank of the animal under semiocclusive conditions. Skin was examined 1, 24, 48, and 72 hours after the removal of the dressing. The mean erythema and edema scores were 0 in all three animals when measured up to 72 hours after treatment. The study authors concluded that erucamide was not irritating to skin (Klimisch 1, reliable without restriction).

Eye Irritation/Corrosivity (IrE) (Group II) Score (vH, H, M, or L): L

Erucamide was assigned a score of Low for eye irritation/corrosivity based on the absence of eye irritation seen in rabbits. GreenScreen® criteria classify chemicals as a Low hazard for eye irritation/corrosivity when adequate data are available, studies are negative, and is not GHS classified (CPA 2018b). The confidence in the score is high as it is based on reliable experimental data.

- Authoritative and Screening Lists
  - *Authoritative:* Not present on any authoritative lists for this endpoint.
  - *Screening:* Not present on any screening lists for this endpoint.
- ECHA 2021b
  - In a GLP-compliant eye irritation study conducted according to EU Method B.5, eyes of three female New Zealand white rabbits were instilled with 0.1 mL of erucamide (99% purity) into the conjunctival sac of the left eye followed by continuous exposure until the last observation at 7 days. Eyes were observed at 1, 24, 48, 72 hours, and 7 days post-instillation. Approximately 1 hour post-application, two animals showed diffuse conjunctival redness and slight chemosis; the third animal showed slight conjunctival redness and slight chemosis. The conjunctival redness was fully reversible in one animal by day 3, and between days 3 and 7 in the other animals. The chemosis resolved within 24, 48, or 72 hours for each animal. As per Annex VI of the EE Council Directive 67/548/EC, erucamide is not classified as an ocular irritant (Klimisch 2, reliable with restrictions).

Ecotoxicity (Ecotox)

Acute Aquatic Toxicity (AA) Score (vH, H, M, or L): *L*

Erucamide was assigned a score of Low for acute aquatic toxicity based no effects expected at saturation. GreenScreen® criteria classify chemicals as a Low hazard for acute aquatic toxicity when no effects are expected at saturation (CPA 2018b). The confidence in the score is low as experimental data were not available for all three trophic levels.

- Authoritative and Screening Lists
  - *Authoritative:* Not present on any authoritative lists for this endpoint.
  - *Screening:* Not present on any screening lists for this endpoint.
- ECHA 2021b
  - 48-hour mobility EC_{50} (*Daphnia magna*) ≥0.13 mg/L (GLP-compliant, OECD Guideline 202) (Klimisch 1, reliable without restriction)
  - 72-hour growth rate EC_{50} (*Pseudokirchneriella subcapitata*, algae) >50 µg/L (GLP-compliant, OECD Guideline 201) (Klimisch 1, reliable without restriction)
- Based on the weight of evidence, a score of Low was assigned. Although no acute aquatic toxicity studies were identified in fish, a chronic study identified a NOEC of 105 µg/L in fish. Erucamide has very low water solubility (< 0.738 µg/L) (ECHA 2021b), and no effects are expected at saturation (approximately 10 times the water solubility or 0.007 mg/L).

Chronic Aquatic Toxicity (CA) Score (vH, H, M, or L): L

Erucamide was assigned a score of Low for chronic aquatic toxicity based on no effects are expected at saturation (< 0.738 µg/L). GreenScreen® criteria classify chemicals as a Low hazard for chronic aquatic toxicity when no effects are expected at saturation (CPA 2018b). The confidence in the score is high as it is based on measured data for all three trophic levels.

- Authoritative and Screening Lists
  - *Authoritative:* Not present on any authoritative lists for this endpoint.
  - *Screening:* Not present on any screening lists for this endpoint.
- ECHA 2021b
  - 28-day growth rate NOEC (*Danio rerio*, freshwater fish) ≥105 µg/L (GLP-compliant, similar OECD Guideline 215) (Klimisch 2, reliable with restrictions)
  - 21-day reproduction NOEC (*Daphnia magna*) ≥75 µg/L (GLP-compliant, OECD Guideline 202) (Klimisch 2, reliable with restrictions)

- 72-hour growth rate NOEC (*P. subcapitata*, algae) ≥50 µg/L (GLP-compliant, OECD Guideline 201) (Klimisch 1, reliable without restriction) (Klimisch 1, reliable without restriction).

Environmental Fate (Fate)

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

Erucamide was assigned a score of Low for persistence based on it reaching the pass level of 60% in an OECD 301B test, and on being predicted to mainly partition to sediment. Although it failed the 10-day window, this is due to its low water solubility. Therefore, it is still considered to have passed the test and hence met the GHS rapid degradability criteria. GreenScreen® criteria classify chemicals as a Low hazard for persistence when they meet the GHS rapid degradation criteria and mainly partition to water, soil or sediment (CPA 2018b). The confidence in the score is high as it is based on reliable measured data.

- Authoritative and Screening Lists
  - *Authoritative:* Not present on any authoritative lists for this endpoint.
  - *Screening:* Not present on any screening lists for this endpoint.
- ECHA 2021b
  - A GLP-compliant ready biodegradability test conducted according to OECD Guideline 301B (CO_2 Evolution Test) was performed with activated domestic sludge (non-activated) exposed to erucamide (purity not reported) at 10 mg/L for 28 days. At the end of the exposure period, the level of degradation was 64%. While the 10-day biodegradation window was not met, the rate of biodegradation was limited by its solubility in the test media. Therefore, ECHA dossier authors stated that the failing of the 10-day window could not be used as failing of the test, and erucamide is readily biodegradable (Klimisch 1, reliable without restriction).
  - A GLP-compliant ready biodegradability test was conducted according to OECD Guideline 301B with one deviation – the test material was not tested in duplicates but tested at two concentrations, 10 mg/L and 20 mg/L based on DOC measurement. Biodegradation was monitored by CO_2 evolution. At 28 days, degradation was 28% and 5% at the initial concentrations of 10 mg/L and 20 mg/L, respectively. The positive control performed as expected. Study authors concluded that erucamide was not readily biodegradable, but the degree of biodegradable may be limited by its water solubility (Klimisch 2, reliable with restrictions).
  - A GLP-compliant ready biodegradability study was conducted according to OECD Guideline 301D. Domestic non-adapted activated sludge was exposed to erucamide adsorbed on silica gel at the initial concentration of 2 mg/L for a total of 140 days. Biodegradation was monitored by O_2 consumption. Erucamide was degraded by 15% at 28 days and 43% at 140 days. The positive control performed as expected. Study authors concluded that erucamide was not readily biodegradable (Klimisch 1, reliable without restriction).
- U.S. EPA 2017
  - The BIOWIN modeling Ready Biodegradable Predictor indicates that erucamide is not expected to be readily biodegradable. Fugacity modeling (EQC default method) predicts 1.91% will partition to water with a half-life of 37.5 days, 69.4% will partition to sediment with a half-life of 337.5 days, and 28.7% will partition to soil with a half-life of 75 days (Appendix H).

- Based on the weight of evidence, a score of Low was assigned. Per OECD guidance, when conflicting data are available from multiple biodegradability tests, the positive tests (i.e., readily biodegradable) of acceptable reliability could be considered valid regardless of the negative studies (OECD 2001). Therefore, the score for this endpoint is based on the best-performing tests. Available ready biodegradability studies indicate erucamide is degradable. In the best-performing test, an OECD 301B study, erucamide met the pass level, but it did not meet the 10-day window. However, this was attributed to the low solubility of erucamide by the ECHA dossier authors, who concluded that erucamide met the ready biodegradability criteria. Therefore, ToxServices considered that erucamide meet the GHS rapid degradability criteria.

Bioaccumulation (B) Score (vH, H, M, L, or vL): *L*
Erucamide was assigned a score of Low for bioaccumulation based on a modeled BAF of 403.3. GreenScreen® criteria classify chemicals as a Low hazard for bioaccumulation when the BAF value is between 100 and 500 (CPA 2018b). The confidence in the score is low as it is based on modeled data.

- Authoritative and Screening Lists
  - *Authoritative:* Not present on any authoritative lists for this endpoint.
  - *Screening:* Not present on any screening lists for this endpoint.
- U.S. EPA 2017
  - BCFBAF predicts a BCF of 1,090 using the regression-based model based on a measured log K_{ow} of 8, and a BAF of 403.3 using the Arnot-Gobas model for the upper trophic level, taking metabolism into consideration (Appendix H).

Physical Hazards (Physical)

Reactivity (Rx) Score (vH, H, M, or L): *L*
Erucamide was assigned a score of Low for reactivity based on the absence of functional groups associated with explosive or self-reactive properties. GreenScreen® criteria classify chemicals as a Low hazard for reactivity when the chemical does not warrant GHS classification as explosive or self-reactive and the chemical is not present on authoritative or screening lists (CPA 2018b). The confidence in the score is low based on the lack of experimental data.

- Authoritative and Screening Lists
  - *Authoritative:* Not present on any authoritative lists for this endpoint.
  - *Screening:* Not present on any screening lists for this endpoint.
- No measured data were identified. Therefore, screening procedures for explosivity were used here to estimate the reactivity property of erucamide. These procedures are listed in the GHS (UN 2019).
  - Based on the structure of its components or moieties, erucamide is not considered explosive or self-reactive due to lack of functional groups associated with explosive or self-reactive properties (See Appendix I).
  - Based on the structure of its components or moieties, erucamide is not considered to have oxidizing properties as it does not contain any structural groups known to be correlated with a tendency to react exothermally with combustible materials.

Flammability (F) Score (vH, H, M, or L): L
Erucamide was assigned a score of Low for flammability based on negative results in a test for the flammability of solids (Klimisch score 1). GreenScreen® criteria classify chemicals as a Low hazard for flammability when available data indicate that the chemical does not warrant GHS classification as a flammable solid and the chemical is not present on authoritative or screening lists (CPA 2018b). The confidence in the score is high as it is based on reliable experimental data.

- Authoritative and Screening Lists
    - *Authoritative:* Not present on any authoritative lists for this endpoint.
    - *Screening:* Not present on any screening lists for this endpoint.
- ECHA 2021b
    - In a non-GLP compliant solid flammability study conducted according to EU Method A.10, erucamide was not considered to be highly flammable. The test substance did not ignite within 2 minutes in the pre-test (Klimisch 1, reliable without restriction).

Use of New Approach Methodologies (NAMs)[10] in the Assessment, Including Uncertainty Analyses of Input and Output

New Approach Methodologies (NAMs) used in this GreenScreen® include *in silico* modeling for carcinogenicity, endocrine activity, respiratory sensitization, and bioaccumulation, and *in vitro* testing for genotoxicity. NAMs are non-animal alternative that can be used alone or in combination to provide information for safety assessment (Madden et al. 2020). At present, there is not a uniformly accepted framework on how to report and apply individual NAMs (U.S. EPA 2020, OECD 2020b). The expanded application of NAMs greatly amplifies the need to communicate uncertainties associated with their use. As defined by EFSA (2018), uncertainty is "a general term referring to all types of limitations in available knowledge that affect the range and probability of possible answers to an assessment question." The quality, utility, and accuracy of NAM predictions are greatly influenced by two primary types of uncertainties (OECD 2020b):

- Type I: Uncertainties related to the input data used
- Type II: Uncertainties related to extrapolations made

As shown in Table 3, Type I (input data) uncertainties in erucamide's NAMs dataset include lack of data on carcinogenicity, endocrine activity, respiratory sensitization, and bioaccumulation. Erucamide's Type II (extrapolation output) uncertainties include limitations of modeling software Toxtree and OECD Toolbox in identifying structural alerts without defining applicability domains, the limitations of *in vitro* genotoxicity assays in mimicking *in vivo* metabolic systems and the uncertain *in vivo* relevance of *in silico* modeling of endocrine receptor binding. Some of erucamide's type II uncertainties were alleviated by the use of *in vitro* test batteries and/or in combination of *in vivo* data.

Table 3: Summary of NAMs Used in the GreenScreen® Assessment, Including Uncertainty Analyses	
Uncertainty Analyses (OECD 2020b)	
Type I Uncertainty: Data/Model Input	**Carcinogenicity**: No experimental data are available. **Endocrine activity:** No experimental data are available. **Respiratory sensitization**: No experimental data are available and there are no validated test methods. **Bioaccumulation:** No experimental data are available and measured log K_{ow} warrants a Very High score.
Type II Uncertainty: Extrapolation Output	**Carcinogenicity**: Toxtree only identifies structural alerts (SAs), and no applicability domain can be defined (Toxtree 2018). **Genotoxicity:** The bacterial reverse mutation assay (as defined in OECD Guideline 471) only tests point-mutation inducing activity in non-mammalian cells, and the exogenous metabolic activation system does not entirely mimic *in vivo* conditions[11]. The mammalian cell gene mutation assay (as defined in OECD Guideline 476) only detects gene mutations, and the exogenous

[10] NAMs refers to any non-animal technology, methodology, approach, or combination thereof that inform chemical hazard and risk assessments. NAMs include *in silico*/computational tools, *in vitro* biological profiling (e.g., cell cultures, 2,3-D organotypic culture systems, genomics/transcriptomics, organs on a chip), and frameworks (i.e., adverse outcome pathways (AOPs), defined approaches (DA), integrated approaches to testing and assessment (IATA).

[11] https://www.oecd-ilibrary.org/docserver/9789264071247-en.pdf?expires=1614097593&id=id&accname=guest&checksum=89925F80B9F4BD2FFC6E90F94A0EE427

	metabolic activation system does not entirely mirror *in vivo* metabolism (i.e., the liver S9 mix contains enzymes present in the endoplasmic reticulum but not the cytosol of liver cells).[12] The *in vitro* chromosome aberration assay (OECD 473) does not measure aneuploidy and it only measures structural chromosomal aberrations. The exogenous metabolic activation system does not entirely mirror *in vivo* metabolism[13]. **Endocrine activity:** the *in vivo* relevance of *in silico* receptor binding activity is unclear due to lack of sufficient data on toxicokinetics. **Respiratory sensitization**: The OECD Toolbox only identifies structural alerts and does not define applicability domains. Additionally, the ECHA guidance (2017), on which the use of OECD Toolbox structural alerts is based, does not evaluate non-immunologic mechanisms for respiratory sensitization.	
Endpoint	**NAMs Data Available and Evaluated? (Y/N)**	**Types of NAMs Data (*in silico* modeling/*in vitro* biological profiling/frameworks)**
Carcinogenicity	Y	*In silico* modeling: VEGA/Toxtree/Danish QSAR
Mutagenicity	Y	*In vitro* data: Bacterial reverse mutation assay/*in vitro* gene mutation assay/*in vitro* chromosome aberration assay
Reproductive toxicity	N	
Developmental toxicity	N	
Endocrine activity	Y	*In silico* modeling: ToxCast/Danish QSAR
Acute mammalian toxicity	N	
Single exposure systemic toxicity	N	
Repeated exposure systemic toxicity	N	
Single exposure neurotoxicity	N	
Repeated exposure neurotoxicity	N	
Skin sensitization	N	
Respiratory sensitization	Y	*In silico* modeling: OECD Toolbox structural alerts
Skin irritation	N	
Eye irritation	N	
Acute aquatic toxicity	N	

[12] https://www.oecd-ilibrary.org/docserver/9789264264809-en.pdf?expires=1614097800&id=id&accname=guest&checksum=C0DE371FB9C5A878E66C9AB7F84E6BBE
[13] https://www.oecd-ilibrary.org/docserver/9789264264649-en.pdf?expires=1614098015&id=id&accname=guest&checksum=6A4F9CE52EA974F5A74793DD54D54352

Chronic aquatic toxicity	N	
Persistence	N	
Bioaccumulation	Y	*In silico* modeling: EPI Suite™

References

ChemIDplus. 2021. Erucamide (CAS #112-84-5). United States National Library of Medicine. Available: http://chem.sis.nlm.nih.gov/chemidplus/chemidheavy.jsp

Clean Production Action (CPA). 2018a. GreenScreen® Assessment Expiration Policy. October 2, 2018.

Clean Production Action (CPA). 2018b. The GreenScreen® for Safer Chemicals Guidance. Version 1.4 Guidance. Dated January 2018. Available: https://www.greenscreenchemicals.org/static/ee_images/uploads/resources/GreenScreen_Guidance_v1_4_2018_01_Final.pdf

Danish Technical University (DTU). 2021. National Food Institute. Danish QSAR Database. Available: http://qsar.food.dtu.dk/.

European Chemicals Agency (ECHA). 2017. Guidance on information requirements and Chemical Safety Assessment. Chapter R.7a: Endpoint specific guidance. Version 6.0. Dated: July 2017. Available: https://echa.europa.eu/documents/10162/13632/information_requirements_r7a_en.pdf

European Chemicals Agency (ECHA). 2021a. C&L Inventory. Available: https://echa.europa.eu/information-on-chemicals/cl-inventory-database/-/discli/details/79192

European Chemicals Agency (ECHA). 2021b. (Z)-Docos-13-enamide (CAS #112-84-5). Available: https://echa.europa.eu/registration-dossier/-/registered-dossier/14899

European Food Safety Authority (EFSA). 2018. Guidance on uncertainty analysis in scientific assessments. *EFSA J.* 16(1): e05123. Available: https://www.ncbi.nlm.nih.gov/pmc/articles/PMC7009727/

Health Canada. 2019. Screening Assessment: Fatty Amides Group. Environment and Climate Change Canada. Dated April 2019. Available: https://www.canada.ca/content/dam/eccc/documents/pdf/pded/fatty-amides/Screening-assessment-fatty-amides-group.pdf

Madden, J.C., S.J. Enoch, A. Paini, and M.T.D. Cronin. "A Review of In Silico Tools as Alternatives to Animal Testing: Principles, Resources, and Applications," *Alt. Lab. Animals* 1-27, (2020). Available: https://journals.sagepub.com/doi/pdf/10.1177/0261192920965977

Organization for Economic Cooperation and Development (OECD). 2001. Guidance document on the use of the harmonised system for the classification of chemicals which are hazardous for the aquatic environment. OECD Series on testing and assessment No. 27.

Organisation for Economic Co-operation and Development (OECD). 2020a. OECD QSAR Toolbox for Grouping Chemicals into Categories Version 4.4.1. Available: http://toolbox.oasis-lmc.org/?section=download&version=latest

Organization for Economic Cooperation and Development (OECD). 2020b. Overview of Concepts and Available Guidance related to Integrated Approaches to Testing and Assessment (IATA), Series on Testing and Assessment, No. 329, Environment, Health and Safety, Environment Directorate. Available: http://www.oecd.org/chemicalsafety/risk-assessment/concepts-and-available-guidance-related-to-integrated-approaches-to-testing-and-assessment.pdf

Pharos. 2021. Pharos Chemical and Material Library Entry for Erucamide (CAS #112-84-5). Available: http://www.pharosproject.net/material/

PubChem. 2021. Erucamide (CAS #112-84-5). United States National Library of Medicine. Available: https://pubchem.ncbi.nlm.nih.gov/compound/Erucamide

ToxServices. 2020. SOP 1.37: GreenScreen® Hazard Assessments. Dated: February 3, 2020.

Toxtree. 2018. Estimation of Toxic Hazard- A Decision Tree Approach v3.1.0. Available: http://toxtree.sourceforge.net

United Nations (UN). 2019. Globally Harmonized System of Classification and Labelling of Chemicals (GHS). Eighth revised edition.

United States Department of Transportation (U.S. DOT). 2008a. Chemicals Listed with Classification. 49 CFR § 172.101. Available: http://www.gpo.gov/fdsys/pkg/CFR-2008-title49-vol2/pdf/CFR-2008-title49-vol2-sec172-101.pdf.

United States Department of Transportation (U.S. DOT). 2008b. Classification Criteria. 49 CFR § 173. Available: http://www.ecfr.gov/cgi-bin/text-idx?c=ecfr&tpl=/ecfrbrowse/Title49/49cfr173_main_02.tpl

United States Environmental Protection Agency (U.S. EPA). 2015. Safer Choice Standard. Available: https://www.epa.gov/saferchoice/standard

United States Environmental Protection Agency (U.S. EPA). 2017. Estimation Programs Interface (EPI) Suite™ Web, v4.11, Washington, DC, USA. Available: http://www.epa.gov/opptintr/exposure/pubs/episuitedl.htm

United States Environmental Protection Agency (U.S. EPA). 2020. New Approach Methods Workplan. Office of Research and Development. Office of Chemical Safety and Pollution Prevention. EPA 615B20001. June 2020. Available: https://www.epa.gov/sites/production/files/2020-06/documents/epa_nam_work_plan.pdf

United States Environmental Protection Agency (U.S. EPA). 2021a. Safer Chemical Ingredients List (SCIL). Available: https://www.epa.gov/saferchoice/safer-ingredients

United States Environmental Protection Agency (U.S. EPA). 2021b. Comptox Dashboard entry for erucamide (CAS #112-84-5). ToxCast Models. Available: https://comptox.epa.gov/dashboard/dsstoxdb/results?search=DTXSID4026929#bioactivity-toxcast-models

Virtual Models for Evaluating the Properties of Chemicals within a Global Architecture (VEGA). 2021. Predictive Model Platform version 1.1.5. Available: http://www.vega-qsar.eu/index.php

APPENDIX A: Hazard Classification 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 Erucamide (CAS #112-84-5)

TOXSERVICES TOXICOLOGY RISK ASSESSMENT CONSULTING

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	Erucamide	112-84-5	*L*	L	DG	L	DG	L	L	L	*L*	L	L	*L*	L	L	*L*	L	L	*L*	*L*	L

Table 3: Hazard Summary Table

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

Table 4

Chemical Name	Preliminary GreenScreen® Benchmark Score
Erucamide	4

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

Table 6

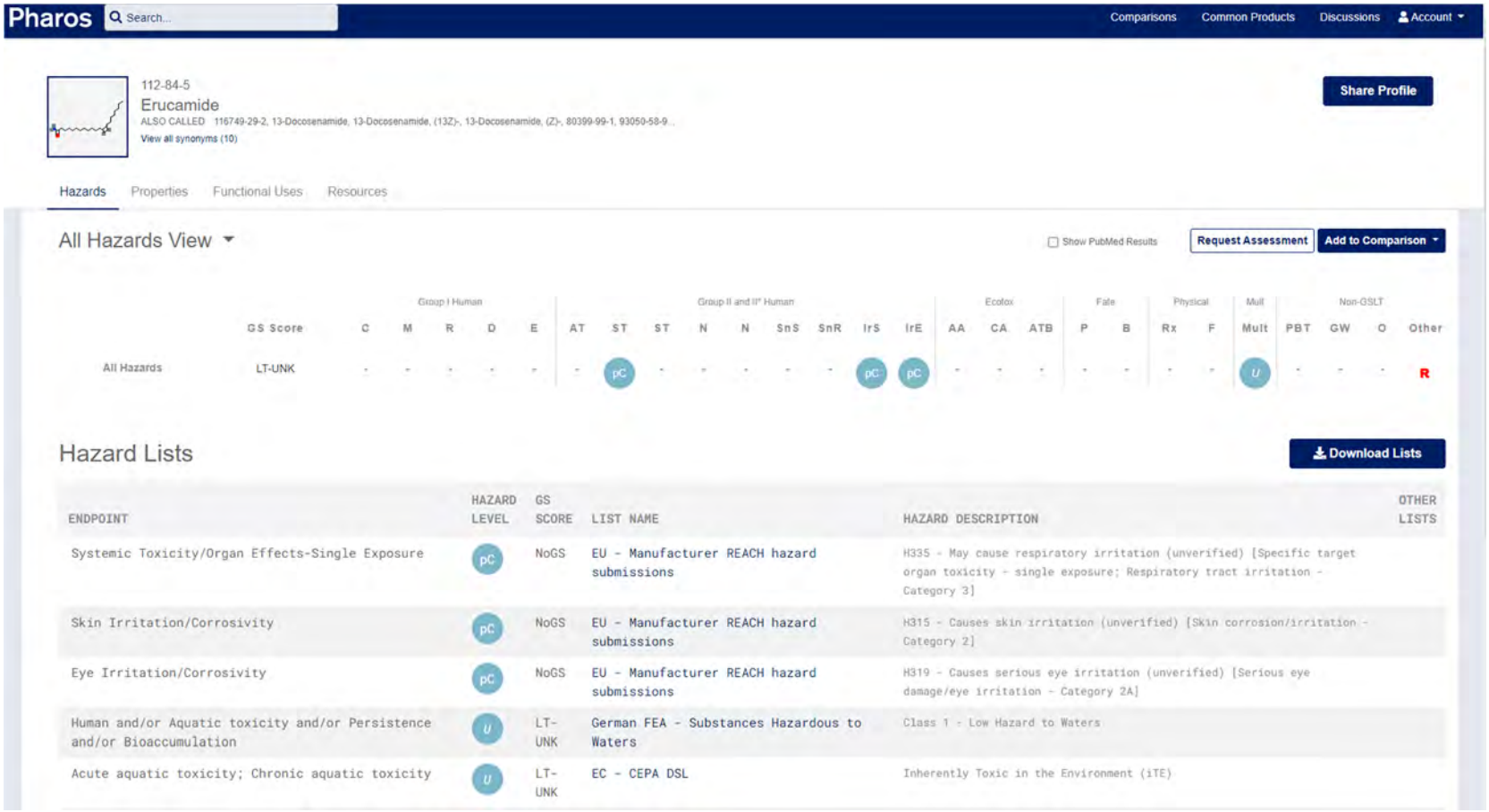
Chemical Name	Final GreenScreen® Benchmark Score
Erucamide	2DG

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

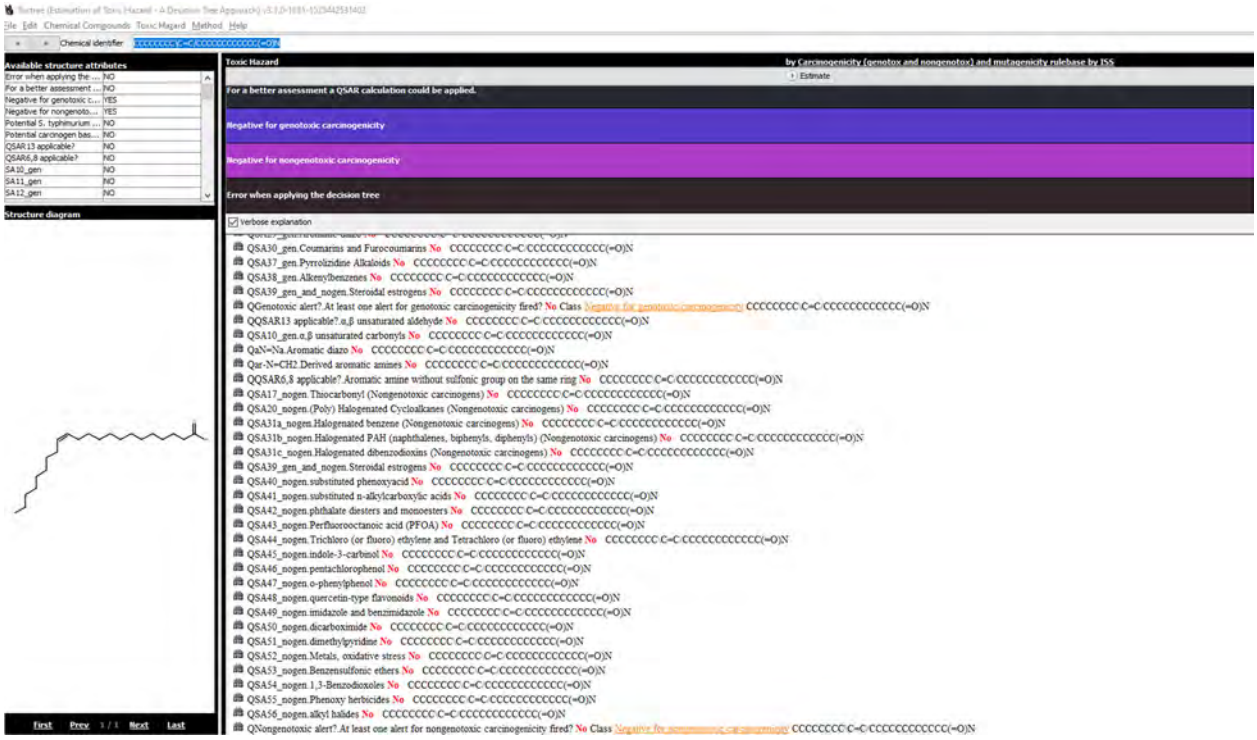
Table 5: Data Gap Assessment Table

Datagap Criteria	a	b	c	d	e	f	g	h	i	j	bm4	End Result
1												
2												
3												
4	Yes	Yes	Yes	Yes	Yes	No	Yes	Yes	Yes	Yes	No	2DG

APPENDIX C: Pharos Output for Erucamide (CAS #112-84-5)



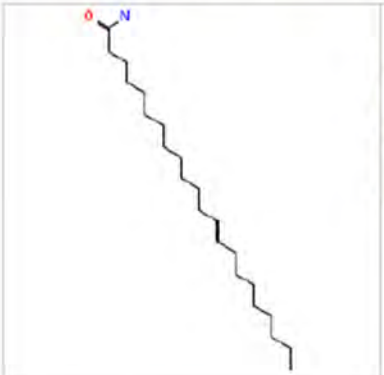
APPENDIX D: Toxtree Carcinogenicity Results for Erucamide (CAS #112-84-5)



APPENDIX E: VEGA Carcinogenicity Modeling Results for Erucamide (CAS #112-84-5)

1. Prediction Summary

Prediction for compound Molecule 0



Prediction: Reliability:

Prediction is NON-Carcinogen, but the result shows some critical aspects, which require to be checked:
- accuracy of prediction for similar molecules found in the training set is not optimal

Compound: Molecule 0
Compound SMILES: O=C(N)CCCCCCCCCCCC=CCCCCCCCC
Experimental value: -
Predicted Carcinogen activity: NON-Carcinogen
P(Carcinogen): 0.13
P(NON-Carcinogen): 0.87
Reliability: the predicted compound could be out of the Applicability Domain of the model
Remarks:
none

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

Compound #1

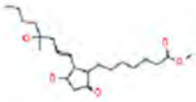
CAS: 112-63-0
Dataset id: 451 (Test set)
SMILES: O=C(OC)CCCCCCCC=CCC=CCCCCC
Similarity: 0.833

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

Compound #2

CAS: 1643-20-5
Dataset id: 273 (Training set)
SMILES: [O-][N+](C)(C)CCCCCCCCCCCC
Similarity: 0.774

Experimental value: NON-Carcinogen
Predicted value: Carcinogen



Compound #3

CAS: 59122-46-2
Dataset id: 485 (Training set)
SMILES: O=C(OC)CCCCCCC1C(=O)CC(O)C1(C=CCC(O)(C)CCCC)
Similarity: 0.769

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

Compound #4

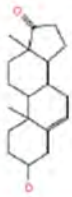
CAS: 2432-99-7
Dataset id: 50 (Test set)
SMILES: O=C(O)CCCCCCCCCCN
Similarity: 0.767

Experimental value: Carcinogen
Predicted value: NON-Carcinogen

Compound #5

CAS: 75881-20-8
Dataset id: 558 (Training set)
SMILES: O=NN(C)CCCCCCCCCCCCCC
Similarity: 0.767

Experimental value: Carcinogen
Predicted value: Carcinogen



Compound #6

CAS: 53-43-0
Dataset id: 198 (Training set)
SMILES: O=C2CCC3C4CC=C1CC(O)CCC1(C)C4(CCC23(C))
Similarity: 0.764

Experimental value: Carcinogen
Predicted value: Carcinogen

3.2 Applicability Domain:
Measured Applicability Domain Scores

⚠	**Global AD Index** AD index = 0.761 Explanation: the predicted compound could be out of the Applicability Domain of the model.
✓	**Similar molecules with known experimental value** Similarity index = 0.801 Explanation: strongly similar compounds with known experimental value in the training set have been found.
⚠	**Accuracy of prediction for similar molecules** Accuracy index = 0.524 Explanation: accuracy of prediction for similar molecules found in the training set is not optimal.
✓	**Concordance for similar molecules** Concordance index = 1 Explanation: similar molecules found in the training set have experimental values that agree with the predicted value.
✓	**Model's 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.
✓	**Atom Centered Fragments similarity check** ACF index = 1 Explanation: all atom centered fragment of the compound have been found in the compounds of the training set.
✓	**Model class assignment reliability** Pos/Non-Pos difference = 0.739 Explanation: model class assignment is well defined.
✓	**Neural map neurons concordance** Neurons concordance = 1 Explanation: predicted value agrees with experimental values of training set compounds laying in the same neuron.

Symbols explanation:

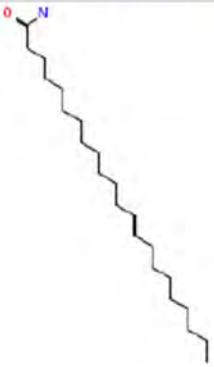
✓ The feature has a good assessment, model is reliable regarding this aspect.

⚠ 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.

1. Prediction Summary

Prediction for compound Molecule 0



Prediction: Reliability:

Prediction is NON-Carcinogen, but the result may be not reliable. A 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
- accuracy of prediction for similar molecules found in the training set is not optimal
- some similar molecules found in the training set have experimental values that disagree with the predicted value

Compound: Molecule 0
Compound SMILES: O=C(N)CCCCCCCCCCCC=CCCCCCCCC
Experimental value: -
Predicted Carcinogen activity: NON-Carcinogen
Structural alerts: -
Reliability: the predicted compound is outside the Applicability Domain of the model
Remarks:
none

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

Compound #1

CAS: 1643-20-5
Dataset id: 879 (Training set)
SMILES: [O-][N+](C)(C)CCCCCCCCCCCC
Similarity: 0.774

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

Compound #2

CAS: 2432-99-7
Dataset id: 36 (Training set)
SMILES: O=C(O)CCCCCCCCCCN
Similarity: 0.767

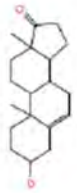
Experimental value: Carcinogen
Predicted value: NON-Carcinogen

Compound #3

CAS: 75881-20-8
Dataset id: 579 (Training set)
SMILES: O=NN(C)CCCCCCCCCCCCCC
Similarity: 0.767

Experimental value: Carcinogen
Predicted value: Carcinogen

Alerts (not found in the target): SA21 Alkyl and aryl N-nitroso groups



Compound #4

CAS: 53-43-0
Dataset id: 836 (Training set)
SMILES: O=C2CCC3C4CC=C1CC(O)CCC1(C)C4(CCC23(C))
Similarity: 0.764

Experimental value: Carcinogen
Predicted value: NON-Carcinogen

Compound #5

CAS: 55090-44-3
Dataset id: 547 (Training set)
SMILES: O=NN(C)CCCCCCCCCCCC
Similarity: 0.741

Experimental value: Carcinogen
Predicted value: Carcinogen

Alerts (not found in the target): SA21 Alkyl and aryl N-nitroso groups

Compound #6

CAS: 68107-26-6
Dataset id: 527 (Training set)
SMILES: O=NN(C)CCCCCCCCCC
Similarity: 0.727

Experimental value: Carcinogen
Predicted value: Carcinogen

Alerts (not found in the target): SA21 Alkyl and aryl N-nitroso groups

3.2 Applicability Domain:
Measured Applicability Domain Scores

✖	**Global AD Index** AD index = 0.623 Explanation: the predicted compound is outside the Applicability Domain of the model.
⚠	**Similar molecules with known experimental value** Similarity index = 0.77 Explanation: only moderately similar compounds with known experimental value in the training set have been found.
⚠	**Accuracy of prediction for similar molecules** Accuracy index = 0.503 Explanation: accuracy of prediction for similar molecules found in the training set is not optimal.
⚠	**Concordance for similar molecules** Concordance index = 0.503 Explanation: some similar molecules found in the training set have experimental values that disagree with the predicted value.
✔	**Atom Centered Fragments similarity check** ACF index = 1 Explanation: all atom centered fragment of the compound have been found in the compounds of the training set.

Symbols explanation:

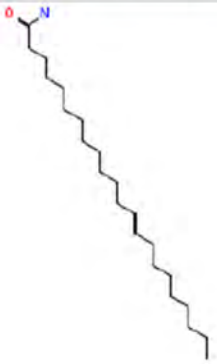
✔ The feature has a good assessment, model is reliable regarding this aspect.

⚠ 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.

1. Prediction Summary

Prediction for compound Molecule 0



Prediction: Reliability:

Prediction is Possible NON-Carcinogen, the result appears reliable. Anyhow, you should check it through the evaluation of the information given in the following sections.

Compound: Molecule 0
Compound SMILES: O=C(N)CCCCCCCCCCCC=CCCCCCCCC
Experimental value: -
Predicted Mutagen activity: Possible NON-Carcinogen
No. alerts for carcinogenicity: 0
Structural alerts: -
Reliability: the predicted compound is into the Applicability Domain of the model
Remarks:
none

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

Compound #1

CAS: N.A.
Dataset id: 1199 (Training set)
SMILES: O=C([O-])CCCCCCCC=CCCCCCCCC
Similarity: 0.876

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

Compound #2

CAS: N.A.
Dataset id: 1366 (Training set)
SMILES: O=C(O)CCCCCCCC=CCCCCCCCC
Similarity: 0.876

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

Compound #3

CAS: N.A.
Dataset id: 1239 (Training set)
SMILES: O=C(N(CCO)CCO)CCCCCCCC=CCCCCCCCC
Similarity: 0.871

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

Compound #4

CAS: N.A.
Dataset id: 1221 (Training set)
SMILES: O=C(O)CCCCCCCC=CCC(O)CCCCCC
Similarity: 0.846

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

Compound #5

CAS: N.A.
Dataset id: 450 (Training set)
SMILES: O=C(OC)CCCCCCCC=CCC=CCCCCC
Similarity: 0.833

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

Compound #6

CAS: N.A.
Dataset id: 273 (Training set)
SMILES: [O-][N+](C)(C)CCCCCCCCCCCC
Similarity: 0.774

Experimental value: NON-Carcinogen
Predicted value: Carcinogen

Alerts (not found in the target): Carcinogenity alert no. 64

3.2 Applicability Domain:
Measured Applicability Domain Scores

Global AD Index
AD index = 0.935
Explanation: the predicted compound is into the Applicability Domain of the model.

Similar molecules with known experimental value
Similarity index = 0.874
Explanation: strongly similar compounds with known experimental value in the training set have been found.

Accuracy of prediction for similar molecules
Accuracy index = 1
Explanation: accuracy of prediction for similar molecules found in the training set is good.

Concordance for similar molecules
Concordance index = 1
Explanation: similar molecules found in the training set have experimental values that agree with the predicted value.

Atom Centered Fragments similarity check
ACF index = 1
Explanation: all atom centered fragment of the compound have been found in the compounds of the training set.

Symbols explanation:

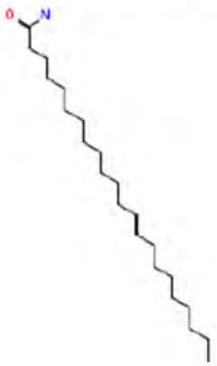
The feature has a good assessment, model is reliable regarding this aspect.

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.

1. Prediction Summary

Prediction for compound Molecule 0



Prediction: Reliability:

Prediction is Carcinogen, but the result shows some critical aspects, which require to be checked:
- only moderately similar compounds with known experimental value in the training set have been found
- accuracy of prediction for similar molecules found in the training set is not optimal
- some similar molecules found in the training set have experimental values that disagree with the predicted value
The following relevant fragments have been found: Carcinogenity alert no. 39

Compound: Molecule 0
Compound SMILES: O=C(N)CCCCCCCCCCCC=CCCCCCCCC
Experimental value: -
Predicted Mutagen activity: Carcinogen
No. alerts for carcinogenicity: 1
Structural alerts: Carcinogenity alert no. 39
Reliability: the predicted compound could be out of the Applicability Domain of the model
Remarks:
none

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

Compound #1

CAS: 1643-20-5
Dataset id: 777 (Training set)
SMILES: [O-][N+](C)(C)CCCCCCCCCCCC
Similarity: 0.774

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

Compound #2

CAS: 2432-99-7
Dataset id: 29 (Training set)
SMILES: O=C(O)CCCCCCCCCCN
Similarity: 0.767

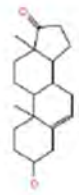
Experimental value: Carcinogen
Predicted value: Possible NON-Carcinogen

Compound #3

CAS: 75881-20-8
Dataset id: 489 (Training set)
SMILES: O=NN(C)CCCCCCCCCCCCCC
Similarity: 0.767

Experimental value: Carcinogen
Predicted value: Carcinogen

Alerts (not found in the target): Carcinogenity alert no. 1; Carcinogenity alert no. 14; Carcinogenity alert no. 27



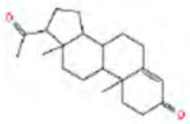
Compound #4

CAS: 53-43-0
Dataset id: 626 (Training set)
SMILES: O=C2CCC3C4CC=C1CC(O)CCC1(C)C4(CCC23(C))
Similarity: 0.764

Experimental value: Carcinogen
Predicted value: Carcinogen

Alerts (found also in the target): Carcinogenity alert no. 39

Alerts (not found in the target): Carcinogenity alert no. 5



Compound #5

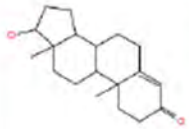
CAS: 57-83-0
Dataset id: 899 (Training set)
SMILES: O=C4C=C3CCC1C(CCC2(C)C(C(C(=O)C)CCC12))C3(C)CC4
Similarity: 0.764

Experimental value: Carcinogen
Predicted value: Carcinogen

Alerts (found also in the target): Carcinogenity alert no. 39

Alerts (not found in the target): Carcinogenity alert no. 5

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



Compound #6

CAS: 58-22-0
Dataset id: 908 (Training set)
SMILES: O=C4C=C3CCC1C(CCC2(C)(C(O)CCC12))C3(C)CC4
Similarity: 0.762

Experimental value: Carcinogen
Predicted value: Carcinogen

Alerts (found also in the target): Carcinogenity alert no. 39

Alerts (not found in the target): Carcinogenity alert no. 5

3.2 Applicability Domain:
Measured Applicability Domain Scores

Global AD Index
AD index = 0.716
Explanation: the predicted compound could be out of the Applicability Domain of the model.

Similar molecules with known experimental value
Similarity index = 0.769
Explanation: only moderately similar compounds with known experimental value in the training set have been found.

Accuracy of prediction for similar molecules
Accuracy index = 0.668
Explanation: accuracy of prediction for similar molecules found in the training set is not optimal.

Concordance for similar molecules
Concordance index = 0.664
Explanation: some similar molecules found in the training set have experimental values that disagree with the predicted value.

Atom Centered Fragments similarity check
ACF index = 1
Explanation: all atom centered fragment of the compound have been found in 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.

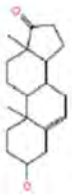
4.1 Reasoning:
Relevant Chemical Fragments and Moieties

(Molecule 0) Reasoning on fragments/structural alerts:

Fragment found: Carcinogenity alert no. 39

Structural alert for carcinogenity defined by the SMARTS: C(=CCC)CC

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

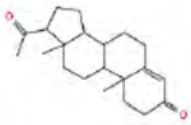


CAS: 53-43-0
Dataset id: 626 (Training set)
SMILES: O=C2CCC3C4CC=C1CC(O)CCC1(C)C4(CCC23(C))
Similarity: 0.764

Experimental value: Carcinogen
Predicted value: Carcinogen

Alerts (found also in the target): Carcinogenity alert no. 39

Alerts (not found in the target): Carcinogenity alert no. 5

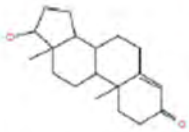


CAS: 57-83-0
Dataset id: 899 (Training set)
SMILES: O=C4C=C3CCC1C(CCC2(C)C(C(=O)C)CCC12))C3(C)CC4
Similarity: 0.764

Experimental value: Carcinogen
Predicted value: Carcinogen

Alerts (found also in the target): Carcinogenity alert no. 39

Alerts (not found in the target): Carcinogenity alert no. 5



CAS: 58-22-0
Dataset id: 908 (Training set)
SMILES: O=C4C=C3CCC1C(CCC2(C)C(O)CCC12))C3(C)CC4
Similarity: 0.762

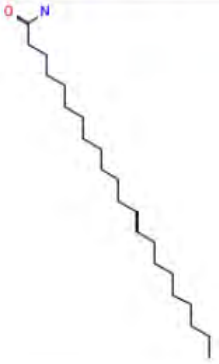
Experimental value: Carcinogen
Predicted value: Carcinogen

Alerts (found also in the target): Carcinogenity alert no. 39

Alerts (not found in the target): Carcinogenity alert no. 5

1. Prediction Summary

Prediction for compound Molecule 0



Prediction: Reliability:

Prediction is Carcinogen, but the result shows some critical aspects, which require to be checked:
- only moderately similar compounds with known experimental value in the training set have been found
- accuracy of prediction for similar molecules found in the training set is not adequate
- similar molecules found in the training set have experimental values that disagree with the predicted value

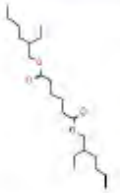
Compound: Molecule 0
Compound SMILES: O=C(N)CCCCCCCCCCCC=CCCCCCCCC
Experimental value: -
Predicted Oral Carcinogenic class: Carcinogen
Reliability: the predicted compound could be out of the Applicability Domain of the model
Remarks:
none

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

Compound #1

CAS: 2439-10-3
Dataset id: 490 (Training set)
SMILES: N(=C(N)N)CCCCCCCCCCCC
Similarity: 0.726

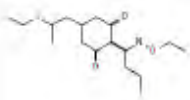
Experimental value: NON-Carcinogen
Predicted value: NON-Carcinogen



Compound #2

CAS: 103-23-1
Dataset id: 94 (Training set)
SMILES: O=C(OCC(CC)CCCC)CCCCC(=O)OCC(CC)CCCC
Similarity: 0.715

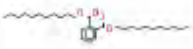
Experimental value: Carcinogen
Predicted value: NON-Carcinogen



Compound #3

CAS: 74051-80-2
Dataset id: 674 (Training set)
SMILES: O=C1C(C(=O)CC(C1)CC(C)SCC)=C(NOCC)CCC
Similarity: 0.702

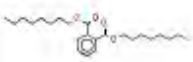
Experimental value: NON-Carcinogen
Predicted value: NON-Carcinogen



Compound #4

CAS: 3648-20-2
Dataset id: 488 (Training set)
SMILES: O=C(OCCCCCCCCCCC)c1ccccc1(C(=O)OCCCCCCCCCCC)
Similarity: 0.668

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



Compound #5

CAS: 117-84-0
Dataset id: 614 (Training set)
SMILES: O=C(OCCCCCCCC)c1ccccc1(C(=O)OCCCCCCCC)
Similarity: 0.66

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

Compound #6

CAS: 78-42-2
Dataset id: 313 (Training set)
SMILES: O=P(OCC(CC)CCCC)(OCC(CC)CCCC)OCC(CC)CCCC
Similarity: 0.653

Experimental value: Carcinogen
Predicted value: Carcinogen

3.2 Applicability Domain:
Measured Applicability Domain Scores

Global AD Index
AD index = 0.6
Explanation: the predicted compound could be out of the Applicability Domain of the model.

Similar molecules with known experimental value
Similarity index = 0.72
Explanation: only moderately similar compounds with known experimental value in the training set have been found.

Accuracy of prediction for similar molecules
Accuracy index = 0.506
Explanation: accuracy of prediction for similar molecules found in the training set is not adequate.

Concordance for similar molecules
Concordance index = 0.494
Explanation: similar molecules found in the training set have experimental values that disagree with the predicted value.

Model's 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.

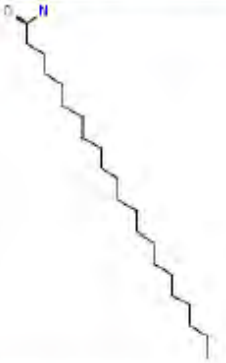
Atom Centered Fragments similarity check
ACF index = 1
Explanation: all atom centered fragment of the compound have been found in the compounds of the training set.

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.

1. Prediction Summary

Prediction for compound Molecule 0



Prediction: Reliability:

Prediction is Carcinogen, but the result may be not reliable. A 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
- accuracy of prediction for similar molecules found in the training set is not adequate
- similar molecules found in the training set have experimental values that disagree with the predicted value

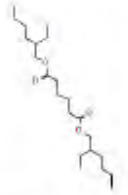
Compound: Molecule 0
Compound SMILES: O=C(N)CCCCCCCCCCCC=CCCCCCCCC
Experimental value: -
Predicted Inhalation Carcinogenic class: Carcinogen
Reliability: the predicted compound is outside the Applicability Domain of the model
Remarks:
none

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

Compound #1

CAS: 2439-10-3
Dataset id: 462 (Training set)
SMILES: N(=C(N)N)CCCCCCCCCCCC
Similarity: 0.726

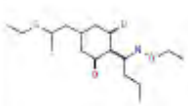
Experimental value: NON-Carcinogen
Predicted value: Carcinogen



Compound #2

CAS: 103-23-1
Dataset id: 391 (Training set)
SMILES: O=C(OCC(CC)CCCC)CCCCC(=O)OCC(CC)CCCC
Similarity: 0.715

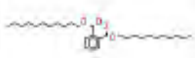
Experimental value: NON-Carcinogen
Predicted value: NON-Carcinogen



Compound #3

CAS: 74051-80-2
Dataset id: 664 (Training set)
SMILES: O=C1C(C(=O)CC(C1)CC(C)SCC)=C(NOCC)CCC
Similarity: 0.702

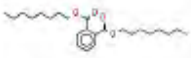
Experimental value: NON-Carcinogen
Predicted value: NON-Carcinogen



Compound #4

CAS: 3648-20-2
Dataset id: 460 (Test set)
SMILES: O=C(OCCCCCCCCCCC)c1ccccc1(C(=O)OCCCCCCCCCCC)
Similarity: 0.668

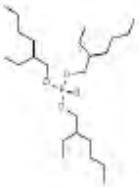
Experimental value: NON-Carcinogen
Predicted value: NON-Carcinogen



Compound #5

CAS: 117-84-0
Dataset id: 597 (Training set)
SMILES: O=C(OCCCCCCCC)c1ccccc1(C(=O)OCCCCCCCC)
Similarity: 0.66

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



Compound #6

CAS: 78-42-2
Dataset id: 741 (Training set)
SMILES: O=P(OCC(CC)CCCC)(OCC(CC)CCCC)OCC(CC)CCCC
Similarity: 0.653

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

3.2 Applicability Domain:
Measured Applicability Domain Scores

Global AD Index
AD index = 0
Explanation: the predicted compound is outside the Applicability Domain of the model.

Similar molecules with known experimental value
Similarity index = 0.72
Explanation: only moderately similar compounds with known experimental value in the training set have been found.

Accuracy of prediction for similar molecules
Accuracy index = 0.494
Explanation: accuracy of prediction for similar molecules found in the training set is not adequate.

Concordance for similar molecules
Concordance index = 0
Explanation: similar molecules found in the training set have experimental values that disagree with the predicted value.

Model's 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.

Atom Centered Fragments similarity check
ACF index = 1
Explanation: all atom centered fragment of the compound have been found in 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.

APPENDIX F: Danish QSAR Predictions Output for Erucamide (CAS #112-84-5)

Carcinogenicity

	E Ultra	Leadscope
FDA RCA Cancer Male Rat	NEG_IN	NEG_IN
FDA RCA Cancer Female Rat	NEG_IN	NEG_IN
FDA RCA Cancer Rat	NEG_IN	NEG_IN
FDA RCA Cancer Male Mouse	NEG_IN	NEG_IN
FDA RCA Cancer Female Mouse	NEG_IN	NEG_IN
FDA RCA Cancer Mouse	NEG_IN	NEG_IN
FDA RCA Cancer Rodent	NEG_IN	NEG_IN

Commercial models from CASE Ultra and Leadscope

FDA RCA: Data from US Food and Drug Administration as part of Research Cooperation Agreement

Carcinogenicity (genotox and nongenotox) alerts by ISS, alerts in:	
- parent only	No alert found
Oncologic Primary Classification, alerts in:	
- parent only	Not classified

OECD QSAR Toolbox v.4.2 profilers

Profiler predictions are supporting information to be used together with the relevant QSAR predictions

	Exp	Battery	CASE Ultra	Leadscope	SciQSAR
Liver Specific Cancer in Rat or Mouse		NEG_IN	NEG_IN	NEG_OUT	NEG_IN

DTU-developed models

Endocrine and Molecular Endpoints

	Exp	Battery	CASE Ultra	Leadscope	SciQSAR
Estrogen Receptor α Binding, Full training set (Human *in vitro*)		INC_OUT	INC_OUT	NEG_OUT	NEG_OUT
Estrogen Receptor α Binding, Balanced Training Set (Human *in vitro*)		INC_OUT	INC_OUT	POS_OUT	NEG_OUT
Estrogen Receptor α Activation (Human *in vitro*)		INC_OUT	INC_OUT	NEG_OUT	POS_OUT
Estrogen Receptor Activation, CERAPP data (*in vitro*)		N/A	N/A	NEG_IN	N/A
Androgen Receptor Inhibition (Human *in vitro*)		NEG_IN	NEG_OUT	NEG_IN	NEG_IN
Androgen Receptor Binding, CoMPARA data (*in vitro*)		N/A	N/A	INC_OUT	N/A
Androgen Receptor Inhibition, CoMPARA data (*in vitro*)		N/A	N/A	NEG_OUT	N/A
Androgen Receptor Activation, CoMPARA data (*in vitro*)		N/A	N/A	NEG_IN	N/A
Thyroperoxidase (TPO) inhibition QSAR1 (Rat *in vitro*)		N/A	N/A	NEG_IN	N/A
Thyroperoxidase (TPO) inhibition QSAR2 (Rat *in vitro*)		N/A	N/A	NEG_IN	N/A
Thyroid Receptor α Binding (Human *in vitro*)					
- mg/L				1318.529	
- µM				3905.711	
- Positive for IC_{50} ≤ 10 µM					
- Positive for IC_{50} ≤ 100 µM					
- Domain				OUT	OUT
Thyroid Receptor β Binding (Human *in vitro*)					
- mg/L				12.76055	
- µM				37.79897	
- Positive for IC_{50} ≤ 10 µM					
- Positive for IC_{50} ≤ 100 µM					
- Domain				OUT	OUT
Arylhydrocarbon (AhR) Activation – Rational final model (Human *in vitro*)		N/A	N/A	INC_OUT	N/A
Arylhydrocarbon (AhR) Activation –		N/A	N/A	INC_OUT	N/A

	Exp	Battery	CASE Ultra	Leadscope	SciQSAR
Random final model (Human *in vitro*)					
Pregnane X Receptor (PXR) Binding (Human *in vitro*)	N/A	INC_OUT	POS_OUT	INC_OUT	NEG_OUT
Pregnane X Receptor (PXR) Binding (Human *in vitro*) NEW		N/A	N/A	INC_OUT	N/A
Pregnane X Receptor (PXR) Activation (Human *in vitro*)		N/A	N/A	NEG_IN	N/A
Pregnane X Receptor (PXR) Activation (Rat *in vitro*)		N/A	N/A	NEG_IN	N/A
Constitutive Androstane Receptor (CAR) Activation at max. 20 µM (*in vitro*)		N/A	N/A	NEG_IN	N/A
Constitutive Androstane Receptor (CAR) Activation at max. 50 µM (*in vitro*)		N/A	N/A	NEG_IN	N/A
Constitutive Androstane Receptor (CAR) Inhibition at max. 20 µM (*in vitro*)		N/A	N/A	NEG_IN	N/A
Constitutive Androstane Receptor (CAR) Inhibition at max. 50 µM (*in vitro*)		N/A	N/A	NEG_IN	N/A
CYP3A4 Induction (Human *in vitro*)		N/A	N/A	NEG_IN	N/A

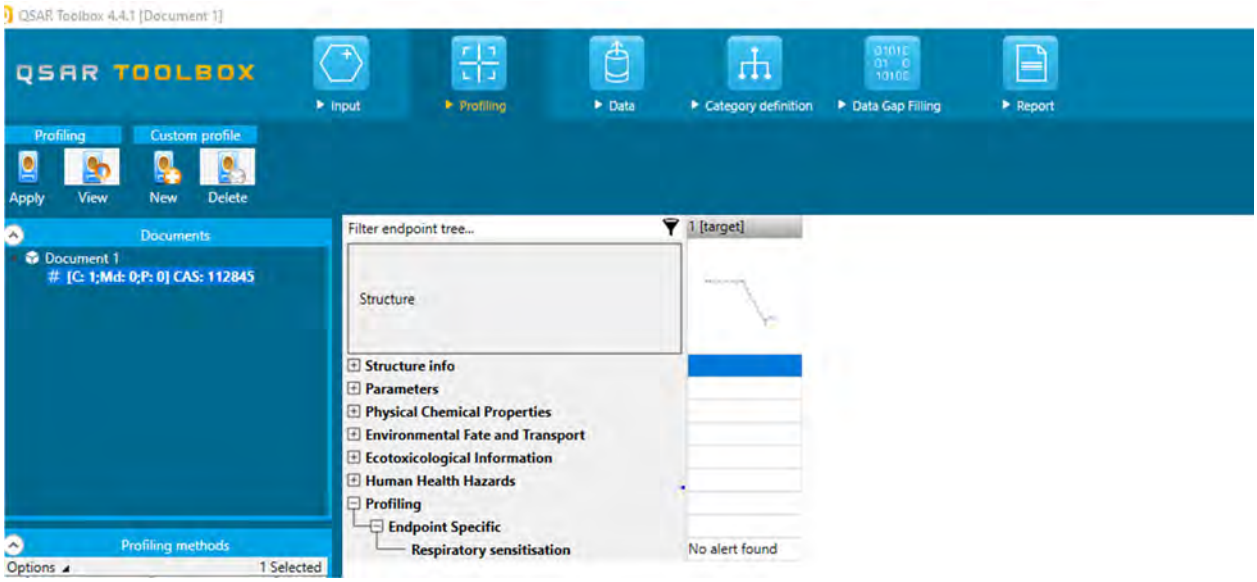
DTU-developed models

Estrogen Receptor Binding, alerts in:	
- parent only	Non binder, non cyclic structure
- metabolites from *in vivo* Rat metabolism simulator only	Non binder, non cyclic structure
- metabolites from Rat liver S9 metabolism simulator only	Non binder, non cyclic structure
rtER Expert System - USEPA, alerts in:	
- parent only	No alert found
- metabolites from *in vivo* Rat metabolism simulator only	No alert found
- metabolites from Rat liver S9 metabolism simulator only	No alert found

OECD QSAR Toolbox v.4.2 profilers

Profiler predictions are supporting information to be used together with the relevant QSAR predictions

APPENDIX G: OECD Toolbox Respiratory Sensitization Results for Erucamide (CAS #112-84-5)



APPENDIX H: EPI Suite™ Modeling Results for Erucamide (CAS #112-84-5)

(Estimated values included in the GreenScreen® are highlighted and bolded)

CAS Number: 000112-84-5
SMILES : O=C(N)CCCCCCCCCCCCC=CCCCCCCCC
CHEM : 13-DECOSENAMIDE (CIS)
MOL FOR: C22 H43 N1 O1
MOL WT : 337.59
---------- EPI SUMMARY (v4.11) ----------
Physical Property Inputs:
Log Kow (octanol-water): 8.00
Boiling Point (deg C) : ------
Melting Point (deg C) : 77.50
Vapor Pressure (mm Hg) : ------
Water Solubility (mg/L): ------
Henry LC (atm-m3/mole) : ------

Log Octanol-Water Partition Coef (SRC):
Log Kow (KOWWIN v1.69 estimate) = 8.44

Boiling Pt, Melting Pt, Vapor Pressure Estimations (MPBPVP v1.43):
Boiling Pt (deg C): 461.05 (Adapted Stein & Brown method)
Melting Pt (deg C): 183.35 (Mean or Weighted MP)
VP(mm Hg,25 deg C): 8.28E-008 (Modified Grain method)
VP (Pa, 25 deg C) : 1.1E-005 (Modified Grain method)
MP (exp database): 77.5 deg C
Subcooled liquid VP: 2.61E-007 mm Hg (25 deg C, Mod-Grain method)
: 3.48E-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): 0.0009962
log Kow used: 8.00 (user entered)
melt pt used: 77.50 deg C

Water Sol Estimate from Fragments:
Wat Sol (v1.01 est) = 0.00060783 mg/L

ECOSAR Class Program (ECOSAR v1.11):
Class(es) found:
Amides

Henrys Law Constant (25 deg C) [HENRYWIN v3.20]:
Bond Method : 2.84E-006 atm-m3/mole (2.88E-001 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: 3.692E-005 atm-m3/mole (3.741E+000 Pa-m3/mole)

VP: 8.28E-008 mm Hg (source: MPBPVP)
WS: 0.000996 mg/L (source: WSKOWWIN)

Log Octanol-Air Partition Coefficient (25 deg C) [KOAWIN v1.10]:
Log Kow used: 8.00 (user entered)
Log Kaw used: -3.935 (HenryWin est)
Log Koa (KOAWIN v1.10 estimate): 11.935
Log Koa (experimental database): None

Probability of Rapid Biodegradation (BIOWIN v4.10):
Biowin1 (Linear Model) : 0.9054
Biowin2 (Non-Linear Model) : 0.9399
Expert Survey Biodegradation Results:
Biowin3 (Ultimate Survey Model): 2.6973 (weeks-months)
Biowin4 (Primary Survey Model) : 3.8352 (days)
MITI Biodegradation Probability:
Biowin5 (MITI Linear Model) : 0.5396
Biowin6 (MITI Non-Linear Model): 0.5340
Anaerobic Biodegradation Probability:
Biowin7 (Anaerobic Linear Model): 0.1917
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): 3.48E-005 Pa (2.61E-007 mm Hg)
Log Koa (Koawin est): 11.935
Kp (particle/gas partition coef. (m3/ug)):
Mackay model : 0.0862
Octanol/air (Koa) model: 0.211
Fraction sorbed to airborne particulates (phi):
Junge-Pankow model : 0.757
Mackay model : 0.873
Octanol/air (Koa) model: 0.944

Atmospheric Oxidation (25 deg C) [AopWin v1.92]:
Hydroxyl Radicals Reaction:
OVERALL OH Rate Constant = 85.7254 E-12 cm3/molecule-sec [Cis-isomer]
OVERALL OH Rate Constant = 93.3254 E-12 cm3/molecule-sec [Trans-isomer]
Half-Life = 1.497 Hrs (12-hr day; 1.5E6 OH/cm3) [Cis-isomer]
Half-Life = 1.375 Hrs (12-hr day; 1.5E6 OH/cm3) [Trans-isomer]
Ozone Reaction:
OVERALL Ozone Rate Constant = 13.000000 E-17 cm3/molecule-sec [Cis-]
OVERALL Ozone Rate Constant = 20.000000 E-17 cm3/molecule-sec [Trans-]
Half-Life = 2.116 Hrs (at 7E11 mol/cm3) [Cis-isomer]
Half-Life = 1.375 Hrs (at 7E11 mol/cm3) [Trans-isomer]
Reaction With Nitrate Radicals May Be Important!
Fraction sorbed to airborne particulates (phi):

0.815 (Junge-Pankow, Mackay avg)
0.944 (Koa method)
Note: the sorbed fraction may be resistant to atmospheric oxidation

Soil Adsorption Coefficient (KOCWIN v2.00):
Koc : 5.103E+005 L/kg (MCI method)
Log Koc: 5.708 (MCI method)
Koc : 2.22E+005 L/kg (Kow method)
Log Koc: 5.346 (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 = 3.038 (BCF = 1090 L/kg wet-wt)
Log Biotransformation Half-life (HL) = 0.3730 days (HL = 2.361 days)
Log BCF Arnot-Gobas method (upper trophic) = 1.655 (BCF = 45.23)
Log BAF Arnot-Gobas method (upper trophic) = 2.606 (BAF = 403.3)
log Kow used: 8.00 (user entered)

Volatilization from Water:
Henry LC: 2.84E-006 atm-m3/mole (estimated by Bond SAR Method)
Half-Life from Model River: 380.7 hours (15.86 days)
Half-Life from Model Lake : 4307 hours (179.4 days)

Removal In Wastewater Treatment:
Total removal: 94.02 percent
Total biodegradation: 0.78 percent
Total sludge adsorption: 93.24 percent
Total to Air: 0.00 percent
(using 10000 hr Bio P,A,S)

Level III Fugacity Model: (MCI Method)
** Note: When the Log Kow is > 7, the model may be underestimating the mass of material in sediment and overestimating the mass of material in the water column (biota). Consider using the results of the default EQC model. **

	Mass Amount (percent)	Half-Life (hr)	Emissions (kg/hr)
Air	0.0329	1.24	1000
Water	8.67	900	1000
Soil	52.2	1.8e+003	1000
Sediment	39.1	8.1e+003	0

Persistence Time: 1.72e+003 hr

Level III Fugacity Model: (MCI Method with Water percents)

	Mass Amount (percent)	Half-Life (hr)	Emissions (kg/hr)

Air	0.0329	1.24	1000
Water	8.67	900	1000
water	(1.28)		
biota	(6.41)		
suspended sediment	(0.981)		
Soil	52.2	1.8e+003	1000
Sediment	39.1	8.1e+003	0

Persistence Time: 1.72e+003 hr

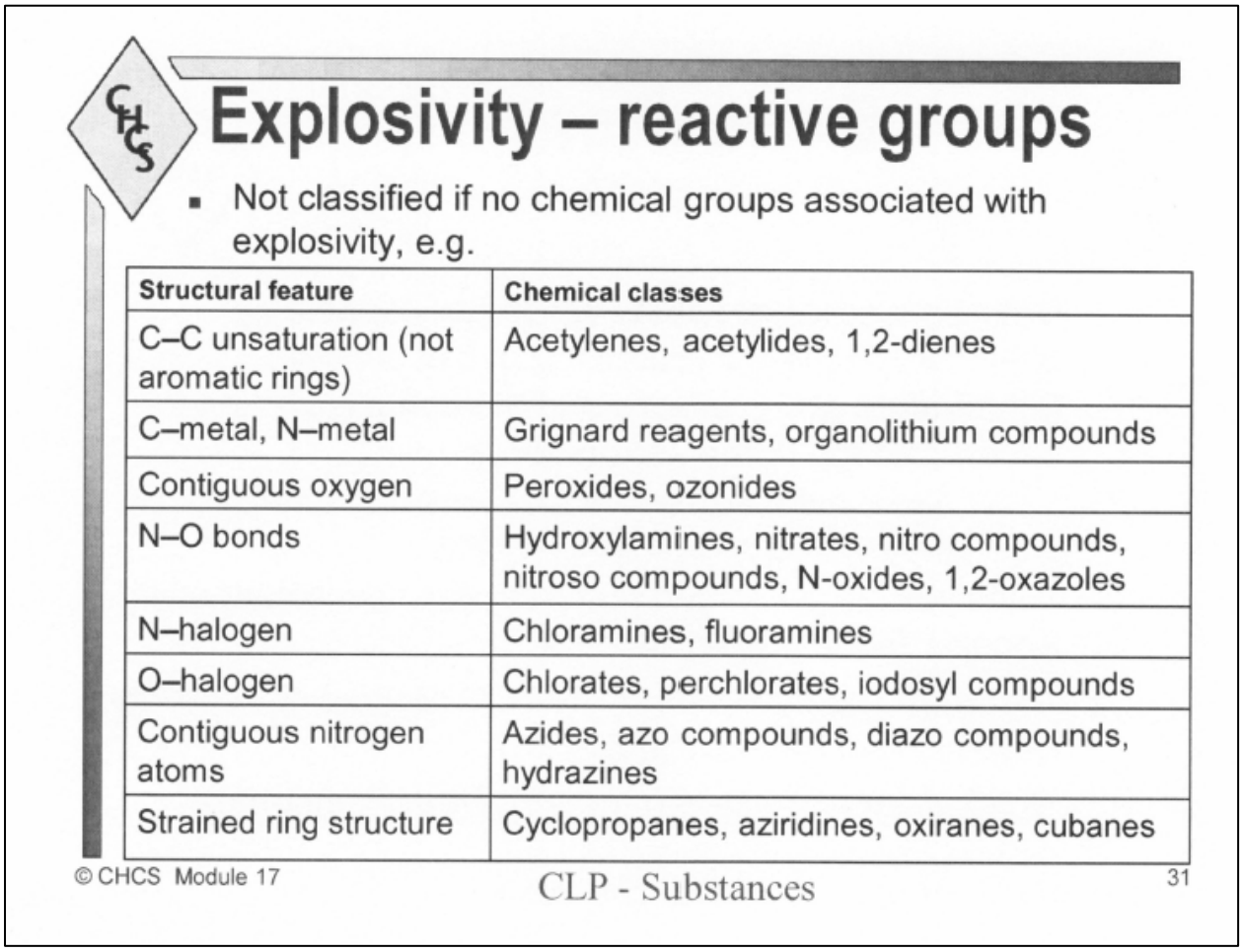
Level III Fugacity Model: (EQC Default)

	Mass Amount (percent)	**Half-Life (hr)**	**Emissions (kg/hr)**
Air	**0.018**	**1.24**	**1000**
Water	**1.91**	**900**	**1000**
water	**(0.0283)**		
biota	**(0.142)**		
suspended sediment	**(1.74)**		
Soil	**28.7**	**1.8e+003**	**1000**
Sediment	**69.4**	**8.1e+003**	**0**

Persistence Time: 3.13e+003 hr

APPENDIX I: Known Structural Alerts for Reactivity

Explosivity – Abbreviated List



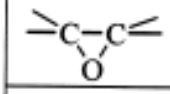
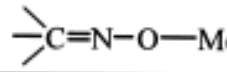
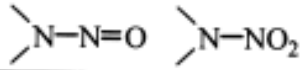
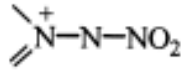
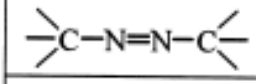
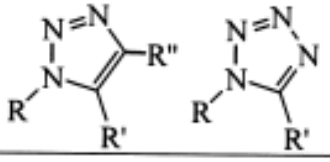
Explosivity – reactive groups

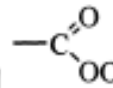
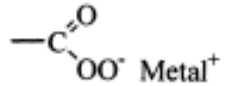
- Not classified if no chemical groups associated with explosivity, e.g.

Structural feature	Chemical classes
C–C unsaturation (not aromatic rings)	Acetylenes, acetylides, 1,2-dienes
C–metal, N–metal	Grignard reagents, organolithium compounds
Contiguous oxygen	Peroxides, ozonides
N–O bonds	Hydroxylamines, nitrates, nitro compounds, nitroso compounds, N-oxides, 1,2-oxazoles
N–halogen	Chloramines, fluoramines
O–halogen	Chlorates, perchlorates, iodosyl compounds
Contiguous nitrogen atoms	Azides, azo compounds, diazo compounds, hydrazines
Strained ring structure	Cyclopropanes, aziridines, oxiranes, cubanes

Explosivity – Full List

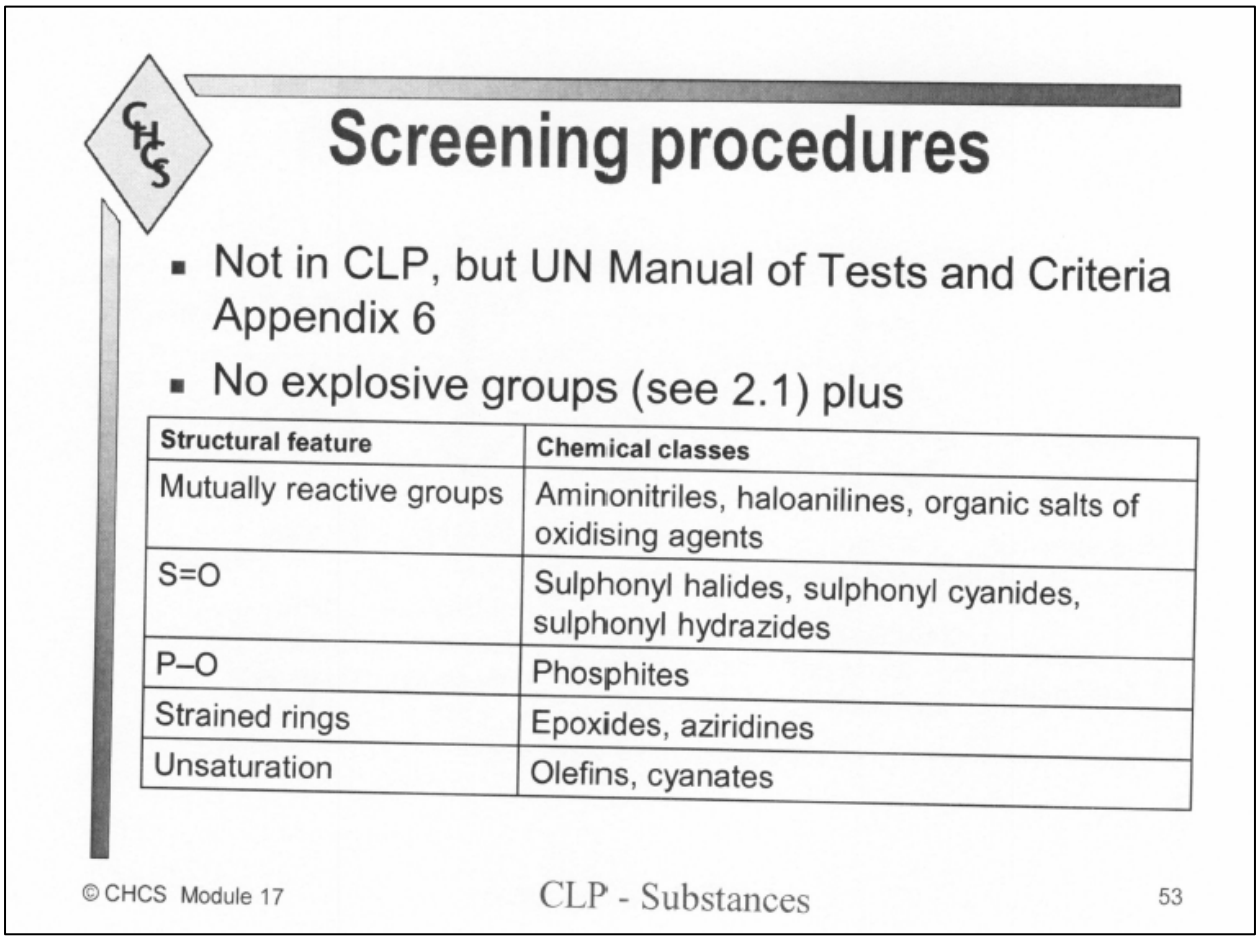
Table R.7.1-28 Chemical groups associated with explosive properties

Chemical group	Chemical Class
-C≡C- -C≡C-Metal -C≡C-Halogen	Acetylenic Compounds Metal Acetylides Haloacetylene Derivatives
	Diazo Compounds
-N=O -NO_2	Nitroso and Nitro Compounds,
R-O-N=O R-O-NO_2	Acyl or Alkyl Nitrites and Nitrates
	1,2-Epoxides
	Metal Fulminates or *aci*-Nitro Salts
	N-Metal Derivatives (especially heavy metals)
	N-Nitroso and N-Nitro Compounds
	N-Azolium Nitroimidates
	Azo Compounds
Ar-N=N-O-Ar	Arene Diazoates
$(ArN=N)_2O$, $(ArN=N)_2S$	Bis-Arenediazo Oxides and Sulfides
RN=N-NR'R''	Triazines
	High-nitrogen Compounds: e.g. Triazoles, Tetrazoles

Chemical group	Chemical Class
[1] ROOR', [2] 	Peroxy Compounds: [1] Alkyl hydroperoxides (R'=H), Peroxides (R'=organic); [2] Peroxo acids (R'=H), Peroxyesters (R'=organic)
[1] ROOMetal, [2] 	Metal peroxides, Peroxoacids salts
$-N_3$	Azides e.g. PbN_6, CH_3N_3
$^{-}O-C-N_2^{+}$	Arenediazonium oxides i.e. inner diazonium salts in which the counter ion is an oxide
Ar-N=N-S- Ar-N=N-S-Ar	Diazonium sulfides and derivatives, Arenediazo Aryl Sulfides
XO_n	Halogen Oxide: e.g. percholrates, bromates, etc
NX_3 e.g. $NC1_3$, $RNC1_2$	N-Halogen Compounds

Adapted from Bretherick (Bretherick's Handbook of Reactive Chemical Hazards 6th Ed., 1999, Butterworths, London)

Self-Reactive Substances



Screening procedures

- Not in CLP, but UN Manual of Tests and Criteria Appendix 6
- No explosive groups (see 2.1) plus

Structural feature	Chemical classes
Mutually reactive groups	Aminonitriles, haloanilines, organic salts of oxidising agents
S=O	Sulphonyl halides, sulphonyl cyanides, sulphonyl hydrazides
P–O	Phosphites
Strained rings	Epoxides, aziridines
Unsaturation	Olefins, cyanates

APPENDIX J: Change in Benchmark Score

Table 4 provides a summary of changes to the GreenScreen® Benchmark™ for erucamide. This GreenScreen® has undergone three rounds of updates and the benchmark score changed from Benchmark 2 to Benchmark U and currently 2DG.

Table 4: Change in GreenScreen® Benchmark™ for Erucamide

Date	GreenScreen® Benchmark™	GreenScreen® Version	Comment
April 28, 2011	BM-2	v. 1.0	New assessment
February 19, 2016	BM-U	v. 1.2	Updated evaluation to the v1.2 Criteria, benchmark score changed to U due to data gap for carcinogenicity
August 11, 2021	BM-2_{DG}	v. 1.4	Updated evaluation to the v1.4 Criteria, benchmark score changed to 2DG due to data gaps for reproductive toxicity and endocrine activity
November 12, 2021	BM-2_{DG}	v. 1.4	Minor updates to various endpoints without changing hazard or benchmark scores.

Licensed GreenScreen® Profilers

Erucamide GreenScreen® Evaluation (v.1.1) Prepared by:

SIGNATURE BLOCK

Kristen Schaefer, M.F.S.
Associate Toxicologist
ToxServices LLC

Erucamide GreenScreen® Evaluation (v.1.1) QC'd by:

SIGNATURE BLOCK

Margaret H. Whittaker, Ph.D., M.P.H., CBiol., F.R.S.B., E.R.T., D.A.B.T.
Managing Director and Chief Toxicologist
ToxServices LLC

Erucamide GreenScreen® Evaluation (v.1.2) Updated by:

SIGNATURE BLOCK

Sara M. Ciotti, Ph.D.
Toxicologist
ToxServices LLC

Erucamide GreenScreen® Evaluation Update (v.1.2) QC'd by:

SIGNATURE BLOCK

Bingxuan Wang, Ph.D., D.A.B.T.
Toxicologist
ToxServices LLC

Erucamide GreenScreen® Evaluation (v.1.4) Updated by:

SIGNATURE BLOCK

Megan B. Boylan, M.S.
Toxicologist
ToxServices LLC

Erucamide GreenScreen® Evaluation Update (v.1.4) QC'd by:

SIGNATURE BLOCK

Bingxuan Wang, Ph.D., D.A.B.T.
Senior Toxicologist
ToxServices LLC