

SCREENING-LEVEL HAZARD CHARACTERIZATION

Hexamethyleneimine (CASRN 111-49-9)

The High Production Volume (HPV) Challenge Program¹ was conceived as a voluntary initiative aimed at developing and making publicly available screening-level health and environmental effects information on chemicals manufactured in or imported into the United States in quantities greater than one million pounds per year. In the Challenge Program, producers and importers of HPV chemicals voluntarily sponsored chemicals; sponsorship entailed the identification and initial assessment of the adequacy of existing toxicity data/information, conducting new testing if adequate data did not exist, and making both new and existing data and information available to the public. Each complete data submission contains data on 18 internationally agreed to “SIDS” (Screening Information Data Set^{1,2}) endpoints that are screening-level indicators of potential hazards (toxicity) for humans or the environment.

The Environmental Protection Agency’s Office of Pollution Prevention and Toxics (OPPT) is evaluating the data submitted in the HPV Challenge Program on approximately 1400 sponsored chemicals by developing hazard characterizations (HCs). These HCs consist of an evaluation of the quality and completeness of the data set provided in the Challenge Program submissions. They are not intended to be definitive statements regarding the possibility of unreasonable risk of injury to health or the environment.

The evaluation is performed according to established EPA guidance^{2,3} and is based primarily on hazard data provided by sponsors; however, in preparing the hazard characterization, EPA considered its own comments and public comments on the original submission as well as the sponsor’s responses to comments and revisions made to the submission. In order to determine whether any new hazard information was developed since the time of the HPV submission, a search of the following databases was made from one year prior to the date of the HPV Challenge submission to the present: (ChemID to locate available data sources including Medline/PubMed, Toxline, HSDB, IRIS, NTP, ATSDR, IARC, EXTOXNET, EPA SRS, etc.), STN/CAS online databases (Registry file for locators, ChemAbs for toxicology data, RTECS, Merck, etc.) and Science Direct. OPPT’s focus on these specific sources is based on their being of high quality, highly relevant to hazard characterization, and publicly available.

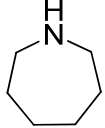
OPPT does not develop HCs for those HPV chemicals which have already been assessed internationally through the HPV program of the Organization for Economic Cooperation and Development (OECD) and for which Screening Initial Data Set (SIDS) Initial Assessment Reports (SIAR) and SIDS Initial Assessment Profiles (SIAP) are available. These documents are presented in an international forum that involves review and endorsement by governmental authorities around the world. OPPT is an active participant in these meetings and accepts these documents as reliable screening-level hazard assessments.

¹ U.S. EPA. High Production Volume (HPV) Challenge Program; <http://www.epa.gov/chemrtk/index.htm>.

² U.S. EPA. HPV Challenge Program – Information Sources; <http://www.epa.gov/chemrtk/pubs/general/guidocs.htm>.

³ U.S. EPA. Risk Assessment Guidelines; <http://cfpub.epa.gov/ncea/raf/rafguid.cfm>.

These hazard characterizations are technical documents intended to inform subsequent decisions and actions by OPPT. Accordingly, the documents are not written with the goal of informing the general public. However, they do provide a vehicle for public access to a concise assessment of the raw technical data on HPV chemicals and provide information previously not readily available to the public.

Chemical Abstract Service Registry Number (CASRN)	111-49-9
Chemical Abstract Index Name	1H-Azepine, hexahydro-
Structural Formula	
Summary	
<p>CASRN 111-49-9 is a clear, colorless liquid with high water solubility and high vapor pressure. It is expected to have moderate mobility in soil. Volatilization is considered negligible since CASRN 111-49-9 will exist as a cation under environmental conditions and cations do not volatilize. The rate of hydrolysis is considered negligible. The rate of atmospheric photooxidation is considered rapid. CASRN 111-49-9 is expected to have moderate persistence (P2) and low bioaccumulation potential (B1).</p> <p>Acute oral and inhalation toxicity of CASRN 111-49-9 in rats is high and moderate, respectively. In a combined oral gavage repeated-dose/reproductive/developmental toxicity screening test in rats, CASRN 111-49-9 showed mucosal thickening in the glandular forestomach at 50 mg/kg-day; the NOAEL for systemic toxicity is 25 mg/kg-day. No reproductive, maternal or developmental effects were observed; the NOAEL for reproductive, maternal and developmental toxicity is 50 mg/kg-day (highest dose tested). CASRN 111-49-9 did not induce gene mutations in bacteria or chromosomal aberrations in mammalian cells, <i>in vitro</i>. CASRN 111-49-9 is corrosive to rabbit skin, irritating to rabbit eyes and is a skin sensitizer in mice.</p> <p>The 96-hr LC₅₀ of CASRN 111-49-9 for fish is > 100 mg/L, the 48-hr EC₅₀ for aquatic invertebrates is >100 mg/L and the 72-hr EC₅₀ for aquatic plants is 43 mg/L (biomass), and 88 mg/L (growth).</p> <p>No data gaps were identified under the HPV Challenge Program.</p>	

The sponsor, E.I. du Pont de Nemours & Company, Inc., submitted a Test Plan and Robust Summaries to EPA for Hexamethyleneimine (CASRN 111-49-9; 9th CI name: 1H-Azepine, hexahydro-) on June 24, 2002. EPA posted the submission on the ChemRTK HPV Challenge website on September 19, 2002

(<http://www.epa.gov/chemrtk/pubs/summaries/hexamthln/c13912tc.htm>). EPA comments on the original submission were posted to the website on January 21, 2003. Public comments were also received and posted to the website. The sponsor submitted updated/revised documents on February 12, 2004, which were posted to the ChemRTK website on April 21, 2004.

Justification for Supporting Chemical

The sponsor submitted data for dibutylamine (CASRN 111-92-2) as a supporting chemical for CASRN 111-49-9, based on similar physical-chemical properties; however, since adequate test data were presented for the sponsored chemical (CASRN 111-49-9), data presented for the proposed analog are not included in this hazard characterization.

1. Chemical Identity

1.1 Identification and Purity

Purity, where indicated in the revised Test Plan and Robust Summaries, ranged from 98.2-99.6%.

1.2 Physical-Chemical Properties

The physical-chemical properties of CASRN 111-49-9 are summarized in Table 1. This chemical is a clear, colorless liquid with high water solubility and high vapor pressure.

Property	Value
CASRN	111-49-9
Molecular Weight	99.18
Physical State	Clear, colorless liquid
Melting Point	-37°C (measured)
Boiling Point	138°C (measured)
Vapor Pressure	8.09 mm Hg at 25°C (measured)
Water Solubility	1.7×10 ⁵ mg/L at 25°C (measured) ²
Dissociation Constant (pK _a)	11.07 (measured) ²
Henry's Law Constant	6.1×10 ⁻⁶ atm·m ³ /mole (measured) ²
Log K _{ow}	-1.60 at pH 1 (estimated) -1.60 at pH 4 (estimated) -1.57 at pH 7 (estimated) -1.37 at pH 8 (estimated) 0.24 at pH 10 (estimated) 1.7 (estimated) ³

¹ INVISTA S.a.r.l. 2008. Revised Test Plan and Robust Summary for Hexamethyleneimine. Available online at <http://www.epa.gov/chemrtk/pubs/summaries/hexamthln/c13912tc.htm> as of June 23, 2010.

² SRC. 2010. The Physical Properties Database (PHYSPROP). SRC: Syracuse, NY. Available online at <http://www.srcinc.com/what-we-do/free-demos.aspx> as of June 23, 2010.

³ U.S. EPA. 2010. Estimation Programs Interface Suite™ for Microsoft® Windows, v4.00. U.S. EPA, Washington, DC, USA. Available online at <http://www.epa.gov/opptintr/exposure/pubs/episuitd.htm> as of June 23, 2010.

2. General Information on Exposure

2.1 Production Volume and Exposure

CASRN 111-49-9 had an aggregated production and/or import volume in the United States between 1 and 10 million pounds during calendar year 2005.

Non-confidential information in the IUR indicated that the industrial processing and uses of the chemical include other basic organic chemical manufacturing as intermediates; pesticide and other agricultural chemical manufacturing as intermediates; and copper, nickel, lead, and zinc mining as processing aid not otherwise listed. Non-confidential commercial and consumer use information was claimed not readily obtainable (NRO).

2.2 Environmental Exposure and Fate

Table 2 lists the environmental fate properties of CASRN 111-49-9.

Table 2. Environmental Fate Characteristics of 1H-Azepine, hexahydro- ¹	
Property	Value
Photodegradation Half-life	1.4 hours (estimated) ²
Hydrolysis Half-life	Stable
Biodegradation	1.19% after 28 days (not readily biodegradable)
Bioaccumulation Factor	BAF = 5.7 (estimated) ²
Log K _{oc}	2.0 (estimated) ²
Fugacity (Level III Model) ²	
Air (%)	0.3
Water (%)	28.1
Soil (%)	71.4
Sediment (%)	0.2
Persistence ³	P2 (moderate)
Bioaccumulation ³	B1 (low)

¹ INVISTA S.a.r.l. 2008. Revised Test Plan and Robust Summary for Hexamethyleneimine. Available online at <http://www.epa.gov/chemrtk/pubs/summaries/hexamthln/c13912tc.htm> as of June 23, 2010.

² U.S. EPA. 2010. Estimation Programs Interface Suite™ for Microsoft® Windows, v4.00. U.S. EPA, Washington, DC, USA. Available online at <http://www.epa.gov/opptintr/exposure/pubs/episuitedi.htm> as of June 23, 2010.

³ Federal Register. 1999. Category for Persistent, Bioaccumulative, and Toxic New Chemical Substances. *Federal Register* 64, Number 213 (November 4, 1999) pp. 60194–60204.

3. Human Health Hazard

A summary of health effects data submitted for SIDS endpoints is provided in Table 3.

Acute Oral Toxicity

(1) Male ChR-CD rats (number of animals not specified) were administered a single dose of aqueous solution of CASRN 111-49-9 (98% purity) via gavage at 0, 300, 450, 670, 1000, 1500, 2250 or 2400 mg/kg and observed for 14 days. Mortality occurred at doses ≥ 1000 mg/kg. An LD₅₀ value was not provided; however, the acute lethal dose (ALD) was 1000 mg/kg.

ALD = 1000 mg/kg

(2) Male CRCD rats (6/dose) were administered a neat single dose of CASRN 111-49-9 (neat; purity not specified) via gavage at 0, 50 or 500 mg/kg. Duration of the observation period was not indicated in the Robust Summaries. All rats treated at 500 mg/kg died on the day of dosing.
50 < LD₅₀ < 500 mg/kg

(3) Sprague-Dawley rats (5/sex/dose) were administered a single dose of CASRN 111-49-9 (purity not specified) via gavage at 3.98, 6.31, 10 or 15.8 mg/kg-day and observed for 14 days. Mortality was 0/5, 2/5, 3/5 and 5/5 at 3.98, 6.31, 10 and 15.8 mg/kg-day, respectively.
LD₅₀ = 9.6 mg/kg-day

(4) Male and female Sprague-Dawley rats (5 combined sexes/dose) received CASRN 111-49-9 (purity not specified) as a single gavage administration at 7.94, 12.6, 20.0 or 31.6 mg/kg and were observed for 14 days. Mortalities (8/20) generally occurred within eight days of exposure. OTS0534842
LD₅₀ = 20.7 mg/kg

Acute Inhalation Toxicity

Male ChR-CD rats received CASRN 111-49-9 (98% purity) via vapor inhalation at 0.52, 1.32, 1.95, 2.45, 2.77 or 3.12 mg/L for 4 hours and were observed for 14 days. Six rats were assigned to all but the lowest treatment group (0.52 mg/L), which had ten rats. Mortality was 0/10, 0/6, 0/6, 1/6, 3/6 and 6/6 at 0.52, 1.32, 1.95, 2.45, 2.77 and 3.12 mg/L, respectively.
LC₅₀ = 2.45 mg/L

Repeated-Dose Toxicity

In a combined repeated-dose/reproductive/developmental toxicity screening test, Wistar rats (5/sex/dose) received CASRN 111-49-9 (aqueous; 99.6% purity) via gavage at 0, 10, 25 or 50 mg/kg-day. Dose selection for the main study was based on results obtained during an initial range finding study, in which rats (3/sex/dose) were gavaged with CASRN 111-49-9 at 50, 200 or 400/500 mg/kg-day. Severe toxicity (clinical signs, decreased body weight, decreased liver and kidney weights, microscopic findings in the stomach and small intestine) was observed at concentrations > 50 mg/kg-day. In the main study, male rats were dosed at 0, 10, 25 or 50 mg/kg-day for six weeks (14 days prior to mating through study termination on day 29) and females were dosed at these concentrations for seven weeks (14 days prior to mating, throughout gestation and the third day of lactation). Five rats/sex/dose were subjected to a Functional Observation Battery (startle, reflex and motor activity), as well as hematology and blood chemistry tests. At necropsy, a thorough examination was made of the cranial, thoracic and abdominal tissues and organs (adrenals, brain, epididymides, heart, kidney, liver, spleen, testes and thymus). Microscopic examinations were completed on selected tissues (and all reproductive organs). Macroscopic examination revealed thickening of the glandular/forestomach mucosal tissue in rats treated at 50 mg/kg-day; however, microscopic examination revealed no correlation for these effects. No treatment-related effects were observed on mortality, clinical signs, body weight, food consumption, functional observations, clinical laboratory investigations, organ weights, or microscopic examinations.
LOAEL = 50 mg/kg-day (based on mucosal thickening in the glandular/forestomach region)
NOAEL = 25 mg/kg-day

Reproductive/Developmental Toxicity

In the combined repeated-dose/reproductive/developmental toxicity screening test described above, male and female Wistar rats (5/sex/dose) received CASRN 111-49-9 (aqueous; 99.6% purity) via gavage administration at 0, 10, 25, or 50 mg/kg-day. Males were dosed for six weeks (14 days prior to mating through study termination on day 29) and females were dosed for seven weeks (14 days prior to mating, throughout gestation and the third day of lactation). No reproductive effects or maternal/developmental toxicity were observed up to and including the highest dose tested (50 mg/kg-day). There were no treatment-related effects on: reproductive performance (mating success, conception), organ weights (testes, epididymides), fertility indices (number of corpora lutea, gestation length), offspring survival or pup body weight.

NOAEL (reproductive toxicity) = 50 mg/kg-day (highest dose tested)

NOAEL (maternal toxicity) = 50 mg/kg-day (highest dose tested)

NOAEL (developmental) = 50 mg/kg-day (highest dose tested)

Genetic Toxicity - Gene mutation

In vitro

(1) In a reverse mutation assay, *Salmonella typhimurium* strains TA97a, TA98, TA100, TA1535 and *Escherichia coli* strain WP2 *uvrA* (pKM101) were exposed to CASRN 111-49-9 (99.5% purity) at 0, 10, 50, 100, 500, 1000, 2500 or 5000 µg/plate, in the presence or absence of metabolic activation. Positive and negative controls were tested concurrently and responded accordingly. No precipitation was observed in this assay. Cytotoxicity occurred in all test strains at 1000 µg/plate in the absence of metabolic activation and at 2500 µg/plate in all strains (except TA 1535) in the presence of metabolic activation. CASRN 111-49-9 did not induce a positive response (*i.e.*, an increase in the frequency of revertant colonies that is at least two times greater than the vehicle control) in the presence or absence of metabolic activation.

CASRN 111-49-9 was not mutagenic in this assay.

(2) In a National Toxicology Program (NTP) reverse mutation assay, in *S. typhimurium* strains TA98 and TA100 were exposed to CASRN 111-49-9 in dimethyl sulfoxide at 0, 33,100, 333, 1000, 1666, 3333 or 6666 µg/plate with and without metabolic activation. *S. typhimurium* strains TA1535 and TA97 were exposed to CASRN 111-49-9 dimethyl sulfoxide at 0, 33,100, 333, 1000, 1666, 3333 µg/plate with and without metabolic activation. Positive and negative controls were tested concurrently and responded appropriately. Precipitation occurred at concentrations \geq 1666 µg/plate. Cytotoxicity was observed only at the highest concentration (6666 µg/plate) in *S. typhimurium* strains TA 98 and TA100 in the presence of metabolic activation. No increase in the frequency of revertant colonies was recorded for any of the bacterial strains at any concentration tested with or without metabolic activation.

CASRN 111-49-9 was not mutagenic in this assay.

Genetic Toxicity - Chromosomal aberrations

In vitro

Human lymphocytes were exposed to CASRN 111-49-9 (99.5% purity) at 100, 333, 992 µg/mL in the presence or absence of metabolic activation. Cultures were treated for 3 hours and fixed 24 hours after the last dose. Positive and negative controls were tested concurrently and

responded accordingly. No cytotoxicity or precipitation was observed in this assay. No increase in the proportion of cells with chromosomal aberrations was observed when compared to vehicle controls. In a confirmatory test, cultures were exposed at 100, 200, 300, 350 or 400 µg/mL for 24 hours in the presence or absence of metabolic activation and fixed 24 hours after the last dose. A second group of cultures was exposed at 100, 200, 300, 350 or 400 µg/mL for 48 hours in the presence or absence of metabolic activation and fixed 48 hours after the last dose. No significant increase in the number of cells with chromosomal aberrations was observed with either exposure regimen. Positive and negative controls were tested concurrently and responded accordingly. **CASRN 111-49-9 did not induce chromosomal aberrations in this assay.**

Additional Information

Skin Irritation

New Zealand White rabbits (number/sex not specified) were administered 0.5 mL undiluted CASRN 111-49-9 (purity not specified) via topical application to areas of abraded and intact skin under occluded conditions for 4 hours (concentration not specified) and observed for 72 hours. CASRN 111-49-9 was severely irritating and corrosive, with a primary irritation score of 8.0/8.0. **CASRN 111-49-9 was corrosive to rabbit skin.**

Eye Irritation

Undiluted CASRN 111-49-9 (0.1 mL; purity not specified) was applied to the corneal surface of six male New Zealand rabbits. The eyes of three rabbits were rinsed for 60 seconds approximately 20-30 seconds after dosing; the eyes of the other three rabbits were not rinsed. Rabbits were observed for 24 hours and evaluated according to the Draize procedure. The mean score in un-rinsed eyes was 80 for the cornea and 19 for the conjunctivae. All rabbits exhibited blanching of the nictitating membrane and eschar formation on eyelids; the iris could not be scored due to opacity. The study was terminated after a 24-hour observation period. **CASRN 111-49-9 was severely irritating to rabbit eyes.**

Skin Sensitization

In a mouse ear swelling test, female CF-1 mice (number not specified) were administered 100 µL CASRN 111-49-9 (98% purity) in acetone via topical application to shaved stomach skin following two intradermal injections of Freund's complete adjuvant emulsion. Topical application of test article was repeated for four consecutive days. One week after the final application, 20 µL of CASRN 111-49-9 (in acetone) was applied to the right ear and 20 µL of the solvent control was applied to the left ear of each animal. Ear thickness was measured 24 and 48 hours after application. Sensitization potential was also evaluated using an occluded patch induction method. In this test, CASRN 111-49-9 (1% in propylene glycol) was applied to shaved, tape-stripped skin under occlusive conditions every other day for 6 days. The criteria used to determine sensitization potential were not described in the Robust Summaries. No evidence of sensitization was observed using the occluded patch induction; however, CASRN 111-49-9 produced a positive response in 40% of mice tested using the mouse ear swelling test. **CASRN 111-49-9 was sensitizing in the mouse ear swelling test, but not the patch induction test.**

Conclusion: Acute oral and inhalation toxicity of CASRN 111-49-9 in rats is high and moderate, respectively. In a combined oral gavage repeated-dose/reproductive/developmental toxicity screening test in rats, CASRN 111-49-9 showed mucosal thickening in the glandular forestomach at 50 mg/kg-day; the NOAEL for systemic toxicity is 25 mg/kg-day. No reproductive, maternal or developmental effects were observed; the NOAEL for reproductive, maternal and developmental toxicity is 50 mg/kg-day (highest dose tested). CASRN 111-49-9 did not induce gene mutations in bacteria or chromosomal aberrations in mammalian cells, *in vitro*. CASRN 111-49-9 is corrosive to rabbit skin, irritating to rabbit eyes and is a skin sensitizer in mice.

Table 3. Summary of the Screening Information Data Set as Submitted under the U.S. HPV Challenge Program - Human Health Data	
Endpoints	SPONSORED CHEMICAL Hexamethyleneimine (111-49-9)
Acute Toxicity Oral LD₅₀ (mg/kg)	9.6
Acute Inhalation Toxicity LC₅₀ (mg/L)	2.45
Repeated-Dose Toxicity NOAEL/LOAEL Oral (mg/kg-day)	NOAEL = 25 LOAEL = 50
Reproductive/ Developmental Toxicity NOAEL/LOAEL Oral (mg/kg-day)	NOAEL = 50 (highest dose tested)
Maternal toxicity	NOAEL = 50 (highest dose tested)
Reproductive/Developmental toxicity	NOAEL = 50 (highest dose tested)
Genetic Toxicity - Gene Mutation <i>In vitro</i>	Negative
Genetic Toxicity - Chromosome Aberrations <i>In vitro</i>	Negative
Additional Information Skin irritation Eye irritation Skin Sensitization	Corrosive Severely irritating Positive

Measured data in bold

4. Hazard to the Environment

A summary of aquatic toxicity data submitted for SIDS endpoints is provided in Table 4.

Acute Toxicity to Fish

Rainbow trout (*Oncorhynchus mykiss*) were exposed to CASRN 111-49-9 at a concentration of 100mg/L (Limit Test) for 96 hours under flow-through conditions. No mortalities were reported.
96-hr LC₅₀ > 100 mg/L

Acute Toxicity to Aquatic Invertebrates

Water fleas (*Daphnia magna*) were exposed to CASRN 111-49-9 at concentrations of 10, 18, 32, 56, and 100 mg/L for 48 hours under flow-through conditions. Treatment related immobilization was observed at 56 (25 %) and 100 (20 %) mg/L.
48-hr EC₅₀ > 100 mg/L

Toxicity to Aquatic Plants

Green algae (*Pseudokirchneriella subcapitata*, formerly known as *Selenastrum capricornutum*) were exposed to CASRN 111-49-9 for 72-hours. Test concentrations were not provided.
72-hr EC₅₀ = 43 mg/L (biomass)
72-hr EC₅₀ = 88 mg/L (growth)

Conclusion: The 96-hr LC₅₀ of CASRN 111-49-9 for fish is > 100 mg/L. The 48-hr EC₅₀ of CASRN 111-49-9 for aquatic invertebrates is >100 mg/L. The 72-hr EC₅₀ of CASRN 111-49-9 for aquatic plants is 43 mg/L (biomass) and 88 mg/L (growth).

Table 4. Summary Table of the Screening Information Data Set as submitted under the U.S. HPV Challenge Program Summary of Environmental Effects – Aquatic Toxicity Data	
Endpoints	SPONSORED CHEMICAL Hexamethyleneimine (111-49-9)
Fish 96-h LC ₅₀ (mg/L)	>100
Aquatic Invertebrates 48-h EC ₅₀ (mg/L)	>100
Aquatic Plants 72-h EC ₅₀ (mg/L) (biomass)	43
(growth)	88

bold = measured data (i.e., derived from testing)