N-ISOPROPYL-N'-PHENYL-P-PHENYLENEDIAMINE (IPPD) (CAS #101-72-4) GREENSCREEN® FOR SAFER CHEMICALS (GREENSCREEN®) ASSESSMENT

Prepared by:

ToxServices LLC

Assessment Date: October 14, 2021

ToxServices Review Date: October 14, 2026[1]

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[1] Although CPA's Assessment Expiration Policy (CPA 2018a) indicates that Benchmark 1 assessments have no expiration date, ToxServices strives to review BM-1s in a five-year period to ensure currency of data presented in the BM-1 GreenScreen® assessments.

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GreenScreen® Executive Summary for N-Isopropyl-N'-phenyl-p-phenylenediamine (IPPD) (CAS #101-72-4)

N-Isopropyl-N'-phenyl-p-phenylenediamine (IPPD) is used as an antioxidant and stabilizer and is used to counteract the degradation of rubber. It is manufactured by the reaction of p-chloronitrobenzene with aniline to yield p-nitrodiphenylamine, which is reductively alkylated with acetone over a nickel/chromium catalyst. IPPD is a solid at room temperature. Its vapor pressure and boiling point indicate it may volatilize. It has low water solubility, and its log K_{ow} indicates it is not expected to bioaccumulate.

IPPD was assigned a **GreenScreen Benchmark™ Score of 1** ("Avoid – Chemical of High Concern"). This score is based on the following hazard score:

- Benchmark 1e
    - High Group I Human Toxicity (reproductive toxicity-R)

A data gap (DG) exists for neurotoxicity (repeated dose)-Nr*. As outlined in GreenScreen® Guidance Section 11.6.2.1 and Annex 5 (Conduct a Data Gap Analysis), IPPD meets requirements for a GreenScreen Benchmark™ Score of 1 despite the hazard data gap. In a worst-case scenario, if IPPD were assigned a High score for the data gap Nr*, it would still 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, aquatic toxicity, persistence and bioaccumulation, and *in vitro* testing for carcinogenicity, genotoxicity, and endocrine activity. 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 IPPD's NAMs dataset include the lack of sufficient data on carcinogenicity, endocrine activity, respiratory sensitization, chronic aquatic toxicity, and persistence along with a lack of validated test methods for respiratory sensitization.

Type II (extrapolation output) uncertainties in IPPD's NAMs dataset include limitations of modeling software Toxtree and OECD Toolbox in identifying structural alerts without defining applicability domains, the limitations of *in vitro* genotoxicity and carcinogenicity assays in mimicking *in vivo* metabolic systems, the uncertain *in vivo* relevance of *in silico* modeling of endocrine receptor binding and *in vitro* high throughput testing data, the limitations in the examination of structural alerts for respiratory sensitization evaluation that does not account for non-immunologic mechanisms of respiratory sensitization, the lack of guidance from the ECHA framework to subclassify respiratory sensitizers to Category 1A and 1B, and the unreliable predictions of chronic aquatic toxicity by ECOSAR. Some of IPPD'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 IPPD

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**	*H*	**M**	*M*	**M**		**M**	*M*	DG	**H**	*M*	**L**	*M*	**vH**	*vH*	*H*	**vL**	*L*	**L**

Note: Hazard levels (Very High (vH), High (H), Moderate (M), Low (L), Very Low (vL)) in *italics* reflect 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 N-Isopropyl-N'-phenyl-p-phenylenediamine (IPPD) (CAS #101-72-4)

Method Version: GreenScreen® Version 1.4
Assessment Type[2]: Certified
Assessor Type: Licensed GreenScreen® Profiler

GreenScreen® Assessment (v.1.4) Prepared By:
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Date: October 13, 2021

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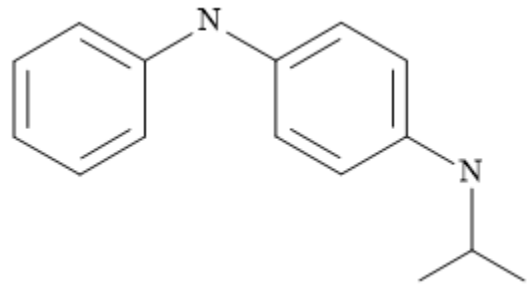
Organization: ToxServices LLC
Date: October 14, 2021

ToxServices Review Date: October 14, 2026[3]

Chemical Name: N-Isopropyl-N'-phenyl-p-phenylenediamine

CAS Number: 101-72-4

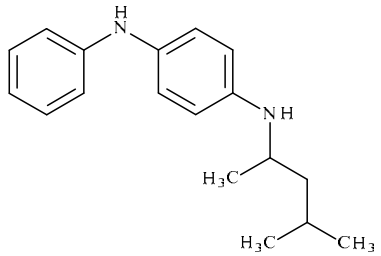
Chemical Structure(s):



Also called: 4-(Isopropylamino)diphenylamine; 1,4-Benzenediamine, N-(1-methylethyl)-N'-phenyl-; 4-Anilino-N-isopropylaniline; 4-Isopropylaminodiphenylamine; 4010 NA; 4010NA; Antigen 3c; Antigene 3C; Antioxidant 4010 NA; Antioxidant 4010NA; Antioxidant 40NA; Antioxidant IP; ASM 4010MA; BRN 2213195; Cyzone; Cyzone IP; Diafen FP; Diaphen FP; Elastozone 34; Flexzone 3C; Ipognox 44; IPPD; N-2-Propyl-N'-phenyl-p-phenylenediamine; N-Isopropyl-N'-phenyl-para-phenylenediamine; N-Phenyl-N'-isopropyl-1,4-phenylenediamine; N-Phenyl-N'-isopropyl-p-phenylenediamine; NA 4010; Nocrac 810NA; Nocrack 810NA; Nonox ZA; NSC 41029; Orflex PP; Ozonon 3C; p-Phenylenediamine, N-isopropyl-N'-phenyl-para-Isopropylaminodiphenylamine; Permanax 115; S-IP; Santoflex 36; Santoflex IP; 1,4-Benzenediamine, N-(1-methylethyl)-N'-phenyl-; 1,4-Benzenediamine, N1-(1-methylethyl)-N4-phenyl-; N-(1-Methylethyl)-N'-phenyl-1,4-benzenediamine; N-Isopropyl-N'-phenyl-1,4-phenylenediamine; p-Phenylenediamine, N-isopropyl-N'-phenyl- (ChemIDplus 2021).

[2] GreenScreen® reports are either "UNACCREDITED" (by unaccredited person), "AUTHORIZED" (by Authorized GreenScreen® Practitioner), or "CERTIFIED" (by Licensed GreenScreen® Profiler or equivalent).
[3] Although CPA's Assessment Expiration Policy (CPA 2018a) indicates that Benchmark 1 assessments have no expiration date, ToxServices strives to review BM-1s in a five-year period to ensure currency of data presented in the BM-1 GreenScreen® assessments.

Suitable surrogates or moieties of chemicals used in this assessment (CAS #'s):
N-isopropyl-N'-phenyl-p-phenylenediamine has a relatively complete toxicological dataset. ToxServices identified N-(1,3-dimethylbutyl)-N'-phenyl-p-phenylenediamine (6PPD) (CAS #793-24-8) as a surrogate, as both compounds are dibenzenediamines connected with a branched alkyl group. The surrogate 6PPD is slightly larger than IPPD as it contains three additional carbons in the alkyl group. The Australian Industrial Chemicals Introduction Scheme (AICIS) used both 6PPD and IPPD as surrogate in its public report on another structurally similar chemical (AICIS 2009). Due to its slightly larger size, ToxServices considers 6PPD to be a weak surrogate.



Surrogate: 6PPD (CAS #793-24-8)

Identify Applications/Functional Uses (ECHA 2021a, Pharos 2021)**:**
1. Stabilizer
2. Antioxidant

Known Impurities[4]:
Chromium and nickel are used as catalysts and may be present as residuals (Pharos 2021). The screen is performed on the theoretical pure substance.

GreenScreen® Summary Rating for IPPD[5,6 7,8]: IPPD was assigned a **GreenScreen Benchmark™ Score of** ("Avoid – Chemical of High Concern") (CPA 2018b). This score is based on the following hazard score:

- Benchmark 1e
    - High Group I Human Toxicity (reproductive toxicity-R)

A data gap (DG) exists for neurotoxicity (repeated dose)-Nr*. As outlined in GreenScreen® Guidance Section 11.6.2.1 and Annex 5 (Conduct a Data Gap Analysis), IPPD meets requirements for a GreenScreen Benchmark™ Score of 1 despite the hazard data gap. In a worst-case scenario, if IPPD were assigned a High score for the data gap Nr*, it would still be categorized as a Benchmark 1 Chemical.

[4] Impurities of the chemical will be assessed at the product level instead of in this GreenScreen®.
[5] 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.
[6] See Appendix A for a glossary of hazard endpoint acronyms.
[7] For inorganic chemicals only, see GreenScreen® Guidance v1.4 Section 12 (Inorganic Chemical Assessment Procedure).
[8] 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 IPPD

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**	*H*	**M**	*M*	**M**		**M**	*M*	DG	**H**	*M*	**L**	*M*	**vH**	*vH*	*H*	**vL**	*L*	**L**

Note: Hazard levels (Very High (vH), High (H), Moderate (M), Low (L), Very Low (vL)) in *italics* reflect 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

Using OECD Toolbox, ToxServices predicted there would be no hydrolysis products (OECD 2021). ToxServices identified no other feasible and/or relevant environmental transformation products for IPPD.

Introduction

IPPD is used as an antioxidant and stabilizer and is used to counteract the degradation of rubber (ECHA 2021a, Pharos 2021). It is manufactured by the reaction of p-chloronitrobenzene with aniline to yield p-nitrodiphenylamine, which is reductively alkylated with acetone over a nickel/chromium catalyst (HSDB 2007). As IPPD is well known as a severe allergen, its use is usually avoided in non-industrial applications (UNEP 2002).

ToxServices assessed IPPD 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).

IPPD is not listed on the U.S. EPA 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),[9] 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 N-isopropyl-N'-phenyl-p-phenylenediamine can be found in Appendix C.

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

- N-Isopropyl-N'-phenyl-p-phenylenediamine is an LT-P1 chemical when screened using Pharos, and therefore a full GreenScreen® is required.
- N-Isopropyl-N'-phenyl-p-phenylenediamine is not listed on the U.S. DOT list.
- N-Isopropyl-N'-phenyl-p-phenylenediamine is on the following lists for multiple endpoints.
    - EU – GHS: H410 – Very toxic to aquatic life with long lasting effects (Hazardous to the aquatic environment (chronic) – Category 1)
    - Australia – GHS: H410 – Very toxic to aquatic life with long lasting effects (Hazardous to the aquatic environment (chronic) – Category 1)
    - Korea – GHS: H410 – Very toxic to aquatic life with long lasting effects (Hazardous to the aquatic environment (chronic) – Category 1)
    - Japan – GHS: H410 – Very toxic to aquatic life with long lasting effects (Hazardous to the aquatic environment (chronic) – Category 1)
    - New Zealand – GHS: 9.1A (algal) – very ecotoxic in the aquatic environment.
    - New Zealand – GHS: 9.1A (fish) – very ecotoxic in the aquatic environment.
    - New Zealand – GHS: 9.1B (crustacean) – very ecotoxic in the aquatic environment.
    - German FEA – Substances hazardous to waters: Class 3 – Severe hazard to waters
- Specified lists for single endpoints are reported in individual hazard endpoints in the hazard assessment section below.

Hazard Statement and Occupational Control

The Globally Harmonized System of Classification and Labelling of Chemicals (GHS) hazard statements that were harmonized across the European Union (EU) for IPPD are indicated in Table 1. General personal protective equipment (PPE) recommendations are presented in Table 2, below. No occupational exposure limits (OELs) were identified.

Table 1: GHS H Statements for IPPD (CAS #101-72-4) (Pharos 2021)

H Statement	H Statement Details
H302	Harmful if swallowed
H317	May cause an allergic skin reaction
H400	Very toxic to aquatic life
H410	Very toxic to aquatic life with long lasting effects

Table 2: Occupational Exposure Limits and Recommended Personal Protective Equipment for IPPD (CAS #101-72-4)

Personal Protective Equipment (PPE)	Reference	Occupational Exposure Limits (OEL)	Reference
Respiratory: Dust protection mask	ECHA 2021a	None identified	
Hand: Chemical-resistant, impervious gloves			
Eye: Safety glasses with side-shields			

Physicochemical Properties of IPPD

IPPD is a solid at room temperature. Its vapor pressure and boiling point indicate it may be volatile. It has low water solubility, and its log K_{ow} indicates it is not expected to bioaccumulate.

Table 3: Physical and Chemical Properties of IPPD (CAS #101-72-4)

Property	Value	Reference
Molecular formula	$C_{15}H_{18}N_2$	ChemIDplus 2021

Table 3: Physical and Chemical Properties of IPPD (CAS #101-72-4)		
Property	**Value**	**Reference**
SMILES Notation	CC(C)Nc1ccc(Nc2ccccc2)cc1	ChemIDplus 2021
Molecular weight	226.321 g/mol	ChemIDplus 2021
Physical state	Solid	ECHA 2021a
Appearance	Dark-gray to black flakes	ECHA 2021a
Melting point	78.5°C	ECHA 2021a
Boiling point	148-152°C	ECHA 2021a
Vapor pressure	0.00007 hPa @ 20°C	ECHA 2021a
Water solubility	15 mg/L	ECHA 2021a
Dissociation constant	pKa = 6.76 @ 20°C	ECHA 2021a
Density/specific gravity	1.04 g/cm^3 @ 25°C	ECHA 2021a
Partition coefficient	Log K_{ow} = 2.77 @ 25°C	ECHA 2021a

Toxicokinetics

There are limited data available regarding the toxicokinetics of IPPD in both humans and animals.

- *Absorption*
  - In one animal study, the tails of mice were ¾ immersed in 50% IPPD oil for a period of time that was not reported; although it was reported that IPPD did not penetrate unbroken skin, no further details were provided (UNEP 2002).
  - In a poorly-reported human study, the dermal absorption of IPPD was evaluated in a human volunteer. The volunteer immersed one hand in 10 liters of cold water that contained 2 g indissoluble IPPD for 90 minutes. Urine samples were collected in intervals over 7 days. IPPD was detected in the urine 3 hours after the end of exposure and was no longer detected 7 days after exposure. This study shows the potential of dermal absorption of IPPD. IPPD was detected in the urine of workers exposed to IPPD by the inhalation or/ and dermal route (UNEP 2002, ECHA 2021a).
- *Distribution*
  - In a poorly-reported human study, urine was collected twice daily (pre- and post-shift) over a 2-week period from 16 people exposed to IPPD during the curing of rubber; no information was available regarding the route, level, or duration of exposure. The weekly mean levels of IPPD in the urine increased from 19.55 to 83.57 mg/L for pre- to post-shift, respectively. There was also some evidence of accumulation of IPPD in the body with the pre-shift urine levels increasing from 10.8 to 25.8 mg/L over the course of the work week; due to the lack and quality of the information, however, few conclusions could be drawn (UNEP 2002, ECHA 2021a).
- *Metabolism*
  - The major metabolite of IPPD identified in rabbits was a N-glucuronide (ECHA 2021a).
- *Excretion*
  - In the previously described human study in 16 workers, a fast and a slower component of IPPD excretion kinetics with urine were suggested (UNEP 2002, ECHA 2021a).

Hazard Classification Summary

Group I Human Health Effects (Group I Human)

Carcinogenicity (C) Score (H, M, or L): *L*
IPPD was assigned a score of Low for carcinogenicity based on negative experimental data on the surrogate 6PPD, the lack of structural alerts in Toxtree, a reliable prediction from one VEGA model, and negative and in domain results from all FDA RCA cancer models in the Danish QSAR database. ToxServices also attempted to use OncoLogic to evaluate IPPD, but the program is not capable of evaluating its structure. GreenScreen® criteria classify chemicals as a Low hazard for carcinogenicity when adequate negative data are available and they are not GHS classified (CPA 2018b). The confidence in the score is low as it is based on data on a weak surrogate and 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
  - IPPD 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 carcinogen with moderate reliability. An applicability domain (AD) index of 0.657 is calculated (Appendix E), indicating that the prediction is not reliable.
  - ISS Carcinogenicity Model (v.1.0.2) predicts that the compound is a non-carcinogen with moderate reliability. An AD index of 0.805 is calculated (Appendix E), indicating that the prediction is reliable.
  - IRFMN/Antares Carcinogenicity Model (v.1.0.0) predicts that the compound is a carcinogen with weak reliability. An AD index of 0.0 is calculated (Appendix E), indicating that the prediction is not reliable.
  - IRFMN/ISSCAN-CGX Carcinogenicity Model (v.1.0.0) predicts that the compound is a carcinogen with weak reliability. An AD index of 0.0 is calculated (Appendix E), indicating that the prediction is not reliable.
  - IRFMN Oral Classification Model (v1.0.0) predicts that the compound is a carcinogen with weak reliability. An AD index of 0.558 is calculated (Appendix E) indicating that the prediction is not reliable.
  - IRFMN Inhalation Classification Model (v1.0.0) predicts that the compound is a carcinogen with moderate reliability. An AD index of 0.657 is calculated (Appendix E) indicating that the prediction is not reliable.
- 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) predict IPPD 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) predict IPPD to be negative, and all predictions are in domain (Appendix F).
  - The liver specific cancer in rat or mouse model battery predicts the compound to be negative (but out of domain), and none of the three models in the model battery produced in domain predictions (Appendix F).
- UNEP 2005, ECHA 2021b

- *Surrogate: 6PPD (CAS #793-24-8):* A non-GLP-compliant chronic feeding study conducted in a manner similar to OECD Guideline 451 was performed with Sprague-Dawley rats (70/sex/group) provided diets containing 6PPD (as Santoflex 13, 100% active ingredient) at 0, 50, 250, or 1,500 ppm (providing doses of 2.6, 13.5, and 84.8 mg/kg/day for males, and 3.2, 16.5, and 109.5 mg/kg/day for females, respectively) for up to two years. After 12 months, 20 rats/sex/group were sacrificed, and the remaining animals were sacrificed after 24 months. A slight, non-statistically significant increase in the incidence of thyroid follicular cell carcinoma was identified in male rats (the control, low, mid, high dose group incidences were 0/70, 0/69, 2/70, and 3/69, respectively). No such increase was identified in female rats. Reviews in the literature suggest that the increased incidence of this neoplasm may be due to increased liver activity and disruption of thyroid-pituitary signaling and may not be relevant for humans. Therefore, the authors concluded that 6PPD is not likely to be carcinogenic (Klimisch 2, reliable with restrictions).
- *Surrogate: 6PPD (CAS #793-24-8):* A non-GLP-compliant feeding study was performed with Charles River (CD Outbred) rats (50/sex/group) provided diets containing 6PPD (as Santoflex 13, purity not specified) at 0, 100, 300, or 1,000 ppm (contributing doses of 8, 23, and 75 mg/kg/day, respectively) for 24 months. Treatment did not increase the tumor frequency or type of tumors relative to those identified in the control group (Klimisch 2, reliable with restrictions).
- *Surrogate: 6PPD (CAS #793-24-8):* A GLP-compliant *in vitro* cell transformation assay was performed with BALB/3T3 cells exposed to 6PPD (purity not specified) at 0.61-1,000 µg/mL (range finding) and 0.165-0.99 µg/mL (cell transformation assay). Exposure to ≥ 0.488 µg/mL resulted in ≤ 32.3% relative survival. Treatment did not increase the frequency of transformed foci relative to the solvent control, whereas the positive control (methylcholanthrene) produced the expected increase in transformed foci (Klimisch 2, reliable with restrictions).

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

IPPD was assigned a score of Low for mutagenicity/genotoxicity based on a weight of evidence of *in vitro* bacterial reverse mutation assays, mammalian cell mutation assays, an *in vitro* chromosomal aberration assay, and an *in vivo* mouse micronucleus assay. 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 2021a
  - *In vitro*: Negative results for mutagenicity were obtained in a GLP-compliant bacterial reverse mutation assay conducted according to Ames et al. (1975) methods. *Salmonella typhimurium* test strains TA98, TA100, TA1535, and TA1537 were exposed to IPPD (92-99% purity, solvent not reported) at concentrations of 0.2-200 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 bacterial reverse mutation assay (GLP compliance, guideline not reported). *S. typhimurium* test strains TA98, TA100, TA1535, TA1537, and TA1538, as well as *Escherichia coli* strain WP_2 *uvr*A, were exposed

to IPPD (purity not reported) at concentrations of 1-200 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 2, reliable with restriction).
    - *In vitro*: Negative results for mutagenicity were obtained in a non-GLP compliant bacterial reverse mutation assay (guideline not reported). *S. typhimurium* test strains TA98, TA100, TA1535, TA1537, and TA1538, as well as *Saccharomyces cerevisiae* strain D4, were exposed to IPPD (purity not reported) at concentrations of 1-500 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 2, reliable with restriction).
    - *In vitro*: Negative results for mutagenicity were obtained in a non-GLP compliant bacterial reverse mutation assay (guideline not reported). *S. typhimurium* test strains TA98, TA100, TA1535, TA1537, and TA1538, as well as *S. cerevisiae* strain D4, were exposed to IPPD (purity not reported) at concentrations of 0.001-5 µL/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 2, reliable with restriction).
    - *In vitro*: Negative results for mutagenicity were obtained in a GLP-compliant mammalian cell gene mutation assay conducted according to OECD Guideline 476. Chinese hamster ovary (CHO) cells were exposed to IPPD (92-99% purity, solvent not reported) at concentrations of 2-30 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 vivo*: Negative results were obtained in a GLP-compliant micronucleus assay conducted according to OECD Guideline 474. Male Crl:CD (SD) rats (6/dose) were exposed to IPPD (97.6% purity) at doses of 0, 37.5, 75, and 150 mg/kg/day via daily gavage for three consecutive days. There were no increases in micronuclei formation seen at any dose level (Klimisch 1, reliable without restrictions).
- NTP 2018
    - *In vitro:* Negative results were obtained in a bacterial reverse mutation assay; GLP compliance and test guidelines not reported. *S. typhimurium* test strains TA98, TA100, TA1535, TA1537 were exposed to IPPD (purity not reported) at concentrations up to 1,000 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.
- U.S. EPA 2011
    - *In vitro:* Negative results were obtained in an *in vitro* mammalian cell gene forward mutation assay in mouse lymphoma L5178Y cells exposed to the test article (97% purity) at 0.156-2.500 µg/mL and 0.625-10.000 µg/mL with metabolic activation. The highest tested concentrations were cytotoxic. Positive and negative controls were valid.
- NTP 1986a
    - *In vitro*: Positive results for clastogenicity were obtained in a chromosomal aberration assay; GLP compliance and test guidelines were not reported. CHO cells were exposed to IPPD (purity not reported) at concentrations of 1.6-50 µg/mL both in the presence and absence of metabolic activation. There was an increase in chromosomal aberrations seen both in the presence and absence of metabolic activation.
- NTP 1986b
    - *In vitro*: Positive results for genotoxicity were obtained in a sister chromatid exchange (SCE) assay; GLP compliance and test guidelines were not reported. CHO cells were exposed to IPPD (purity not reported) at concentrations of 0.05-50 µg/mL both in the

presence and absence of metabolic activation. There was an increase in SCEs seen both in the presence and absence of metabolic activation.

- Based on the weight of evidence, IPPD is not expected to be genotoxic. Although IPPD was clastogenic in an *in vitro* chromosomal aberration assay, an *in vivo* mouse micronucleus assay was negative for clastogenicity. As the *in vivo* study was negative, IPPD is not expected to be mutagenic or genotoxic.

Reproductive Toxicity (R) Score (H, M, or L): *H*

IPPD was assigned a score of High for reproductive toxicity based on ToxServices classifying it as a GHS Category 1B reproductive toxicant. GreenScreen® criteria classify chemicals as a High hazard for reproductive toxicity when they are classified as GHS Category 1B reproductive toxicants (CPA 2018b). The confidence in the score is low as it is based on data on a weak surrogate.

- 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
  - *Surrogate: 6PPD (CAS #793-24-8):* A GLP-compliant, OECD Guideline 443 extended one-generation reproductive toxicity study was performed with Sprague-Dawley rats (25-30/sex/group) administered gavage doses of 6PPD (95.7% purity) in corn oil at 7, 20, or 60 mg/kg/day. F0 males were dosed for 70 consecutive days prior to mating and through mating for a minimum of 10 weeks. F0 females were dosed for 70 consecutive days prior to mating, during mating, gestation, and lactation, and until weaning of the F1 pups. The parental animals were evaluated for clinical signs of toxicity, estrous cyclicity, sperm parameters (numbers, production rate, motility, progressive motility, and morphology), gross pathology, histopathology, and reproductive performance. Treatment did not adversely affect sperm parameters or male reproductive performance. Two and five females in the mid and high dose groups, respectively, were found dead or euthanized *in extremis* on gestation day 21 through lactation day 2. The authors attributed to deaths and moribund condition to prolonged labor and/or dystocia (difficult birth). Therefore, the authors identified a female reproductive toxicity NOAEL of 7 mg/kg/day based on the dystocia identified at 20 and 60 mg/kg/day (Klimisch Score 1, reliable without restriction).
  - *Surrogate: 6PPD (CAS #793-24-8):* A GLP-compliant reproduction / developmental toxicity screening test conducted in a manner similar to OECD Guideline 421 was used as the dose range-finding study for the OECD Guideline 443 study discussed above. Sprague-Dawley rats (15/sex/group) were administered gavage does of 6PPD (96.9% purity) in corn oil at 0, 50, 75, or 100 mg/kg/day. Males were dosed for at least 14 days prior to mating and through mating for 28 days. Females were dosed for at least 14 days prior to mating and through mating, gestation, and lactation. Over the course of the study, one female each in the low and mid dose groups were found dead and one and three females each in the low and high dose groups were euthanized *in extremis*. No treatment-related effects were identified for body weight, food consumption, thyroid hormone levels [triiodothyronine (T3), thyroxine (T4), and thyroid stimulating hormone (TSH)], or histopathological findings. Treatment did not adversely affect mating, fertility, or copulation/conception indices or the mean estrous cycle lengths, but mean gestation lengths in the treatment group were greater than the concurrent control group (statistical significance not provided). Dystocia was identified for one, one, and five females in the low, mid, and high dose groups, respectively, including for the three high dose females sacrificed *in extremis*. As this was a dose range-

finding study, the authors did not identify a reproductive toxicity NOAEL (Klimisch Score 1, reliable with restrictions).

- UNEP 2005, ECHA 2021b
    - *Surrogate: 6PPD (CAS #793-24-8):* A GLP-compliant, OECD Guideline 421 reproduction/developmental toxicity screening test was performed with Crj: CD(SD) rats (12/sex/group) administered gavage doses of 6PPD (99.4% purity) in corn oil at 0, 6, 25, or 100 mg/kg/day. Males were dosed for 48 days, and females were dosed for 14 days prior to mating until lactation/postnatal day 3. Treatment did not affect body weight gain, and food consumption rates increased intermittently in high dose males and in females in all dose groups during lactation only. Treatment did not adversely affect the copulation or fertility index or estrus cyclicity, but the gestation length was statistically significantly greater in the high dose group (22.7 days) compared to the concurrent control group (22.2 days). The authors identified a reproductive toxicity NOAEL of 100 mg/kg/day based on the lack of adverse effects on fertility (Klimisch Score 1, reliable without restrictions).
    - *Surrogate: 6PPD (CAS #793-24-8):* A non-GLP-compliant three-generation reproduction toxicity test was performed with Charles River CD rats (8 males and 15 females per group per generation) provided diets 6PPD (as Santoflex 13) at 0, 100, 300, or 1,000 ppm (contributing doses of 0, 8, 23, and 75 mg/kg/day, respectively). The F0 males and females were treated for 11 weeks prior to mating, and the exposure continued through mating, gestation, and lactation for two successive litters. The mating and fertility indices, incidence of parturition, mean number of live and dead pups at birth, and number of pups weaned were comparable between the control and treatment groups. The fertility indices for mid dose F1b males and F2a females were lower than controls but the authors attributed these findings to their poor health (decreased body weights and decreased survival). The authors concluded that treatment did not adversely affect fertility in this study and identified a reproductive toxicity NOAEL of 1,000 ppm (75 mg/kg/day) the highest dose tested (Klimisch Score 2, reliable with restrictions).
- In summary, while surrogate 6PPD did not adversely affect fertility, several studies identified increased gestation length and/or an increased incidence of dystocia with treatment. As multiple studies identified dystocia with treatment and due to the potential adverse impacts on the health of the mother and offspring, the REACH dossier authors for 6PPD classified it as a GHS Category 1B reproductive toxicant. ToxServices agrees with this classification and assigned the hazard score for this endpoint based on this classification.

Developmental Toxicity incl. Developmental Neurotoxicity (D) Score (H, M, or L): M

IPPD was assigned a score of Moderate for developmental toxicity based on the authoritative listing for Pregnancy Group C and retarded ossification seen in the absence of apparent maternal toxicity in an OECD 414 study in rats. This effect warrants a classification to GHS Category 2. GreenScreen® criteria classify chemicals as a Moderate hazard for developmental toxicity when associated with Pregnancy Group C (MAK) and when classified to GHS Category 2 (CPA 2018b). The confidence in the score is high as it is based on an authoritative listing and based on reliable experimental data.

- Authoritative and Screening Lists
    - *Authoritative:* MAK: Pregnancy Risk Group C.
    - *Screening:* Not present on any screening lists for this endpoint.
- ECHA 2021a, U.S. EPA 2011
    - *Oral*: In a GLP-compliant developmental toxicity study conducted according to OECD Guideline 414, pregnant Sprague-Dawley rats (24/dose) received doses of 12.5, 62.5, and 125 mg/kg IPPD (97.2% purity) in polyethylene glycol (PEG) by gavage on gestation days

(GD) 6-15. Parameters evaluated include clinical observations, body weight, food consumption, maternal examinations, uterine/implantation data, litter data, and fetal examinations. There were no treatment-related effects seen in the maternal animals according to the ECHA record, however, U.S. EPA reported slight maternal toxicity at the high dose, including reduced food intake, pre-dosing salivation and soft, dark feces. In the fetuses, there were statistically significant increases in the retardation of ossification seen in high-dose and mid-dose animals. REACH dossier authors identified a fetotoxic NOAEL of 62.5 mg/kg/day (Klimisch 1, reliable without restriction). U.S. EPA identified a NOAEL of 62.5 mg/kg/day and LOAEL of 125 mg/kg/day for maternal toxicity based on reduced food intake, pre-dosing salivation and soft, dark feces, and a NOAEL of 12.5 mg/kg/day and LOAEL of 62.5 mg/kg/day for developmental toxicity based on incomplete bone ossification.

- U.S. EPA 2011
    - In a prenatal developmental toxicity study, pregnant Sprague-Dawley rats (24/dose) were exposed to IPPD by gavage in polyethylene glycol 400 at doses of 0 10, 50 or 100 mg/kg/day on GDs 6-15. Maternal animals exhibited post-dosing salivation and lethargy and a slight reduction in food consumption between GDs 6 and 9. There were no unscheduled mortality, and no treatment related effects on the developing fetus. U.S. EPA identified a NOAEL of 50 mg/kg/day and LOAEL of 100 mg/kg/day for maternal toxicity based on post-dosing salivation and lethargy, and a NOAEL of 100 mg/kg/day for developmental toxicity, which was the highest dose tested.

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

IPPD was assigned a score of Moderate for endocrine activity based on altered female pubertal development in rats for the surrogate 6PPD. In addition, *in silico* modeling of IPPD and/or its metabolites may be endocrine receptor modulators. However, there does not appear to be endocrine-mediated carcinogenicity, reproductive or developmental toxicity, or systemic toxicity that warrant raising the final score to High. While the score for reproductive toxicity endpoint is High, there is no evidence that the critical reproductive effect, dystocia, is mediated via endocrine disruption for the surrogate 6PPD. GreenScreen® criteria classify chemicals as a Moderate hazard for endocrine activity when there is evidence of endocrine activity and there are no linked health effects that warrant raising the score (CPA 2018b). The confidence in the score is low as it is based on modeled data and experimental data on a weak surrogate.

- 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
    - N-Isopropyl-N'-phenyl-p-phenylenediamine was active in 4/28 estrogen receptor (ER) assays, 6/16 androgen receptor (AR) assays, 2/2 steroidogenesis assays, and 3/9 thyroid receptor assays performed as part of the U.S. EPA's Endocrine Disruptor Screening Program (EDSP) in the 21st Century.
- DTU 2021 (only results that are in domain are described below)
    - IPPD has no structural alerts for estrogen receptor binding. However, its predicted metabolites are expected to be strong binders of estrogen receptors (Appendix F).
    - IPPD is predicted to be negative for estrogen receptor α binding (full and balanced training set, human *in vitro*) by the respective model batteries (Appendix F).
    - IPPD is predicted to be negative for estrogen receptor α activation by SciQSAR model (human *in vitro* and CERAPP data *in vitro*)

  - IPPD is predicted to be negative for androgen receptor activation (human *in vitro*) with CoMPARA data by the Leadscope model (Appendix F).
  - Benzoyl peroxide is predicted to be positive for thyroperoxidase (TPO) inhibition by the QSAR 1 and QSAR 2 (rat *in vitro*) models in Leadscope (Appendix F).
- ECHA 2021b
  - *Surrogate: 6PPD (CAS #793-24-8):* A GLP-compliant U.S. EPA OPPTS 890.1450 pubertal study was performed with juvenile female Sprague-Dawley rats (15/group) administered gavage doses of N-(1,3-dimethylbutyl)-N'-phenyl-p-phenylenediamine (6PPD) (91% purity) in corn oil at 0, 250, or 500 mg/kg/day for 21 days (postnatal days 22 to 42 or 43). The animals were evaluated for clinical signs of toxicity, body weights, vaginal opening, estrous cyclicity, thyroid hormone levels (T4 and TSH), and histopathology (kidney, thyroid, ovary, and uterus). Treatment-related clinical signs of toxicity included salivation prior to dosing, clear material around the mouth approximately two hours after dosing, and yellow material around the urogenital region (high dose only). Decreased mean body weight gains were identified during the first two (low dose) or three (high dose) days of dosing, resulting in mean body weights that were 8.73% and 14.83% less than the control group for the low and high dose groups, respectively, during the treatment period. Vaginal opening was achieved at an earlier date for the high dose group (33.2 days) than the concurrent control group (35.2 days), and lower body weights were noted for females in both dose groups at the time of vaginal opening. Treatment increased the age at first estrus for the high dose group (39.2 days) compared to the concurrent controls (36.3 days), and a lower number of animals were cycling by the end of study period relative to the control group (estrous cycle lengths could not be evaluated). Treatment in both dose groups increased serum TSH and cholesterol levels and decreased serum T4, AST< and triglyceride levels. High dose females also exhibited increased total bilirubin and gamma-glutamyl transferase (GGT) levels. High dose females exhibited decreased ovary weights, and mid and high dose females exhibited decreased uterine (blotted and unblotted) weights and increased liver, kidney, and thyroid weights. Treatment-related histopathological alterations included lower colloid area and increased follicular cell height in thyroid glands of mid and high dose females, and vacuolation of the liver, absence of corpora lutea with increased tertiary follicles in the ovaries (i.e., non-cycling), and immature uterus of high dose females. The authors postulated that the increased liver weights, alterations to thyroid gland histopathology and T4 and TSH levels were secondary to hepatomegaly, but the liver histopathology was not evaluated. The authors concluded that oral dosing with N-(1,3-dimethylbutyl)-N'-phenyl-p-phenylenediamine (6PPD) produced evidence of endocrine-mediated effects on pubertal development and thyroid function in juvenal female rats (Klimisch Score 1, reliable without restriction).
  - *Surrogate: 6PPD (CAS #793-24-8):* A GLP-compliant OPPTS 890.1500 Endocrine Disruption test was performed with juvenile male Sprague-Dawley rats (15/group) administered gavage doses of N-(1,3-dimethylbutyl)-N'-phenyl-p-phenylenediamine (6PPD) (91.0% purity) in corn oil at 250 or 500 mg/kg/day for 30 days (postnatal days 23 to 53 or 54). The males were evaluated for clinical signs of toxicity and body weights, balanopreputial separation (beginning on postnatal day 30), serum T4, TSH, and testosterone levels, and histopathology of the kidney, thyroid, testis, and epididymis. One high dose male was euthanized *in extremis* on postnatal day 25 due to severe body weight loss. Treatment-related clinical signs of toxicity included salivation prior to dosing and red and/or clear material around the mouth approximately two hours after dosing. Decreased body weight gains were noted in both dose groups, with mean final body weights for the low and

high dose group animals up to 8.69% and 22.33% lower than the control group, respectively. High dose males exhibited a delayed mean age of balanopreputial separation attainment, and lower body weights on the day of attainment of balanopreputial separation was noted for both dose groups. The authors attributed these findings to the decreased body weights for these groups. High dose males exhibited higher GGT and ALT activities, and mid and high dose males exhibited decreased T4 and testosterone and increased TSH levels. Treatment-related organ weight changes included increased liver weights and decreased testes, epididymides, prostate, and seminal vesicle/coagulating gland weights in males of both dose groups. Treatment-related histopathological changes were limited to lower colloid area and increased follicular cell height in the thyroid gland in both dose groups. The authors considered the histopathological changes in the thyroid gland, increased liver weights, TSH, ALT, and GGT levels, and decreased T4 to be secondary to hepatomegaly, although the histopathology of the liver was not evaluated. Additionally, the authors attributed the decreased testosterone levels and male reproductive organ weights to be secondary to systemic stress (decreased body weights). "Therefore, there was no clear evidence of any direct test-substance-related endocrine effects." (Klimisch Score 1, reliable without restriction)

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): M

IPPD was assigned a score of Moderate for acute toxicity based on oral LD_{50} values of 491-900 mg/kg and being associated with EU H Statement H302. GreenScreen® criteria classify chemicals as a Moderate hazard for acute toxicity when associated with H Statement H302 and oral LD_{50} values are >300-2,000 mg/kg (CPA 2018b). The confidence in the score is high as it is based on an authoritative listing and based on reliable experimental data.

- Authoritative and Screening Lists
  - *Authoritative:* EU – GHS: H302 – Harmful if swallowed – Acute toxicity (oral) – Category 4
  - *Screening:*
    - GHS – Australia: H302 – Harmful if swallowed – Acute toxicity (oral) – Category 4
    - GHS – Japan: H302 – Harmful if swallowed – Acute toxicity (oral) – Category 4
    - GHS – Korea: H302 – Harmful if swallowed – Acute toxicity (oral) – Category 4
    - GHS – New Zealand: 6.1D (oral) – Acutely toxic
- ECHA 2021a
  - *Oral*: LD_{50} = 522 mg/kg bw, GLP-compliant, OECD Guideline 401, Crj: CD(SD) rat, male and female (Klimisch 2, reliable with restrictions).
  - *Oral*: LD_{50} = 900 mg/kg bw, non-GLP compliant, acute oral toxicity study, Sprague-Dawley rat, male and female (Klimisch 2, reliable with restrictions).
  - *Oral*: LD_{50} = 491 mg/kg bw (male), 422 mg/kg bw (female), non-GLP compliant, acute oral toxicity study, Sprague-Dawley rat, male and female (Klimisch 2, reliable with restrictions).
  - *Dermal*: LD_{50} >7,940 mg/kg, non-GLP compliant, acute dermal toxicity study, New Zealand white rabbit, male and female (Klimisch 2, reliable with restrictions).

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

IPPD was assigned a score of Data Gap for systemic toxicity (single dose) based on insufficient data.

- Authoritative and Screening Lists
  - *Authoritative:* Not present on any authoritative lists for this endpoint.
  - *Screening:* Not present on any authoritative lists for this endpoint.
- ECHA 2021a
  - *Oral:* A GLP-compliant study was conducted according to OECD Guideline 401 where male and female Crj: CD(SD) rats (5/sex/dose) received IPPD (99.5% purity) in 0.5% sodium carboxymethylcelluose via gavage at 0, 269, 350, 455, 592, 769, and 1,000 mg/kg. The study found that animals died 2-4 days after male animals received 269 mg/kg, as well as 592 mg/kg and above for females. Clinical signs included brownish urine, a crouching position, eyelid closure, a decrease in fecal volume, pale skin, and abdominal distension; additionally, some animals that subsequently died also showed an adoption of a prone position, decreased respiration, lacrimation, and hypothermia. There was a decrease in body weight in dosed groups, with a return to normal starting at day 8. Gross pathology of deceased animals showed an enlargement of the liver, enlargement and pale coloration of the kidney, pleural effusion, ascites, edematous lung, shrinking and pale coloration of the spleen, detachment of red-colored areas in the forestomach mucosa, thickening and pale coloration in the mucosa of glandular stomach, and yellowish-colored change of the subcutis. Histopathology of deceased animals showed necrosis or degeneration of centrilobular hepatocytes and hypertrophy of hepatocytes, necrosis or degeneration in the proximal tubular epithelium of the kidneys, alveolar edema in the lung, and hemorrhage and edema in the submucosa of the forestomach and in the mucosa of the glandular stomach. The authors established an LD_{50} of 522 mg/kg, and classified IPPD as GHS Category 4 (Klimisch 2, reliable with restrictions).
  - *Oral:* A non-GLP compliant study was conducted according to no specified guidelines where male and female Sprague-Dawley rats (2-3 per sex/dose) received IPPD (purity not reported) in corn oil via gavage at 0, 631, 794, 1,000, and 1,260 mg/kg. At the top two doses, 4/5 and 5/5 animals died within 2 days of dosing, respectively. Clinical signs included reduced appetite and activity (3-5 days in survivors), increasing weakness, collapse, and finally death. Gross necropsy of deceased animals showed lung hyperemia, slight liver discoloration, and acute gastrointestinal inflammation. The authors established an LD_{50} of 900 mg/kg (Klimisch 2, reliable with restrictions).
  - *Dermal*: A non-GLP compliant study conducted according to no specified guidelines found no deaths when male and female New Zealand white rabbits (1-2/dose) were exposed to IPPD (purity not reported) in corn oil at 5,010 and 7,940 mg/kg under semi-occlusive conditions. While there was some reduced appetite and activity seen for 3 to 5 days, gross necropsy did not show any effects to the viscera. The authors established an LD_{50} >7,940 mg/kg (Klimisch 2, reliable with restrictions).
- In summary, most studies did not provide sufficient data to determine what dose level(s) the gross pathological alterations were identified at and what effects were observed in survivors. Therefore, ToxServices assigned a Data Gap for this endpoint.

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

IPPD was assigned a score of Moderate for systemic toxicity (repeated dose) based on a LOAEL of 13.5 mg/kg/day from a 90-day feeding study in rats. GreenScreen® criteria classify chemicals as a Moderate

hazard for systemic toxicity (repeated dose) when oral LOAEL values are >10-100 mg/kg/day (CPA 2018b). The confidence in the score is high as it is based on reliable experimental data. It should be noted that the LOAEL was the lowest dose in the study. Therefore, there is a possibility that IPPD may be classified to GHS Category 1 if lower doses were tested.

- Authoritative and Screening Lists
    - *Authoritative:* Not present on any authoritative lists for this endpoint.
    - *Screening:* Not present on any authoritative lists for this endpoint.
- ECHA 2021a
    - *Oral*: A GLP-compliant subacute feeding study was conducted according to an unspecified guideline. Male and female Sprague-Dawley rats (5/sex/dose) were administered diets containing 0, 500, 1,000, 1,750, and 2,500 ppm (equivalent to 0, 43.8, 92.6, 149.5, and 215.1 mg/kg/day in males and 0, 48.1, 92.8, 160.3, and 231.5 mg/kg/day in females, respectively, according to the ECHA record) for 4 weeks. Parameters evaluated include mortality, clinical observations, body weights, feed consumption, hematology, clinical chemistry, organ weights, and pathology. The mean body weights and body weight gains, as well as mean food consumption values, were reduced in the top two dose groups of male animals. Hematology findings included reduced hemoglobin concentration and hematocrit values in the top three dose groups in both males and females, as well as elevated platelet counts and total leukocyte counts in males at the top three doses; the total leukocyte counts were associated with an increase in the mean absolute segmented neutrophil counts. The treatment-related effects on clinical chemistry include increased albumin, globulin, and calcium in the top three dose groups in both males and females; slightly reduced glucose in males at the top two doses; elevated total protein in females at the top three doses; and elevated total protein in males at all dose levels. Based on the effects seen, a LOAEL of 500 ppm (equivalent to 43.8 mg/kg/day) in males and a NOAEL of 500 ppm (equivalent to 48.1 mg/kg/day) in females were established.
    - *Oral*: A GLP-compliant 90-day study was conducted according to OECD Guideline 408. Male and female Sprague-Dawley rats (10/sex/dose) were administered diets containing, 0, 180, 360, and 720 ppm (equivalent to 0, 13.5, 26.5, and 54.0 mg/kg/day in males, and 0, 15.6, 30.0, and 59.0 mg/kg/day in females, respectively, according to the ECHA record) daily for 90 days. Parameters evaluated include mortality, physical observations, ophthalmology, body weight, hematology, clinical chemistry, organ weights, pathology, and histology. In high-dose males, there was soft stool seen in high-dose males, as well as a slight decrease in mean body weight gain that was determined to be treatment-related. There were several statistically significant treatment-related effects on hematological parameters: a significant reduction in hemoglobin concentration and hematocrit values in mid- and high-dose males and females; a significant reduction in hemoglobin concentration in high-dose females at 13 weeks; elevated platelet counts in mid- and high-dose males at 6 weeks; and reduced mean erythrocyte counts in mid- and high-dose females at 6 weeks and high-dose females at 13 weeks. The treatment-related effects on clinical chemistry include: a significant increase in total protein in mid- and high-dose males and females at week 6, and males at study termination; a significant increase in albumin in all males, as well as mid- and high-dose females at week 6; a significant increase in calcium levels in all animals at week 6, as well as in low- and mid-dose males at the termination of the study; and a significant decrease in chloride levels in mid- and high-dose males at the study termination, in low- and high-dose females at week 6, and in all females at the termination of the study. There was a significant increase in mean liver weights, liver to body weight ratio, and liver to brain weight ratio in mid- and high-dose males, as well as all females. As there were treatment

related effects seen at all dose levels, a LOAEL of 180 ppm was established (equivalent to 13.5 mg/kg/day in males and 15.6 mg/kg/day in females (Klimisch 1, reliable without restriction).

Neurotoxicity (single dose, N-single) (Group II) Score (vH, H, M, or L): *M*
IPPD was assigned a score of Moderate for neurotoxicity (single dose) based on ToxServices classifying it as a Category 3 specific target organ toxicant following single exposures for narcotic effects. GreenScreen® criteria classify chemicals as a Moderate hazard for neurotoxicity (single dose) when they are classified as GHS Category 3 specific target organ toxicant following single exposures for narcotic effects (CPA 2018b). The confidence in the score is low as it is not clear if the observed effects were specific neurotoxicity or just a manifestation of general toxicity and weakness after receiving a large dose of a chemical.

- 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 2021a
  - *Oral:* A GLP-compliant study was conducted according to OECD Guideline 401 where male and female Crj: CD(SD) rats (5/sex/dose) received IPPD (99.5% purity) in 0.5% sodium carboxymethylcelluose via gavage at 0, 269, 350, 455, 592, 769, and 1,000 mg/kg. The study found that animals died 2-4 days after male animals received 269 mg/kg, as well as 592 mg/kg and above for females. Clinical signs included brownish urine, a crouching position, eyelid closure, a decrease in fecal volume, pale skin, and abdominal distension; additionally, some animals that subsequently died also showed an adoption of a prone position, decreased respiration, lacrimation, and hypothermia. There was a decrease in body weight in dosed groups, with a return to normal starting at day 8 (Klimisch 2, reliable with restrictions).
  - *Oral:* A non-GLP compliant study was conducted according to no specified guidelines where male and female Sprague-Dawley rats (2-3 per sex/dose) received IPPD (purity not reported) in corn oil via gavage at 0, 631, 794, 1,000, and 1,260 mg/kg. At the top two doses, 4/5 and 5/5 animals died within 2 days of dosing, respectively. Clinical signs included reduced appetite and activity (3-5 days in survivors), increasing weakness, collapse, and finally death (Klimisch 2, reliable with restrictions).
  - *Dermal*: A non-GLP compliant study conducted according to no specified guidelines found no deaths when male and female New Zealand white rabbits (1-2/dose) were exposed to IPPD (purity not reported) in corn oil at 5,010 and 7,940 mg/kg under semi-occlusive conditions. While there was some reduced appetite and activity seen for 3 to 5 days, gross necropsy did not show any effects to the viscera (Klimisch 2, reliable with restrictions).

Neurotoxicity (repeated dose, N-repeated) (Group II*) Score (H, M, or L): DG
IPPD was assigned a score of Data Gap for neurotoxicity (repeated dose) based on a lack of neurotoxicity 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 data were identified.

Skin Sensitization (SnS) (Group II*) Score (H, M, or L): H
IPPD was assigned a score of High for skin sensitization based on the authoritative listings for MAK Sh and EU GHS H-Statement H317, as well as positive results in a guinea pig maximization test and local

lymph node assays in mice leading to GHS Category 1A classification. GreenScreen® criteria classify chemicals as a High hazard for skin sensitization when classified as Category 1A (CPA 2018b). The confidence in the score is high as it is based on two authoritative lists and is based on reliable experimental data.

- Authoritative and Screening Lists
    - *Authoritative:*
        - MAK: Sensitizing Substance Sh – Danger of skin sensitization
        - EU – GHS: H317 – May cause an allergic skin reaction (skin sensitization – Category 1)
    - *Screening:*
        - Japan – GHS: H317 – May cause an allergic skin reaction (skin sensitization – Category 1)
        - Korea – GHS: H317 – May cause an allergic skin reaction (skin sensitization – Category 1)
        - Australia – GHS: H317 – May cause an allergic skin reaction (skin sensitization – Category 1)
        - New Zealand – GHS: 6.5B (contact) – Contact sensitizers (Category 1)
- ECHA 2021a
    - In a guinea pig maximization test conducted according to Magnusson & Kligman (1969) (GLP compliance not specified), IPPD (purity not reported) was applied to female Hartley guinea pigs (20/dose) at 1% in Vaseline under intradermal and epicutaneous conditions. The first induction was 0.5% intracutaneously, the second induction was 1% epicutaneously, and the animals were challenged at 0.05% and 0.5% epicutaneously. IPPD was categorized as a strong sensitizer, with 90% of animals having positive reactions. The authors classified IPPD as a GHS Category 1 sensitizer (Klimisch 2, reliable with restrictions).
        - Based on 90% of the animals responding following an intradermal dose of 0.5%, IPPD warrants classification as a GHS Category 1A skin sensitizer. GHS criteria define Category 1A skin sensitizers as chemicals that produce positive reactions ≥ 60% animals at > 0.1% to ≤ 1% intradermal doses (UN 2021).
    - In a mouse local lymph node assay (LLNA) according to Yamano et al. (2003) (GLP compliance not specified), IPPD (purity not reported) was applied to female Balb/c mice (4/dose) at concentrations of 0, 0.01, 0.03, 0.1, and 0.3% (vehicle not reported). IPPD was found to be a skin sensitizer under the conditions of the study, with stimulation indices (SIs) of 1, 1.3, 1.68, 3.23, and 3.99. Therefore, the authors classified IPPD as a GHS Category 1 sensitizer (Klimisch 2, reliable with restrictions).
        - While the EC3 was not reported, the threshold SI of 3 was exceeded at the concentration of 0.1%, indicating that the EC3 would be less than 0.1%. This is less than the GHS guidance value of 2% for Category 1A classification (UN 2021).
    - In a mouse LLNA conducted according to a non-specified guideline (GLP compliance not specified), IPPD (purity not reported) was applied to female Balb/c mice (3/dose) at concentrations of 0, 0.1, 0.5, 1, and 2% in acetone/olive oil (4:1 v/v). IPPD was found to be a skin sensitizer under the conditions of the study, with SIs of 1.5, 3.85, 2.39, and 1.42. Therefore, the authors classified IPPD as a GHS Category 1 sensitizer (Klimisch 2, reliable with restrictions).
        - ToxServices could not determine an EC3 for this study as no dose response was observed.
- UNEP 2002

- IPPD is well known as a severe allergen, and its use is usually avoided in non-industrial applications.

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

IPPD was assigned a score of Moderate for respiratory sensitization based on positive skin sensitization results and the presence of a structural alert for respiratory sensitization. GreenScreen® criteria classify chemicals as a Moderate hazard for respiratory sensitization when there is low to moderate frequency of concern (CPA 2018b). Confidence in the score is reduced as there are no data to subclassify the compound to Category 1A and 1B, which translate to Moderate and High scores, respectively.

- 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 2021
  - IPPD contains a structural alert for respiratory sensitization: Pro-Michael Addition (Appendix G)
- No data were identified for the target compound for this endpoint. Therefore, ToxServices attempted to evaluate the respiratory sensitization potential of IPPD according to ECHA's guideline (ECHA 2017), which 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). IPPD contains a structural alert for respiratory sensitization and is a skin sensitizer based on positive experimental and human data. According to the ECHA guidance, this warrants classification as a GHS Category 1 respiratory sensitizer. However, ECHA did not provide guidance on subcategorization to GHS Category 1A (high potency) and 1B (low potency). Due to the lack of specific respiratory sensitization data, ToxServices classified it to GHS Category 1B with low confidence.

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

IPPD 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 studies tested under more aggressive conditions (i.e., occlusion and 24 hours) than required by guidelines (i.e., semi-occlusive, 4 hours).

- 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 2021a
  - In a non-GLP compliant *in vivo* skin irritation study (guideline not reported), six New Zealand white rabbits (sex not reported) were administered IPPD (purity not reported) at an undisclosed dose on abraded and intact skin for 24 hours under semiocclusive conditions. Skin was observed at 24 hours, 72 hours, and 7 days following treatment. An overall irritation sore of 0.0/8.0 was established. The study authors concluded that IPPD was not irritating to the skin (Klimisch 2, reliable with restrictions).
- U.S. EPA 2011
  - In an *in vivo* skin irritation study (GLP and guideline not reported), six New Zealand white rabbits (sex not reported) were administered IPPD (purity 97%) at 0.5 mL on abraded and

intact skin for 24 hours under occlusive conditions. Skin was scored at 24 hours, 48 hours, 72 hours, and 7 days following treatment. The scores for all animals were 0. The study authors concluded that IPPD was not irritating to the skin.

Eye Irritation/Corrosivity (IrE) (Group II) Score (vH, H, M, or L): *M*
IPPD was assigned a score of Moderate for eye irritation/corrosivity based on slight eye irritation seen in rabbits. GreenScreen® criteria classify chemicals as a Moderate hazard for eye irritation/corrosivity when they are classified to GHS Category 2B (mild) (CPA 2018b). The confidence in the score is low as scores for individual animals for individual sub-endpoints were not reported for a definitive GHS classification.

- Authoritative and Screening Lists
  - *Authoritative:* Not present on any authoritative lists for this endpoint.
  - *Screening:* Japan – GHS: H320 – Causes eye irritation (Category 2B).
    - Based on conjunctival erythema observed after 24 hours in all tested animals that was reversible within 48 hours in the SIDS dossier (NITE 2017).
- ECHA 2021a, U.S. EPA 2011
  - In a non-GLP compliant *in vivo* acute eye irritation study (guideline not reported), the eyes of six New Zealand white rabbits (sex not reported) were instilled with neat IPPD (purity not reported) for 24 hours. Eyes were observed at 24 hours, 48 hours, 72 hours, and 7 days following treatment. There was slight erythema and discharge seen that was fully reversed within 48 hours. An overall irritation score of 1.3/110 was established (Klimisch 2, reliable with restrictions).

Ecotoxicity (Ecotox)

Acute Aquatic Toxicity (AA) Score (vH, H, M, or L): vH
IPPD was assigned a score of Very High for acute aquatic toxicity based on being listed with EU GHS H-Statement H-400 and the most conservative L/EC_{50} values of 0.34-0.98 mg/L. GreenScreen® criteria classify chemicals as a Very High hazard for acute aquatic toxicity when listed on EU GHS with H-Statement H400 and L/EC_{50} values <1 mg/L (CPA 2018b). The confidence in the score is high as it is listed on an authoritative list and is based on reliable experimental data.

- Authoritative and Screening Lists
  - *Authoritative:* EU – GHS: H400 – Very toxic to aquatic life (Hazardous to the aquatic environment (acute) – Category 1)
  - *Screening:*
    - Korea – GHS: H400 – Very toxic to aquatic life (Hazardous to the aquatic environment (acute) – Category 1)
    - New Zealand – GHS: 9.1A (algal) – Very ecotoxic in the aquatic environment
    - New Zealand – GHS: 9.1A (fish) – Very ecotoxic in the aquatic environment
- ECHA 2021a
  - 96-hour LC_{50} (*Pimephales promelas*) = 0.41 mg/L (GLP-compliant, OECD Guideline 204) (Klimisch 1, reliable without restriction)
  - 48-hour mortality EC_{50} (*Daphnia magna*) = 0.98 mg/L (GLP-compliant, EU Method C.2) (Klimisch 1, reliable without restriction)
  - 48-hour EC_{50} (nominal) (*D. magna*) =1.1 mg/L (GLP compliance not specified, EPA-660/3-75-009) (Klimisch 2, reliable with restriction)
  - 48-hour mortality EC_{15} (*Paratanytarsus parthenogenetica*) = 10 mg/L (GLP-compliant, EPA-660/3-75-000) (Klimisch 2, reliable with restriction)

  - 72-hour growth rate EbC_{50} (*Desmodesmus subspicatus*) = 7.73 mg/L (GLP-compliant, EU Method C.3) (Klimisch 1, reliable without restriction)
  - 72-hour growth rate ErC_{50} (*D. subspicatus*) = 26.5 mg/L (GLP-compliant, EU Method C.3) (Klimisch 1, reliable without restriction)
- UNEP 2002
  - 96-hour LC_{50} (*Lepomis macrochirus*) = 0.43 mg/L (GLP compliance and test guidelines not reported) (Klimisch score not reported)
  - 96-hour LC_{50} (*Oncorhynchus mykiss*) = 0.34 mg/L (GLP compliance and test guidelines not reported) (Klimisch score not reported)

Chronic Aquatic Toxicity (CA) Score (vH, H, M, or L): *vH*
IPPD was assigned a score of Very High for chronic aquatic toxicity based on a measured chronic aquatic toxicity value of 0.004 mg/L for fish for the surrogate 6PPD. GreenScreen® criteria classify chemicals as a Very High hazard for chronic aquatic toxicity when chronic aquatic toxicity data are ≤ 0.1 mg/L (CPA 2018b). The confidence in the score is low as it is based on data on a weak surrogate.

- Authoritative and Screening Lists
  - *Authoritative:* Not present on any screening lists for this endpoint.
  - *Screening:* Not present on any screening lists for this endpoint.
- ECHA 2021a
  - 72-hour growth rate NOECb (*D. subspicatus*) = 2 mg/L (GLP-compliant, EU Method C.3) (Klimisch 1, reliable without restriction)
  - 72-hour growth rate NOECr (*D. subspicatus*) = 4 mg/L (GLP-compliant, EU Method C.3) (Klimisch 1, reliable without restriction)
- U.S. EPA 2017a
  - IPPD belongs to the neutral organics ECOSAR chemical class. The most conservative predicted chronic values (ChVs) are 4.02 mg/L in fish, 2.74 mg/L in daphnia, and 7.07 mg/L in green algae (Appendix H).
- ECHA 2021b
  - <u>*Surrogate: 6PPD (CAS #101-72-4):*</u> 30-day NOEC (*O. latipes*, Japanese rice fish) = 0.004 mg/L (measured) (GLP-compliant, OECD Guideline 210) (Klimisch Score 1, reliable without restriction).
- In summary, while modeled chronic values on IPPD suggests a Moderate score (1 – 10 mg/L), measured acute LC_{50} values are < 1 mg/L for all three trophic levels, and the chronic values are expected to be lower than acute values. The chronic values of < 1 mg/L would translate to a High score. Therefore, ToxServices did not rely on the modeled data on IPPD to score this endpoint. Instead, ToxServices relied on a measured NOEC for the surrogate 6PPD to score this endpoint, which leads to a Very High score.

<u>Environmental Fate (Fate)</u>

Persistence (P) Score (vH, H, M, L, or vL): *H*
IPPD was assigned a score of High for persistence based on biodegradation taking 75 days in the main compartment of soil. GreenScreen® criteria classify chemicals as a High hazard for persistence when the half-life in soil is >60 to 180 days (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.

- ECHA 2021a
    - A ready biodegradability study conducted according to a method similar to 40 CFR 796.3100 and OECD Guideline 301B (GLP compliance not reported) was performed with acclimated bacterial seed (adaption not reported) exposed to IPPD (80.35% purity) at 30.4 mg/L for 32 days under aerobic conditions. At the end of the exposure period, the level of degradation was 18.9% (Klimisch 2, reliable with restriction).
    - A ready biodegradability test conducted according to OECD Guideline 301C (Modified MITI Test) (GLP compliance not reported) was performed with activated sludge (adaption not reported) exposed to IPPD (purity not reported) at 100 mg/L for 2 weeks. At the end of the exposure period, the level of degradation was 2.2% (Klimisch 2, reliable with restriction).
- U.S. EPA 2017b
    - The BIOWIN modeling Ready Biodegradable Predictor indicates that IPPD is not expected to be readily biodegradable. Fugacity modeling (MCI method) predicts 12% will partition to water with a half-life of 37.5 days, 2.98% will partition to sediment with a half-life of 337.5 days, and 85.1% will partition to soil with a half-life of 75 days (Appendix I).

Bioaccumulation (B) Score (vH, H, M, L, or vL): vL
IPPD was assigned a score of Very Low for bioaccumulation based on a measured log K_{ow} of 2.77. GreenScreen® criteria classify chemicals as a Very Low hazard for bioaccumulation when the log K_{ow} <4 (CPA 2018b). The confidence in the score is high as it is based on measured log K_{ow} 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 2021a
    - Low K_{ow} = 2.77 (measured)
- U.S. EPA 2017b
    - BCFBAF predicts a BCF of 31.24 using the regression-based model based on a measured log K_{ow} of 2.77, and a BCF/BAF of 38 using the Arnot-Gobas model for the upper trophic level, taking metabolism into consideration (Appendix I).

Physical Hazards (Physical)

Reactivity (Rx) Score (vH, H, M, or L): *L*
IPPD 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 IPPD. These procedures are listed in the GHS (UN 2021).
    - Based on the structure of its components or moieties, IPPD is not considered explosive or self-reactive due to lack of functional groups associated with explosive or self-reactive properties (See Appendix J).

- Based on the structure of its components or moieties, IPPD 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

IPPD 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 2021a
  - In a GLP-compliant solid flammability study conducted according to UN Guideline N.1/EU Method A.10, IPPD was not considered to be readily combustible (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, aquatic toxicity, persistence and bioaccumulation, and *in vitro* testing for carcinogenicity, genotoxicity, and endocrine activity. 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 2020). 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 2020):

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

As shown in Table 4, Type I (input data) uncertainties in IPPD's NAMs dataset include the lack of sufficient data on carcinogenicity, endocrine activity, respiratory sensitization, chronic aquatic toxicity, and persistence along with a lack of validated test methods for respiratory sensitization. IPPD'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 and carcinogenicity assays in mimicking *in vivo* metabolic systems, the uncertain *in vivo* relevance of *in silico* modeling of endocrine receptor binding and *in vitro* high throughput testing data, the limitations in the examination of structural alerts for respiratory sensitization evaluation that does not account for non-immunologic mechanisms of respiratory sensitization, the lack of guidance from the ECHA framework to subclassify respiratory sensitizers to Category 1A and 1B, and the unreliable predictions of chronic aquatic toxicity by ECOSAR. Some of IPPD's type II uncertainties were alleviated by the use of *in vitro* test batteries and/or in combination of *in vivo* data.

Table 4: Summary of NAMs Used in the GreenScreen® Assessment, Including Uncertainty Analyses

Uncertainty Analyses (OECD 2020)	
Type I Uncertainty: Data/Model Input	**Carcinogenicity**: Experimental data are available for a weak surrogate. **Endocrine activity**: Experimental data are available for a weak surrogate. **Genotoxicity:** The UDS assay method (OECD Guideline 482) has been deleted due to lack of use and poorer performance compared to other standard tests.[11] **Respiratory sensitization**: No experimental data are available and there are no validated test methods.

[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.org/env/ehs/testing/Draft_Intro_Genotoxicity%20TGs%20September%202014.pdf

	Chronic aquatic toxicity: Experimental data are available for only one trophic level and a weak surrogate. **Persistence**: Insufficient experimental data are available for hazard classification.
Type II Uncertainty: Extrapolation Output	**Carcinogenicity**: Toxtree only identifies structural alerts (SAs), and no applicability domain can be defined (Toxtree 2018). Identification of morphologically transformed colonies in the *in vitro* mammalian cell transformation assay could be subjective. The mechanism leading to cell transformations is not fully understood. The test does not inform *in vivo* potency, species-specificity or tissue-specificity of cell transformations, and is being validated for mono-constituent substances only[12]. **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[13]. The mammalian cell gene mutation assay (as defined in OECD Guideline 476) only detects gene mutations, and the exogenous 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).[14] The *in vitro* chromosome aberration assay (as defined in OECD Guideline 473) does not measure aneuploidy and it only measures structural chromosomal aberrations. The exogenous metabolic activation system does not entirely mirror *in vivo* metabolism[15]. The *in vitro* SCE assay (as defined in OECD 479, a guideline deleted in 2014) detects reciprocal exchange of DNA without providing the underlying mechanism of action[16]. **Endocrine activity:** The *in vivo* relevance of EDSP Tox 21 screening assays and QSAR modeling of receptor binding is unknown due to lack of consideration of metabolism and other toxicokinetic factors. **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-

[12] https://www.oecd.org/env/ehs/testing/Guidance-Document-on-the-in-vitro-Syrian-Hamster-Embryo-Cell-Transformation-Assay.pdf
[13] https://www.oecd-ilibrary.org/docserver/9789264071247-en.pdf?expires=1614097593&id=id&accname=guest&checksum=89925F80B9F4BD2FFC6E90F94A0EE427
[14] https://www.oecd-ilibrary.org/docserver/9789264264809-en.pdf?expires=1614097800&id=id&accname=guest&checksum=C0DE371FB9C5A878E66C9AB7F84E6BBE
[15] https://www.oecd-ilibrary.org/docserver/9789264264649-en.pdf?expires=1614098015&id=id&accname=guest&checksum=6A4F9CE52EA974F5A74793DD54D54352
[16] https://www.oecd.org/env/ehs/testing/Draft_Intro_Genotoxicity%20TGs%20September%202014.pdf

	immunologic mechanisms for respiratory sensitization. The ECHA framework did not provide guidance on how to subclassify chemicals to Category 1A and 1B. **Chronic aquatic toxicity:** The modeled chronic aquatic toxicity values are greater than the most conservative measured acute LC_{50} values, rendering the predictions unreliable.	
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 *In vitro* data: cell transformation assay
Mutagenicity	Y	*In vitro* data: Bacterial reverse mutation assay/*in vitro* gene mutation assay/*in vitro* chromosome aberration assay/ *in vitro* SCE assay
Reproductive toxicity	N	
Developmental toxicity	N	
Endocrine activity	Y	*In vitro* high throughput data: EDSP Tox 21 screening assays; *In silico* modeling: 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	
Chronic aquatic toxicity	Y	*In silico* modeling: ECOSAR
Persistence	Y	*In silico* modeling: EPI Suite™
Bioaccumulation	Y	*In silico* modeling: EPI Suite™

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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 IPPD (CAS #101-72-4)

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	IPPD	101-72-4	*L*	L	*H*	M	*M*	M		M	*M*	DG	H	*M*	L	*M*	vH	*vH*	*H*	vL	*L*	L

Table 3: Hazard Summary Table

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

Table 4

Chemical Name	Preliminary GreenScreen® Benchmark Score
IPPD	1

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

Table 6

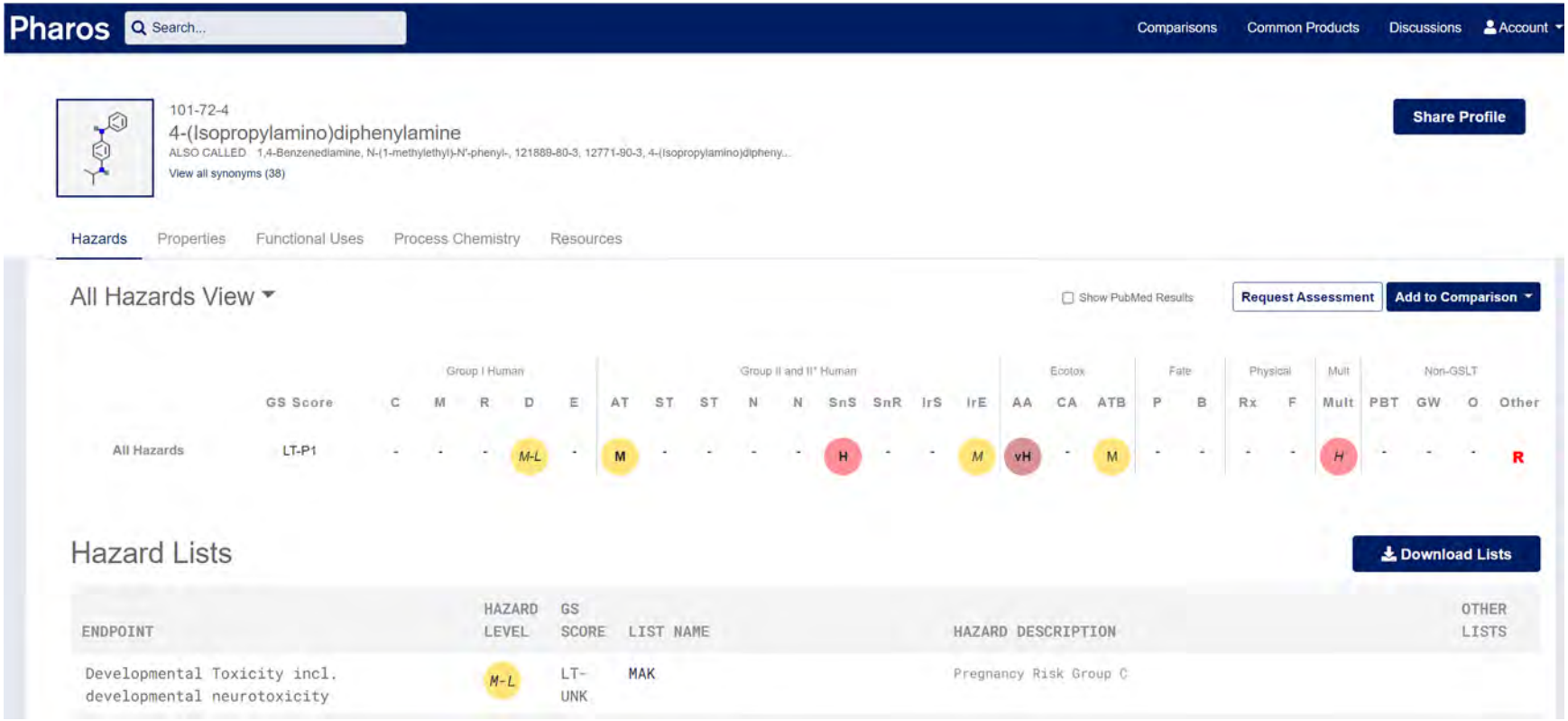
Chemical Name	Final GreenScreen® Benchmark Score
IPPD	1

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												1
2												
3												
4												

APPENDIX C: Pharos Output for IPPD (CAS #101-72-4)



Acute Mammalian Toxicity	M	LT-UNK	EU - GHS (H-Statements)	H302 - Harmful if swallowed [Acute toxicity (oral) - Category 4]	+5
	H-M	LT-UNK	GHS - Australia	H302 - Harmful if swallowed [Acute toxicity (oral) - Category 4]	
	M	LT-UNK	GHS - Japan	H302 - Harmful if swallowed [Acute Toxicity (oral) - Category 4]	
	M	LT-UNK	GHS - Korea	H302 - Harmful if swallowed [Acute toxicity (oral) - Category 4]	
	M	LT-UNK	GHS - New Zealand	6.1D (oral) - Acutely toxic	
	pC	NoGS	EU - Manufacturer REACH hazard submissions	H302 - Harmful if swallowed (unverified) [Acute toxicity (oral) - Category 4]	
Skin Sensitization	H	LT-UNK	MAK	Sensitizing Substance Sh - Danger of skin sensitization	+6
	H-M	LT-UNK	EU - GHS (H-Statements)	H317 - May cause an allergic skin reaction [Skin sensitization - Category 1]	
	H	LT-UNK	GHS - Japan	H317 - May cause an allergic skin reaction [Skin sensitizer - Category 1]	
	H	LT-UNK	GHS - Korea	H317 - May cause an allergic skin reaction [Skin sensitization - Category 1]	
	H	LT-UNK	GHS - New Zealand	6.5B (contact) - Contact sensitisers (Cat. 1)	
	H-M	LT-UNK	GHS - Australia	H317 - May cause an allergic skin reaction [Skin sensitization - Category 1]	
	pC	NoGS	EU - Manufacturer REACH hazard submissions	H317 - May cause an allergic skin reaction (unverified) [Skin sensitization - Category 1]	

Eye Irritation/Corrosivity	M	LT-UNK	GHS - Japan	H319 - Causes serious eye irritation [Serious eye damage / eye irritation – Category 2B]	
Acute Aquatic Toxicity	vH	LT-UNK	EU - GHS (H-Statements)	H400 - Very toxic to aquatic life [Hazardous to the aquatic environment (acute) – Category 1]	+3
	vH	LT-UNK	GHS - Japan	H400 - Very toxic to aquatic life [Hazardous to the aquatic environment (acute) – Category 1]	
	vH	LT-UNK	GHS - Korea	H400 - Very toxic to aquatic life [Hazardous to the aquatic environment (acute) – Category 1]	
	pC	NoGS	EU - Manufacturer REACH hazard submissions	H400 - Very toxic to aquatic life (unverified) [Hazardous to the aquatic environment (acute) - Category 1]	
Terrestrial Ecotoxicity	M	NoGS	GHS - New Zealand	9.3B - Ecotoxic to terrestrial vertebrates	
T & P and/or B [(Chronic Aquatic Toxicity and Persistence) or (Acute Aquatic Toxicity and Persistence and/or Bioaccumulation)]	U	LT-P1	EU - GHS (H-Statements)	H410 - Very toxic to aquatic life with long lasting effects [Hazardous to the aquatic environment (chronic) - Category 1]	+6
	U	LT-P1	GHS - Australia	H410 - Very toxic to aquatic life with long lasting effects [Hazardous to the aquatic environment (chronic) - Category 1]	
	U	LT-P1	GHS - New Zealand	9.1A (algal) - Very ecotoxic in the aquatic environment	
	U	LT-P1	GHS - New Zealand	9.1A (fish) - Very ecotoxic in the aquatic environment	
	U	LT-P1	GHS - Korea	H410 - Very toxic to aquatic life with long lasting effects [Hazardous to the aquatic environment (chronic) - Category 1]	
	U	LT-P1	GHS - New Zealand	9.1B (crustacean) - Very ecotoxic in the aquatic environment	
	pC	NoGS	EU - Manufacturer REACH hazard submissions	H410 - Very toxic to aquatic life with long lasting effects (unverified) [Hazardous to the aquatic environment (chronic)	

Human and/or Aquatic toxicity and/or Persistence and/or Bioaccumulation	U	LT-P1	German FEA - Substances Hazardous to Waters	Class 3 - Severe Hazard to Waters	
Systemic Toxicity/Organ Effects [Single Exposure] and/or Neurotoxicity [Single Exposure]	H	LT-UNK	GHS - Japan	H371 - May cause damage to organs [Specific target organs/systemic toxicity following single exposure - Category 2]	+1
	H	LT-UNK	GHS - New Zealand	6.9B (oral) - Harmful to human target organs or systems (Cat. 2)	
T & P and/or B [(Chronic Aquatic Toxicity and sometimes Persistence) or (Acute Aquatic Toxicity and Persistence and/or Bioaccumulation)]	U	LT-P1	GHS - Japan	H410 - Very toxic to aquatic life with long lasting effects [Hazardous to the aquatic environment (chronic) - Category 1]	
Systemic Toxicity/Organ Effects [Repeated Exposure] and/or Neurotoxicity [Repeated Exposure]	H	LT-UNK	GHS - Japan	H372 - Causes damage to organs through prolonged or repeated exposure [Specific target organs/systemic toxicity following repeated exposure – Category 1]	

Restricted Substance Lists (4)

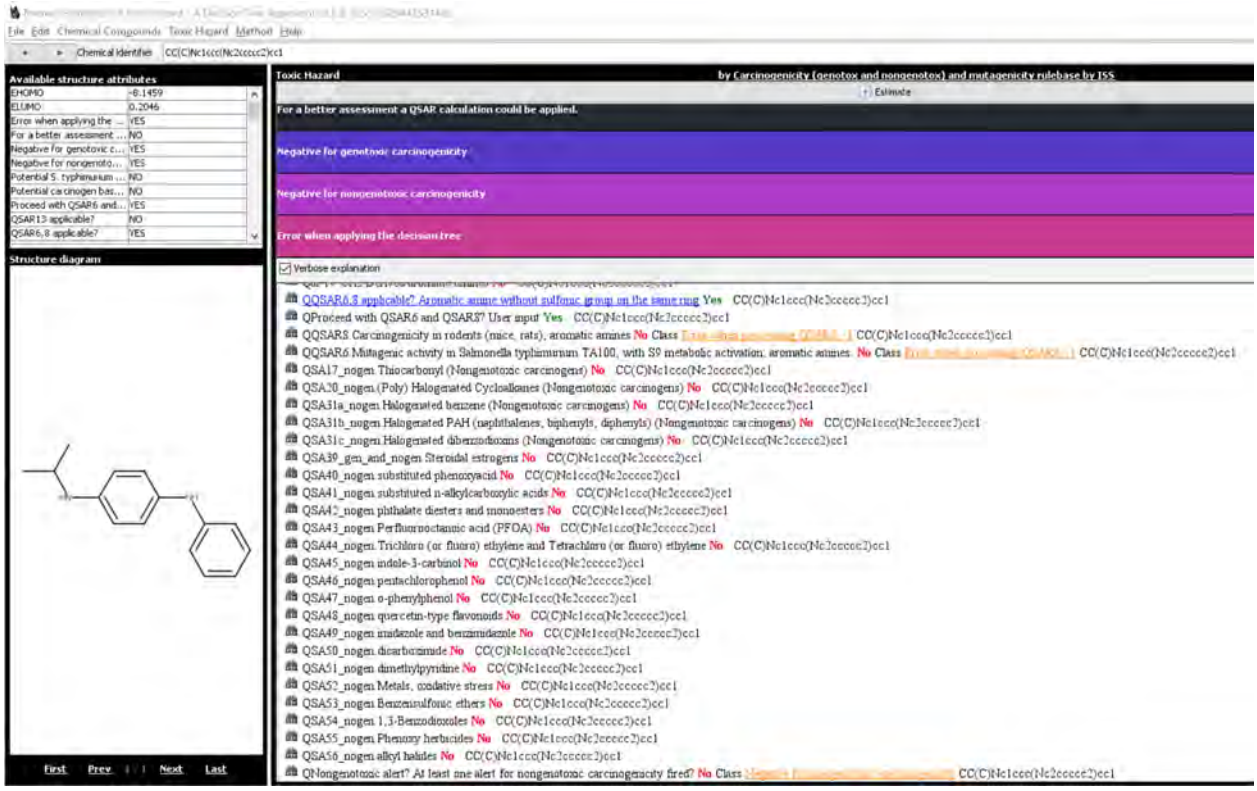
- EU - PACT-RMOA Substances: Substances selected for RMOA or hazard assessment
- Food Contact Chemicals Database (FCCdb): Food Contact Chemicals Database Version 5.0
- Food Contact Chemicals of Concern (FCCoCL): Food Contact Chemicals of Concern List (FCCoCL)
- Food Contact Chemicals of Concern (FCCoCL): Food Contact Chemicals of Concern List (FCCoCL) - TIER 3

Discussions

No discussions have been posted yet.

Ask a question about this chemical in the forums >

APPENDIX D: Toxtree Carcinogenicity Results for IPPD (CAS #101-72-4)

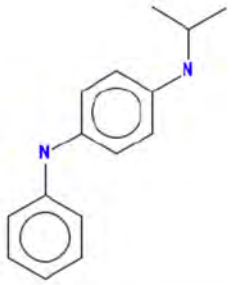


APPENDIX E: VEGA Carcinogenicity Results for IPPD (CAS #101-72-4)

VEGA Carcinogenicity model (CAESAR) 2.1.9 page 1

1. Prediction Summary

Prediction for compound Molecule 0

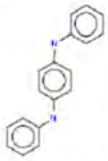


Prediction: Reliability:

Prediction is Carcinogen, but the result shows some critical aspects, which require to be checked:
- 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: c1ccc(cc1)Nc2ccc(cc2)NC(C)C
Experimental value: -
Predicted Carcinogen activity: Carcinogen
P(Carcinogen): 0.585
P(NON-Carcinogen): 0.415
Reliability: the predicted compound could be out of the Applicability Domain of the model
Remarks:
none

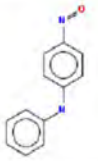
3.1 Applicability Domain:
Similar Compounds, with Predicted and Experimental Values



Compound #1

CAS: 74-31-7
Dataset id: 285 (Test set)
SMILES: c1ccc(cc1)Nc2ccc(cc2)Nc3ccccc3
Similarity: 0.915

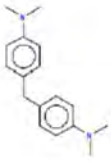
Experimental value: NON-Carcinogen
Predicted value: Carcinogen



Compound #2

CAS: 156-10-5
Dataset id: 583 (Training set)
SMILES: O=Nc1ccc(cc1)Nc2ccccc2
Similarity: 0.868

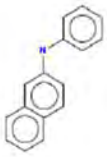
Experimental value: Carcinogen
Predicted value: Carcinogen



Compound #3

CAS: 101-61-1
Dataset id: 467 (Test set)
SMILES: c1cc(ccc1N(C)C)Cc2ccc(cc2)N(C)C
Similarity: 0.854

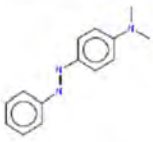
Experimental value: Carcinogen
Predicted value: Carcinogen



Compound #4

CAS: 135-88-6
Dataset id: 643 (Training set)
SMILES: c1ccc(cc1)Nc2ccc3ccccc3(c2)
Similarity: 0.844

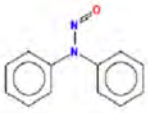
Experimental value: NON-Carcinogen
Predicted value: Carcinogen



Compound #5

CAS: 60-11-7
Dataset id: 262 (Training set)
SMILES: N(=Nc1ccc(cc1)N(C)C)c2ccccc2
Similarity: 0.836

Experimental value: Carcinogen
Predicted value: Carcinogen



Compound #6

CAS: 86-30-6
Dataset id: 582 (Training set)
SMILES: O=NN(c1ccccc1)c2ccccc2
Similarity: 0.836

Experimental value: Carcinogen
Predicted value: Carcinogen

3.2 Applicability Domain:
Measured Applicability Domain Scores

Global AD Index
AD index = 0.657
Explanation: the predicted compound could be out of the Applicability Domain of the model.

Similar molecules with known experimental value
Similarity index = 0.89
Explanation: strongly similar compounds with known experimental value in the training set have been found.

Accuracy of prediction for similar molecules
Accuracy index = 0.485
Explanation: accuracy of prediction for similar molecules found in the training set is not adequate.

Concordance for similar molecules
Concordance index = 0.485
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.

Model class assignment reliability
Pos/Non-Pos difference = 0.17
Explanation: model class assignment is well defined.

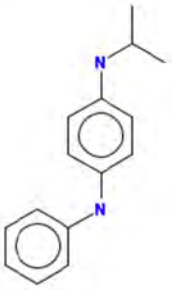
Neural map neurons concordance
Neurons concordance = 1
Explanation: predicted value agrees with experimental values of training set compounds laying in the same neuron.

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

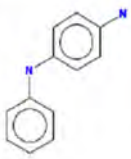


Prediction: Reliability:

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

Compound: Molecule 0
Compound SMILES: c1ccc(cc1)Nc2ccc(cc2)NC(C)C
Experimental value: -
Predicted Carcinogen activity: NON-Carcinogen
Structural alerts: -
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

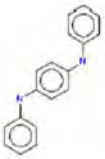


Compound #1

CAS: 101-54-2
Dataset id: 133 (Training set)
SMILES: Nc1ccc(cc1)Nc2ccccc2
Similarity: 0.92

Experimental value: NON-Carcinogen
Predicted value: Carcinogen

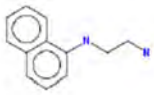
Alerts (not found in the target): SA28 Primary aromatic amine, hydroxyl amine and its derived esters (with restrictions)



Compound #2

CAS: 74-31-7
Dataset id: 887 (Training set)
SMILES: c1ccc(cc1)Nc2ccc(cc2)Nc3ccccc3
Similarity: 0.915

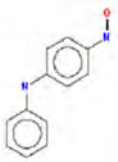
Experimental value: NON-Carcinogen
Predicted value: NON-Carcinogen



Compound #3

CAS: 1465-25-4
Dataset id: 124 (Training set)
SMILES: NCCNc2cccc1ccccc12
Similarity: 0.878

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

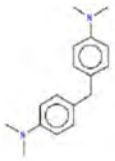


Compound #4

CAS: 156-10-5
Dataset id: 212 (Training set)
SMILES: O=Nc1ccc(cc1)Nc2ccccc2
Similarity: 0.868

Experimental value: Carcinogen
Predicted value: Carcinogen

Alerts (not found in the target): SA25 Aromatic nitroso group



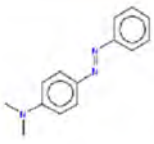
Compound #5

CAS: 101-61-1
Dataset id: 201 (Training set)
SMILES: c1cc(ccc1N(C)C)Cc2ccc(cc2)N(C)C
Similarity: 0.854

Experimental value: Carcinogen
Predicted value: Carcinogen

Alerts (not found in the target): SA28bis Aromatic mono- and dialkylamine

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



Compound #6

CAS: 60-11-7
Dataset id: 1 (Training set)
SMILES: N(=Nc1ccc(cc1)N(C)C)c2ccccc2
Similarity: 0.836

Experimental value: Carcinogen
Predicted value: Carcinogen

Alerts (not found in the target): SA28bis Aromatic mono- and dialkylamine; SA29 Aromatic diazo

3.2 Applicability Domain:
Measured Applicability Domain Scores

Global AD Index
AD index = 0.805
Explanation: the predicted compound could be out of the Applicability Domain of the model.

Similar molecules with known experimental value
Similarity index = 0.917
Explanation: strongly similar compounds with known experimental value in the training set have been found.

Accuracy of prediction for similar molecules
Accuracy index = 0.499
Explanation: accuracy of prediction for similar molecules found in the training set is not adequate.

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

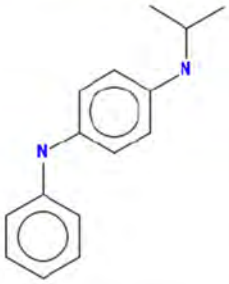
Atom Centered Fragments similarity check
ACF index = 1
Explanation: all atom centered fragment of the compound have been found in the compounds of the training set.

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

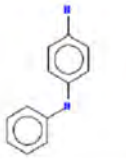


Prediction: Reliability:

Prediction is Carcinogen, but the result may be not reliable. A check of the information given in the following section should be done, paying particular attention to the following issues:
- 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. 36

Compound: Molecule 0
Compound SMILES: c1ccc(cc1)Nc2ccc(cc2)NC(C)C
Experimental value: -
Predicted Mutagen activity: Carcinogen
No. alerts for carcinogenicity: 1
Structural alerts: Carcinogenity alert no. 36
Reliability: the predicted compound is outside the Applicability Domain of the model
Remarks:
none

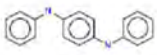
3.1 Applicability Domain:
Similar Compounds, with Predicted and Experimental Values



Compound #1

CAS: N.A.
Dataset id: 1299 (Training set)
SMILES: Nc1ccc(cc1)Nc2ccccc2
Similarity: 0.92

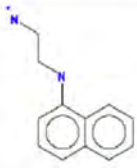
Experimental value: NON-Carcinogen
Predicted value: Possible NON-Carcinogen



Compound #2

CAS: N.A.
Dataset id: 285 (Training set)
SMILES: c1ccc(cc1)Nc2ccc(cc2)Nc3ccccc3
Similarity: 0.915

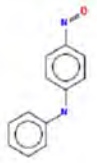
Experimental value: NON-Carcinogen
Predicted value: Possible NON-Carcinogen



Compound #3

CAS: N.A.
Dataset id: 1180 (Training set)
SMILES: c1ccc2c(c1)cccc2(NCC[NH3+])
Similarity: 0.876

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

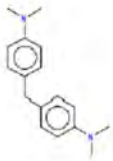


Compound #4

CAS: N.A.
Dataset id: 588 (Training set)
SMILES: O=Nc1ccc(cc1)Nc2ccccc2
Similarity: 0.868

Experimental value: Carcinogen
Predicted value: Carcinogen

Alerts (not found in the target): Carcinogenity alert no. 63



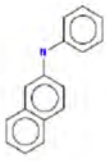
Compound #5

CAS: N.A.
Dataset id: 466 (Training set)
SMILES: c1cc(ccc1N(C)C)Cc2ccc(cc2)N(C)C
Similarity: 0.854

Experimental value: Carcinogen
Predicted value: Carcinogen

Alerts (not found in the target): Carcinogenity alert no. 29; Carcinogenity alert no. 30; Carcinogenity alert no. 31

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

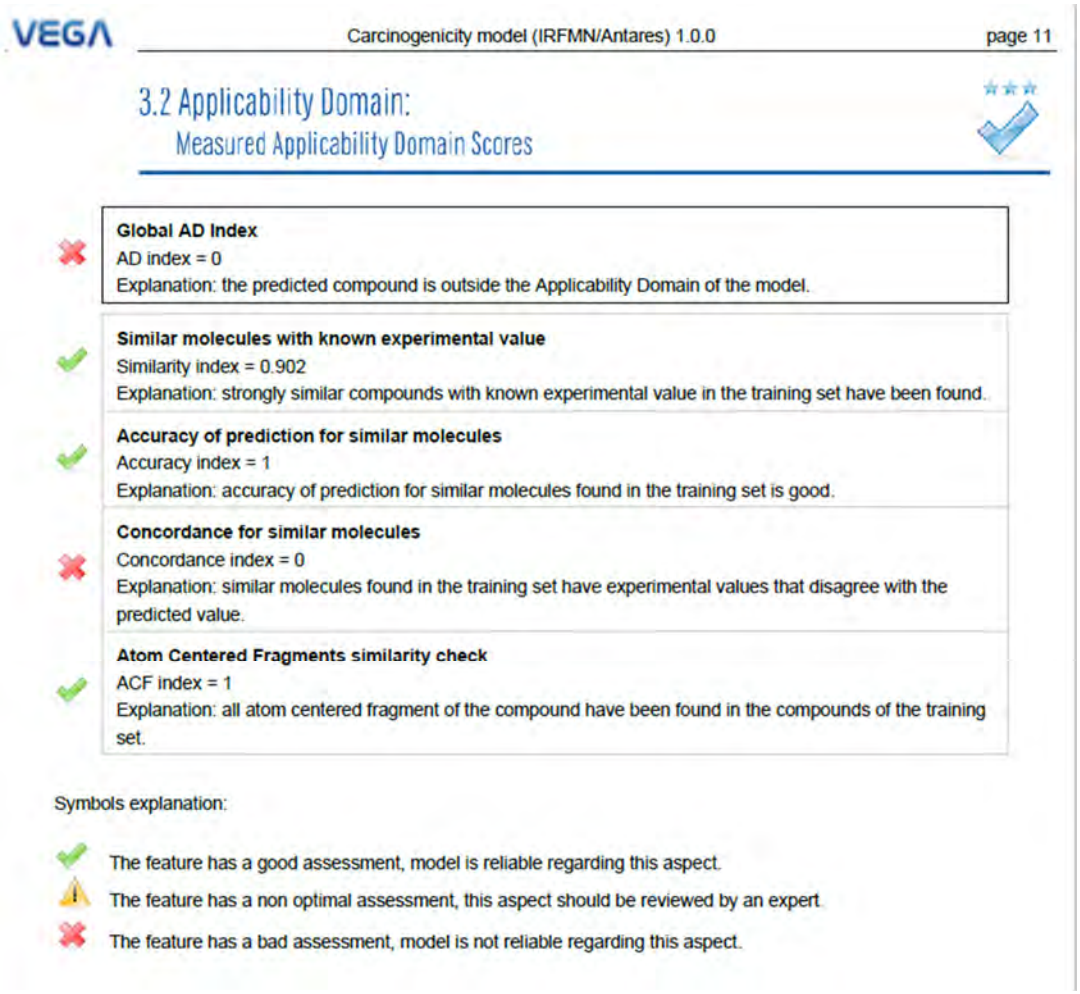


Compound #6

CAS: N.A.
Dataset id: 654 (Training set)
SMILES: c1ccc(cc1)Nc2ccc3ccccc3(c2)
Similarity: 0.844

Experimental value: NON-Carcinogen
Predicted value: Carcinogen

Alerts (not found in the target): Carcinogenity alert no. 35



VEGA Carcinogenicity model (IRFMN/Antares) 1.0.0 page 11

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 = 0.902
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 = 0
Explanation: 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.

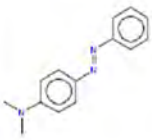
4.1 Reasoning:
Relevant Chemical Fragments and Moieties

(Molecule 0) Reasoning on fragments/structural alerts:

Fragment found: Carcinogenity alert no. 36

Structural alert for carcinogenity defined by the SMARTS: CNc1ccc(N)cc1

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

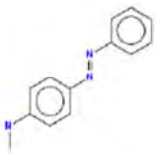


CAS: N.A.
Dataset id: 262 (Training set)
SMILES: N(=Nc1ccc(cc1)N(C)C)c2ccccc2
Similarity: 0.836

Experimental value: Carcinogen
Predicted value: Carcinogen

Alerts (found also in the target): Carcinogenity alert no. 36

Alerts (not found in the target): Carcinogenity alert no. 71

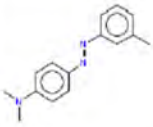


CAS: N.A.
Dataset id: 1248 (Training set)
SMILES: N(=Nc1ccc(cc1)NC)c2ccccc2
Similarity: 0.834

Experimental value: Carcinogen
Predicted value: Carcinogen

Alerts (found also in the target): Carcinogenity alert no. 36

Alerts (not found in the target): Carcinogenity alert no. 71



CAS: N.A.
Dataset id: 447 (Training set)
SMILES: N(=Nc1cccc(c1)C)c2ccc(cc2)N(C)C
Similarity: 0.827

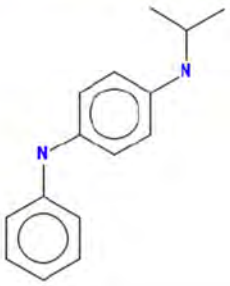
Experimental value: Carcinogen
Predicted value: Carcinogen

Alerts (found also in the target): Carcinogenity alert no. 36

Alerts (not found in the target): Carcinogenity alert no. 33; Carcinogenity alert no. 71

1. Prediction Summary

Prediction for compound Molecule 0

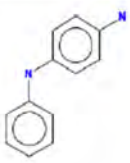


Prediction: Reliability:

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:
- 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
The following relevant fragments have been found: Carcinogenity alert no. 42

Compound: Molecule 0
Compound SMILES: c1ccc(cc1)Nc2ccc(cc2)NC(C)C
Experimental value: -
Predicted Mutagen activity: Carcinogen
No. alerts for carcinogenicity: 1
Structural alerts: Carcinogenity alert no. 42
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

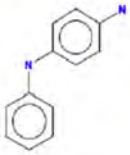


Compound #1

CAS: 101-54-2
Dataset id: 657 (Training set)
SMILES: Nc1ccc(cc1)Nc2ccccc2
Similarity: 0.92

Experimental value: NON-Carcinogen
Predicted value: Carcinogen

Alerts (found also in the target): Carcinogenity alert no. 42

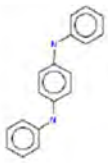


Compound #2

CAS: 2198-59-6
Dataset id: 973 (Training set)
SMILES: Nc1ccc(cc1)Nc2ccccc2
Similarity: 0.92

Experimental value: NON-Carcinogen
Predicted value: Carcinogen

Alerts (found also in the target): Carcinogenity alert no. 42

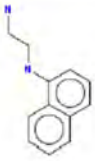


Compound #3

CAS: 74-31-7
Dataset id: 639 (Training set)
SMILES: c1ccc(cc1)Nc2ccc(cc2)Nc3ccccc3
Similarity: 0.915

Experimental value: NON-Carcinogen
Predicted value: Carcinogen

Alerts (found also in the target): Carcinogenity alert no. 42



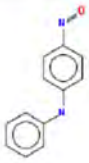
Compound #4

CAS: 1465-25-4
Dataset id: 98 (Training set)
SMILES: NCCNc2cccc1ccccc12
Similarity: 0.878

Experimental value: NON-Carcinogen
Predicted value: Carcinogen

Alerts (found also in the target): Carcinogenity alert no. 42

Alerts (not found in the target): Carcinogenity alert no. 30



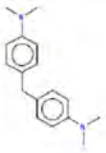
Compound #5

CAS: 156-10-5
Dataset id: 173 (Training set)
SMILES: O=Nc1ccc(cc1)Nc2ccccc2
Similarity: 0.868

Experimental value: Carcinogen
Predicted value: Carcinogen

Alerts (found also in the target): Carcinogenity alert no. 42

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



Compound #6

CAS: 101-61-1
Dataset id: 162 (Training set)
SMILES: c1cc(ccc1N(C)C)Cc2ccc(cc2)N(C)C
Similarity: 0.854

Experimental value: Carcinogen
Predicted value: Carcinogen

Alerts (found also in the target): Carcinogenity alert no. 42

Alerts (not found in the target): Carcinogenity alert no. 41

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 = 0.918
Explanation: strongly similar compounds with known experimental value in the training set have been found.

Accuracy of prediction for similar molecules
Accuracy index = 0
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.

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.

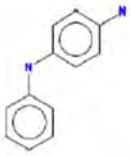
4.1 Reasoning:
Relevant Chemical Fragments and Moieties

(Molecule 0) Reasoning on fragments/structural alerts:

Fragment found: Carcinogenity alert no. 42

Structural alert for carcinogenity defined by the SMARTS: Nc1ccccc1

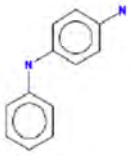
Following, the most similar compounds from the model's dataset having the same fragment.



CAS: 101-54-2
Dataset id: 657 (Training set)
SMILES: Nc1ccc(cc1)Nc2ccccc2
Similarity: 0.92

Experimental value: NON-Carcinogen
Predicted value: Carcinogen

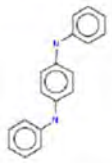
Alerts (found also in the target): Carcinogenity alert no. 42



CAS: 2198-59-6
Dataset id: 973 (Training set)
SMILES: Nc1ccc(cc1)Nc2ccccc2
Similarity: 0.92

Experimental value: NON-Carcinogen
Predicted value: Carcinogen

Alerts (found also in the target): Carcinogenity alert no. 42



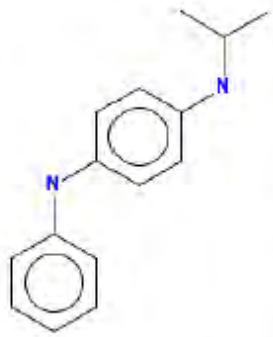
CAS: 74-31-7
Dataset id: 639 (Training set)
SMILES: c1ccc(cc1)Nc2ccc(cc2)Nc3ccccc3
Similarity: 0.915

Experimental value: NON-Carcinogen
Predicted value: Carcinogen

Alerts (found also in the target): Carcinogenity alert no. 42

1 Prediction Summary

Prediction for compound Molecule 0

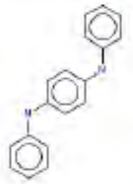


Prediction: Reliability:

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:
- 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
- some atom centered fragments of the compound have not been found in the compounds of the training set or are rare fragments (1 infrequent fragments found)

Compound: Molecule 0
Compound SMILES: c1ccc(cc1)Nc2ccc(cc2)NC(C)C
Experimental value: -
Predicted Oral Carcinogenic class: Carcinogen
Reliability: the predicted compound is outside the Applicability Domain of the model
Remarks:
none

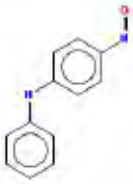
3.1 Applicability Domain:
Similar Compounds, with Predicted and Experimental Values



Compound #1

CAS: 74-31-7
Dataset id: 598 (Training set)
SMILES: c1ccc(cc1)Nc2ccc(cc2)Nc3ccccc3
Similarity: 0.915

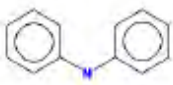
Experimental value: NON-Carcinogen
Predicted value: Carcinogen



Compound #2

CAS: 156-10-5
Dataset id: 227 (Test set)
SMILES: O=Nc1ccc(cc1)Nc2ccccc2
Similarity: 0.868

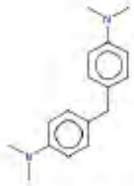
Experimental value: Carcinogen
Predicted value: Carcinogen



Compound #3

CAS: 122-39-4
Dataset id: 484 (Test set)
SMILES: c1ccc(cc1)Nc2ccccc2
Similarity: 0.856

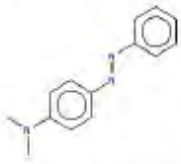
Experimental value: NON-Carcinogen
Predicted value: Carcinogen



Compound #4

CAS: 101-61-1
Dataset id: 201 (Training set)
SMILES: c1cc(ccc1N(C)C)Cc2ccc(cc2)N(C)C
Similarity: 0.854

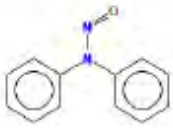
Experimental value: Carcinogen
Predicted value: Carcinogen



Compound #5

CAS: 60-11-7
Dataset id: 122 (Training set)
SMILES: N(=Nc1ccc(cc1)N(C)C)c2ccccc2
Similarity: 0.836

Experimental value: Carcinogen
Predicted value: Carcinogen



Compound #6

CAS: 86-30-6
Dataset id: 226 (Training set)
SMILES: O=NN(c1ccccc1)c2ccccc2
Similarity: 0.836

Experimental value: Carcinogen
Predicted value: Carcinogen

3.2 Applicability Domain:
Measured Applicability Domain Scores

Global AD Index
AD index = 0.558
Explanation: the predicted compound is outside the Applicability Domain of the model.

Similar molecules with known experimental value
Similarity index = 0.89
Explanation: strongly similar compounds with known experimental value in the training set have been found.

Accuracy of prediction for similar molecules
Accuracy index = 0.485
Explanation: accuracy of prediction for similar molecules found in the training set is not adequate.

Concordance for similar molecules
Concordance index = 0.485
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 = 0.85
Explanation: some atom centered fragments of the compound have not been found in the compounds of the training set or are rare fragments (1 infrequent fragments found).

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.

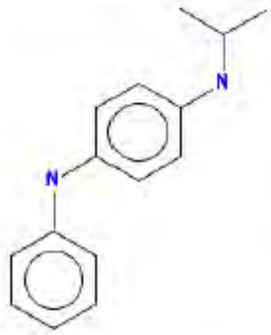
4.1 Reasoning:
Relevant Chemical Fragments and Moieties

(Molecule 0) Reasoning on rare and missing Atom Centered Fragments.
The following Atom Centered Fragments have been found in the molecule, but they are not found or rarely found in the model's training set:

Fragment defined by the SMILES: N(c)c
The fragment has less than 3 occurrences in the model's training set

1 Prediction Summary

Prediction for compound Molecule 0

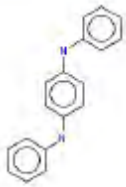


Prediction: Reliability:

Prediction is Carcinogen, but the result shows some critical aspects, which require to be checked:
- 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: c1ccc(cc1)Nc2ccc(cc2)NC(C)C
Experimental value: -
Predicted Inhalation Carcinogenic class: Carcinogen
Reliability: the predicted compound could be out of the Applicability Domain of the model
Remarks:
none

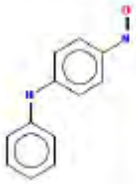
3.1 Applicability Domain:
Similar Compounds, with Predicted and Experimental Values



Compound #1

CAS: 74-31-7
Dataset id: 579 (Training set)
SMILES: c1ccc(cc1)Nc2ccc(cc2)Nc3ccccc3
Similarity: 0.915

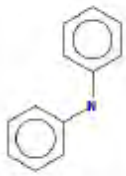
Experimental value: NON-Carcinogen
Predicted value: Carcinogen



Compound #2

CAS: 156-10-5
Dataset id: 198 (Training set)
SMILES: O=Nc1ccc(cc1)Nc2ccccc2
Similarity: 0.868

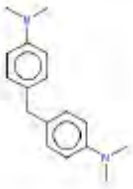
Experimental value: Carcinogen
Predicted value: Carcinogen



Compound #3

CAS: 122-39-4
Dataset id: 456 (Training set)
SMILES: c1ccc(cc1)Nc2ccccc2
Similarity: 0.856

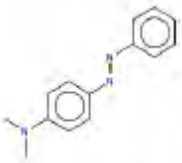
Experimental value: NON-Carcinogen
Predicted value: NON-Carcinogen



Compound #4

CAS: 101-61-1
Dataset id: 170 (Training set)
SMILES: c1cc(ccc1N(C)C)Cc2ccc(cc2)N(C)C
Similarity: 0.854

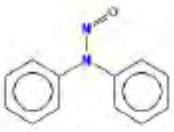
Experimental value: Carcinogen
Predicted value: Carcinogen



Compound #5

CAS: 60-11-7
Dataset id: 100 (Training set)
SMILES: N(=Nc1ccc(cc1)N(C)C)c2ccccc2
Similarity: 0.836

Experimental value: Carcinogen
Predicted value: Carcinogen



Compound #6

CAS: 86-30-6
Dataset id: 197 (Training set)
SMILES: O=NN(c1ccccc1)c2ccccc2
Similarity: 0.836

Experimental value: Carcinogen
Predicted value: Carcinogen

3.2 Applicability Domain:
Measured Applicability Domain Scores

Global AD Index
AD index = 0.657
Explanation: the predicted compound could be out of the Applicability Domain of the model.

Similar molecules with known experimental value
Similarity index = 0.89
Explanation: strongly similar compounds with known experimental value in the training set have been found.

Accuracy of prediction for similar molecules
Accuracy index = 0.485
Explanation: accuracy of prediction for similar molecules found in the training set is not adequate.

Concordance for similar molecules
Concordance index = 0.485
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 IPPD (CAS #101-72-4)

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	Aromatic Amine Type Compounds

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		INC_OUT	NEG_OUT	POS_OUT	INC_OUT

DTU-developed models

Endocrine and Molecular Endpoints

	Exp	Battery	CASE Ultra	Leadscope	SciQSAR
Estrogen Receptor α Binding, Full training set (Human *in vitro*)		NEG_IN	NEG_IN	NEG_OUT	NEG_IN
Estrogen Receptor α Binding, Balanced Training Set (Human *in vitro*)		NEG_IN	NEG_IN	NEG_IN	NEG_IN
Estrogen Receptor α Activation (Human *in vitro*)		NEG_OUT	NEG_OUT	INC_OUT	NEG_IN
Estrogen Receptor Activation, CERAPP data (*in vitro*)		N/A	N/A	NEG_IN	N/A
Androgen Receptor Inhibition (Human *in vitro*)		NEG_IN	NEG_IN	NEG_IN	NEG_IN
Androgen Receptor Binding, CoMPARA data (*in vitro*)		N/A	N/A	NEG_OUT	N/A
Androgen Receptor Inhibition, CoMPARA data (*in vitro*)		N/A	N/A	INC_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	POS_IN	N/A
Thyroperoxidase (TPO) inhibition QSAR2 (Rat *in vitro*)		N/A	N/A	POS_IN	N/A
Thyroid Receptor α Binding (Human *in vitro*)					
- mg/L			36201.2	633.4217	20.85081
- µM			159955.8	2798.788	92.12978
- Positive for $IC_{50} \leq 10$ µM					
- Positive for $IC_{50} \leq 100$ µM					
- Domain		OUT	OUT	OUT	OUT
Thyroid Receptor β Binding (Human *in vitro*)					
- mg/L			7323.576	9.165854	360.017
- µM			32359.38	40.49953	1590.743
- Positive for $IC_{50} \leq 10$ µM					

	Exp	Battery	CASE Ultra	Leadscope	SciQSAR
- Positive for $IC_{50} \leq 100$ µM					
- Domain		OUT	OUT	OUT	OUT
Arylhydrocarbon (AhR) Activation – Rational final model (Human *in vitro*)		N/A	N/A	NEG_IN	N/A
Arylhydrocarbon (AhR) Activation – Random final model (Human *in vitro*)		N/A	N/A	INC_OUT	N/A
Pregnane X Receptor (PXR) Binding (Human *in vitro*)	N/A	NEG_OUT	POS_OUT	INC_OUT	NEG_IN
Pregnane X Receptor (PXR) Binding (Human *in vitro*) NEW		N/A	N/A	NEG_IN	N/A
Pregnane X Receptor (PXR) Activation (Human *in vitro*)		N/A	N/A	NEG_OUT	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_OUT	N/A
Constitutive Androstane Receptor (CAR) Inhibition at max. 20 µM (*in vitro*)		N/A	N/A	INC_OUT	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*)	POS	N/A	N/A	INC_OUT	N/A

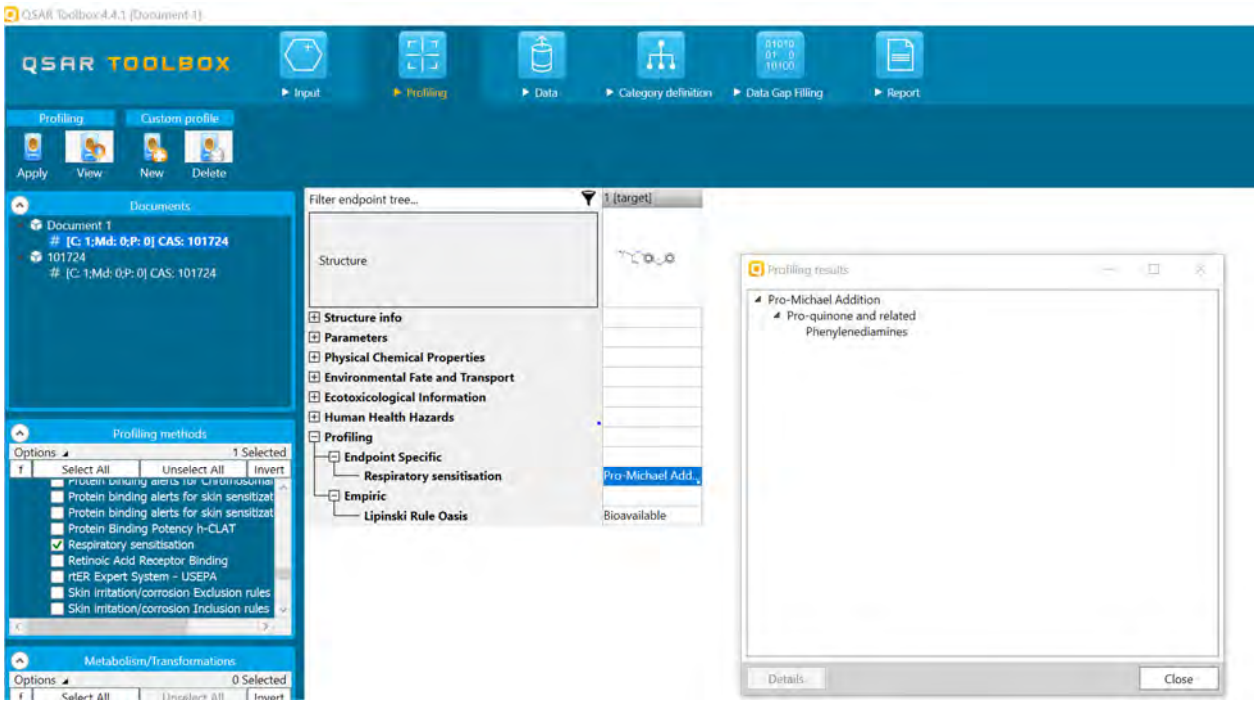
DTU-developed models

Estrogen Receptor Binding, alerts in:	
- parent only	Non binder, without OH or NH2 group
- metabolites from *in vivo* Rat metabolism simulator only	Strong binder, NH2 group; Strong binder, OH group; Moderate binder, NH2 group; Weak binder, NH2 group; Weak binder, OH group
- metabolites from Rat liver S9 metabolism simulator only	Strong binder, NH2 group; Strong binder, OH group; Moderate binder, NH2 group; Moderate binder, OH grooup
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 IPPD (CAS #101-72-4)



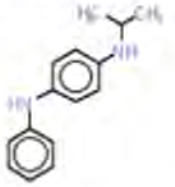
APPENDIX H: ECOSAR Modeling Results for IPPD (CAS #101-72-4)

Organic Module Report

Results of Organic Module Evaluation

CAS	Name	SMILES
101724	1,4-Benzenediamine, N-(1-methylethyl)-N -phenyl-	N(c(ccc(Nc(cccc1)c1)c2)c2)C(C)C

Structure



Details	
Mol Wt	226.32
Selected LogKow	2.77
Selected Water Solubility (mg/L)	15
Selected Melting Point (°C)	74
Estimated LogKow	3.28
Estimated Water Solubility (mg/L)	168.68
Measured LogKow	
Measured Water Solubility (mg/L)	
Measured Melting Point (°C)	74

Class Results:	

Neutral Organics

Organism	Duration	End Point	Concentration (mg/L)	Max Log Kow	Flags

Class Results:	

Organism	Duration	End Point	Concentration (mg/L)	Max Log Kow	Flags
Fish	96h	LC50	37.83	5	• Chemical may not be soluble enough to measure this predicted effect. If the effect level exceeds the water solubility by 10X, typically no effects at saturation (NES) are reported
Daphnid	48h	LC50	23.06	5	• Chemical may not be soluble enough to measure this predicted effect. If the effect level exceeds the water solubility by 10X, typically no effects at saturation (NES) are reported
Green Algae	96h	EC50	23.05	6.4	• Chemical may not be soluble enough to measure this predicted effect. If the effect level exceeds the water solubility by 10X, typically no effects at saturation (NES) are reported
Fish		ChV	4.02	8	
Daphnid		ChV	2.74	8	
Green Algae		ChV	7.07	8	
Fish (SW)	96h	LC50	47.84	5	• Chemical may not be soluble enough to measure this predicted effect. If the effect level exceeds the water solubility by 10X, typically no effects at saturation (NES) are reported
Mysid	96h	LC50	21.1	5	• Chemical may not be soluble enough to measure this predicted effect. If the effect level exceeds the water solubility by 10X, typically no effects at saturation (NES) are reported
Fish (SW)		ChV	8.31	8	
Mysid (SW)		ChV	1.45	8	

Class Results:	

Organism	Duration	End Point	Concentration (mg/L)	Max Log Kow	Flags
Earthworm	14d	LC50	327.4	6	• Chemical may not be soluble enough to measure this predicted effect. If the effect level exceeds the water solubility by 10X, typically no effects at saturation (NES) are reported

APPENDIX I: EPI Suite™ Modeling Results for IPPD (CAS #101-72-4)

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

CAS Number: 101-72-4
SMILES : N(c(ccc(Nc(cccc1)c1)c2)c2)C(C)C
CHEM : 1,4-Benzenediamine, N-(1-methylethyl)-N -phenyl-
MOL FOR: C15 H18 N2
MOL WT : 226.32
---------- EPI SUMMARY (v4.11) ----------
Physical Property Inputs:
Log Kow (octanol-water): 2.77
Boiling Point (deg C) : 150.00
Melting Point (deg C) : 78.50
Vapor Pressure (mm Hg) : ------
Water Solubility (mg/L): 15
Henry LC (atm-m3/mole) : ------

Log Octanol-Water Partition Coef (SRC):
Log Kow (KOWWIN v1.69 estimate) = 3.28

Boiling Pt, Melting Pt, Vapor Pressure Estimations (MPBPVP v1.43):
Boiling Pt (deg C): 341.75 (Adapted Stein & Brown method)
Melting Pt (deg C): 104.37 (Mean or Weighted MP)
VP(mm Hg,25 deg C): 1.14 (Modified Grain method)
VP (Pa, 25 deg C) : 151 (Modified Grain method)
MP (exp database): 74 deg C
BP (exp database): 161 @ 1 mm Hg deg C
Subcooled liquid VP: 3.67 mm Hg (25 deg C, Mod-Grain method)
: 489 Pa (25 deg C, Mod-Grain method)

Water Solubility Estimate from Log Kow (WSKOW v1.42):
Water Solubility at 25 deg C (mg/L): 153.3
log Kow used: 2.77 (user entered)
melt pt used: 78.50 deg C

Water Sol Estimate from Fragments:
Wat Sol (v1.01 est) = 58.071 mg/L

ECOSAR Class Program (ECOSAR v1.11):
Class(es) found:
Neutral Organics

Henrys Law Constant (25 deg C) [HENRYWIN v3.20]:
Bond Method : 1.44E-009 atm-m3/mole (1.46E-004 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: 2.263E-002 atm-m3/mole (2.293E+003 Pa-m3/mole)
VP: 1.14 mm Hg (source: MPBPVP)
WS: 15 mg/L (source: User-Entered)

Log Octanol-Air Partition Coefficient (25 deg C) [KOAWIN v1.10]:
Log Kow used: 2.77 (user entered)
Log Kaw used: -7.230 (HenryWin est)
Log Koa (KOAWIN v1.10 estimate): 10.000
Log Koa (experimental database): None

Probability of Rapid Biodegradation (BIOWIN v4.10):
Biowin1 (Linear Model) : 0.3004
Biowin2 (Non-Linear Model) : 0.0980
Expert Survey Biodegradation Results:
Biowin3 (Ultimate Survey Model): 2.4511 (weeks-months)
Biowin4 (Primary Survey Model) : 3.3093 (days-weeks)
MITI Biodegradation Probability:
Biowin5 (MITI Linear Model) : -0.0385
Biowin6 (MITI Non-Linear Model): 0.0120
Anaerobic Biodegradation Probability:
Biowin7 (Anaerobic Linear Model): -0.6852
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): 489 Pa (3.67 mm Hg)
Log Koa (Koawin est): 10.000
Kp (particle/gas partition coef. (m3/ug)):
Mackay model : 6.13E-009
Octanol/air (Koa) model: 0.00245
Fraction sorbed to airborne particulates (phi):
Junge-Pankow model : 2.21E-007
Mackay model : 4.9E-007
Octanol/air (Koa) model: 0.164

Atmospheric Oxidation (25 deg C) [AopWin v1.92]:
Hydroxyl Radicals Reaction:
OVERALL OH Rate Constant = 218.3766 E-12 cm3/molecule-sec
Half-Life = 0.049 Days (12-hr day; 1.5E6 OH/cm3)
Half-Life = 0.588 Hrs
Ozone Reaction:
No Ozone Reaction Estimation
Fraction sorbed to airborne particulates (phi):
3.56E-007 (Junge-Pankow, Mackay avg)
0.164 (Koa method)
Note: the sorbed fraction may be resistant to atmospheric oxidation

Soil Adsorption Coefficient (KOCWIN v2.00):
Koc : 4326 L/kg (MCI method)
Log Koc: 3.636 (MCI method)
Koc : 246.8 L/kg (Kow method)
Log Koc: 2.392 (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 = 1.495 (BCF = 31.24 L/kg wet-wt)
Log Biotransformation Half-life (HL) = -0.6063 days (HL = 0.2476 days)
Log BCF Arnot-Gobas method (upper trophic) = 1.580 (BCF = 38)
Log BAF Arnot-Gobas method (upper trophic) = 1.580 (BAF = 38)
log Kow used: 2.77 (user entered)

Volatilization from Water:
Henry LC: 1.44E-009 atm-m3/mole (estimated by Bond SAR Method)
Half-Life from Model River: 6.117E+005 hours (2.549E+004 days)
Half-Life from Model Lake : 6.673E+006 hours (2.78E+005 days)

Removal In Wastewater Treatment:
Total removal: 4.15 percent
Total biodegradation: 0.11 percent
Total sludge adsorption: 4.04 percent
Total to Air: 0.00 percent
(using 10000 hr Bio P,A,S)

Level III Fugacity Model: (MCI Method)

	Mass Amount (percent)	Half-Life (hr)	Emissions (kg/hr)
Air	**0.0086**	**1.18**	**1000**
Water	**12**	**900**	**1000**
Soil	**85.1**	**1.8e+003**	**1000**
Sediment	**2.98**	**8.1e+003**	**0**

Persistence Time: 1.68e+003 hr

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

	Mass Amount (percent)	Half-Life (hr)	Emissions (kg/hr)
Air	0.0086	1.18	1000
Water	12	900	1000
water	(11.9)		
biota	(0.00035)		
suspended sediment	(0.077)		
Soil	85.1	1.8e+003	1000
Sediment	2.98	8.1e+003	0

Persistence Time: 1.68e+003 hr

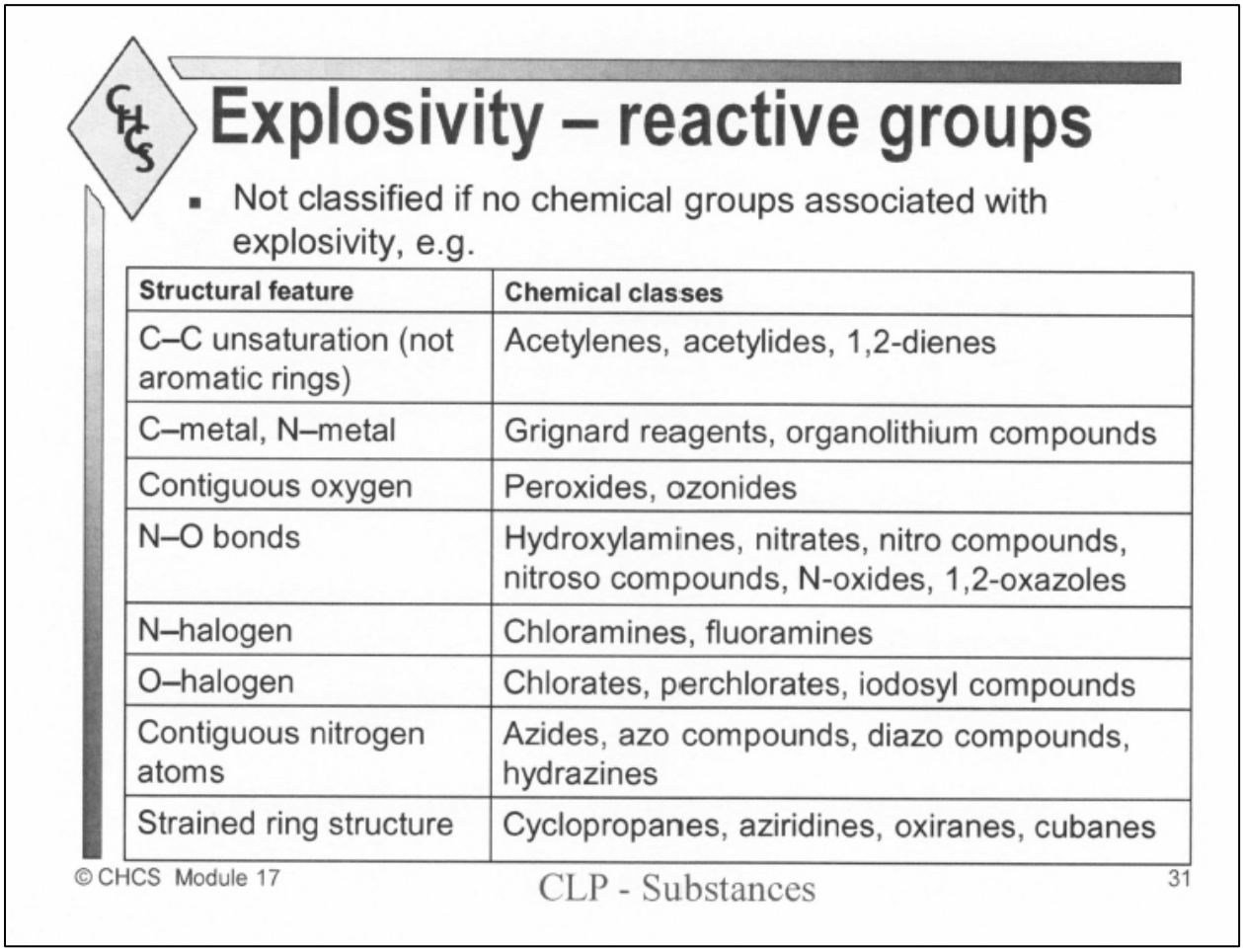
Level III Fugacity Model: (EQC Default)

	Mass Amount (percent)	Half-Life (hr)	Emissions (kg/hr)
Air	0.00943	1.18	1000
Water	15.1	900	1000
water	(15.1)		
biota	(0.000444)		
suspended sediment	(0.00547)		
Soil	84.7	1.8e+003	1000
Sediment	0.21	8.1e+003	0

Persistence Time: 1.54e+003 hr

APPENDIX J: 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

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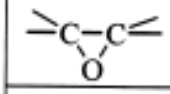
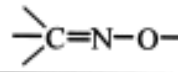
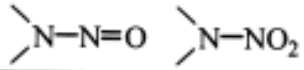
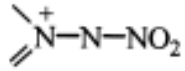
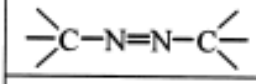
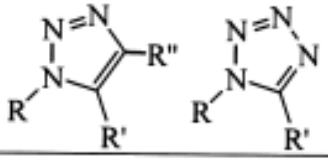
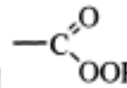
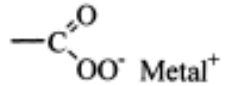
Explosivity – Full List

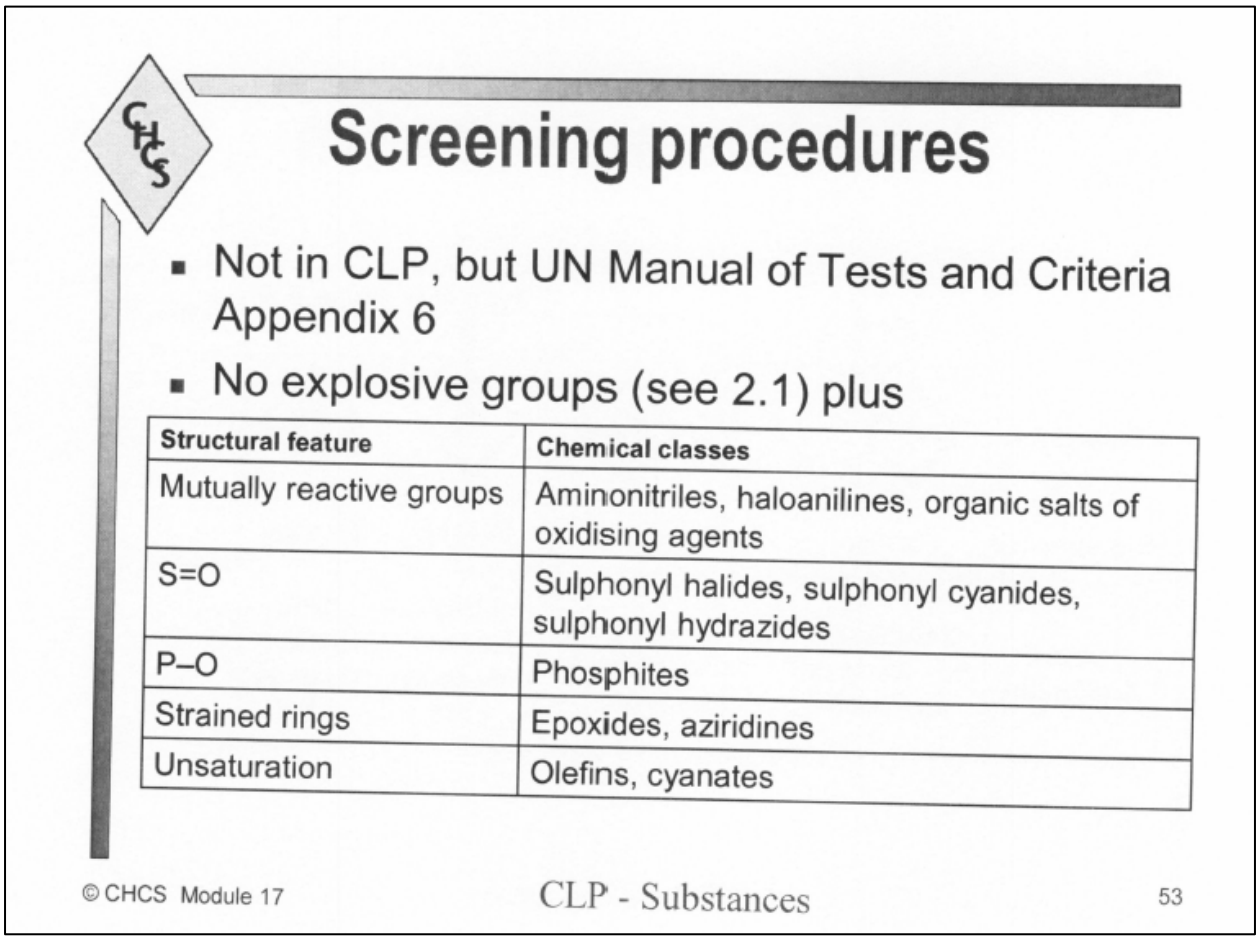
Table R.7.1-28 Chemical groups associated with explosive properties

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



APPENDIX K: Change in Benchmark Score

Table 5 provides a summary of changes to the GreenScreen® Benchmark™ for IPPD. This is a new GreenScreen® assessment.

Table 5: Change in GreenScreen® Benchmark™ for IPPD

Date	GreenScreen® Benchmark™	GreenScreen® Version	Comment
October 14, 2021	BM-1	v. 1.4	New assessment

Licensed GreenScreen® Profilers

IPPD GreenScreen® Evaluation Prepared by:

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