

SCREENING-LEVEL HAZARD CHARACTERIZATION

Nitroglycerin (CASRN 55-63-0)

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.

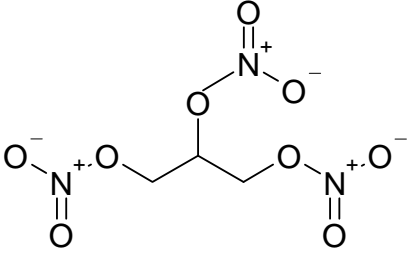
OPPT does not develop HCs for those HPV chemicals which have already been assessed internationally through the HPV program of the Organization for Economic Cooperation and Development (OECD) and for which Screening Initial Data Set (SIDS) Initial Assessment Reports (SIAR) and SIDS Initial Assessment 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.

<p>Chemical Abstract Service Registry Number (CASRN)</p>	<p>55-63-0</p>
<p>Chemical Abstract Index Name</p>	<p>1,2,3-Propanetriol, trinitrate</p>
<p>Structural Formula</p>	
<p style="text-align: center;">Summary</p> <p>CASRN 55-63-0 is also a prescription drug. For medicinal use, a medical provider should be consulted. Additional information is available on the U.S. Food and Drug Administration (FDA) website (http://www.fda.gov/Drugs/default.htm).</p> <p>CASRN 55-63-0 is an explosive hazard. It is a liquid with high water solubility and moderate vapor pressure. It is expected to have moderate mobility in soil. Volatilization is considered low based on the Henry's Law constant of this compound. The rate of hydrolysis is considered negligible. The rate of atmospheric photooxidation is considered slow. CASRN 55-63-0 is expected to have low persistence (P1) and low bioaccumulation potential (B1).</p> <p>Acute oral toxicity of CASRN 55-63-0 to rats and mice and acute dermal toxicity to rabbits is low. Repeated oral exposure of rats to CASRN 55-63-0 for 2 years resulted in histopathologic findings as well as correlative liver weight findings. In females, histopathological changes in the liver (including liver cancer) were observed at 38.1 mg/kg/day and above. Histopathological changes among females were also noted in the spleen and mammary glands at this dose. In males, other histopathological changes in the liver, as well as histopathological findings of the kidney, were observed at doses as low as 3.04 mg/kg/day. The NOAEL for systemic toxicity was 3.99 mg/kg/day for females, and a NOAEL for males could not be established. Repeated oral exposures of CASRN 55-63-0 to mice for 2 years showed decreased survival among females at 9.72 mg/kg/day and above, and methemoglobinemia among males at 114.6 mg/kg/day, and above; the NOAEL for systemic toxicity could not be established for females, and was 11.1 mg/kg/day for males. Oral administration of CASRN 55-63-0 to dogs for 1 year did not show toxicity; the NOAEL for systemic toxicity was 25 mg/kg/day (highest dose tested). In a three-generation dietary reproductive toxicity study in rats, decreased food consumption was observed (at an unspecified dose), as well as signs of reproductive toxicity including severe aspermatogenesis, severe infertility and decreased testicular size, and decreased litter size and birth weights at ~408 mg/kg/day. The NOAEL for systemic toxicity of the females was ~452 mg/kg/day (highest dose tested); the NOAEL for reproductive toxicity was ~39 mg/kg/day. In a prenatal developmental toxicity study, CASRN 55-63-0 was orally administered to pregnant rats during gestation days 6-15. Signs of maternal toxicity included significantly reduced body</p>	

weight and increased liver weights at ~60 mg/kg/day. Developmental toxicity included increased incidences of diaphragmatic hernias and increased incidences of skeletal anomalies in offspring at ~60 mg/kg/day. The NOAEL for maternal and developmental toxicity was ~ 6 mg/kg/day. CASRN 55-63-0 was mutagenic in several bacterial reverse mutation assays and induced chromosomal aberrations in mammalian cells *in vitro*. CASRN 55-63-0 is non-irritating to rabbit eyes, a mild irritant to rabbit skin and a dermal sensitizer in guinea pigs.

The 96-hour LC₅₀ of CASRN 55-63-0 to fish is 1.90 mg/L, the 48-hour EC₅₀ for aquatic invertebrates is 17.83 mg/L, and the 96-hour EC₅₀ for aquatic plants is 1.15 mg/L (biomass).

No data gaps were identified under the HPV Challenge Program.

The sponsor, the Synthetic Organic Chemicals Manufacturers Association's U.S. Nitroglycerin Producers Consortium, submitted a Test Plan and Robust Summaries to EPA for nitroglycerin (CASRN 55-63-0) on September 25, 2002. EPA posted the submission on the ChemRTK HPV Challenge website on October 29, 2002

(<http://www.epa.gov/chemrtk/pubs/summaries/nitroglnc13997tc.htm>). EPA comments on the original submission were posted to the website on February 25, 2003. The sponsor submitted updated/revised documents on April 25, 2003 and October 19, 2004, which were posted to the ChemRTK website on June 3, 2003 and November 15, 2004, respectively. Public comments were also received and posted to the website.

1. Chemical Identity

1.1 Identification and Purity

Purity, when reported, was $\leq 100\%$.

1.2 Physical-Chemical Properties

The physical-chemical properties of CASRN 55-63-0 are summarized in Table 1. CASRN 55-63-0 is a liquid with high water solubility and moderate vapor pressure.

Table 1. Physical-Chemical Properties of CASRN 55-63-0¹	
Property	Value
CASRN	55-63-0
Molecular Weight	227.09
Physical State	Liquid
Melting Point	2.0°C (triclinic structure, measured) 13.5°C (rhombic structure, measured)
Boiling Point	125°C at 2 mm Hg (measured) 180°C at 50 mm Hg (measured) Decomposes upon heating at atmospheric pressure
Vapor Pressure	2.7×10^{-4} mm Hg at 20°C (measured)
Water Solubility	1.38×10^3 – 2.0×10^3 mg/L at 20°C (measured) 1.25×10^3 mg/L at 25°C (measured)
Dissociation Constant (pK _a)	Not applicable
Henry's Law Constant	8.7×10^{-8} atm·m ³ /mole (estimated) ²
Log K _{ow}	2.04 (measured)

¹U.S. Nitroglycerin Producers Consortium. 2004. Revised Test Plan and Robust Summary for Nitroglycerin. Available online at <http://www.epa.gov/chemrtk/pubs/summaries/nitroglnc13997tc.htm> as of June 25, 2010.

²U.S. EPA. 2010. Estimation Programs Interface Suite™ for Microsoft® Windows, v4.00. U.S. Environmental Protection Agency, Washington, DC, USA. Available online at <http://www.epa.gov/opptintr/exposure/pubs/episuiteld.htm> as of June 25, 2010.

2. General Information on Exposure

2.1 Production Volume and Use Pattern

CASRN 55-63-0 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 explosives manufacturing as “other.” Non-confidential commercial and consumer uses of this chemical include “other.”

Precautionary Note: In addition to CASRN 55-63-0’s described uses, it is sold as a prescription drug. This hazard characterization is not intended to address CASRN 55-63-0’s medical uses. Under no circumstances should information contained herein be used in lieu of information obtained from a medical provider. Additional information is available on the U.S. Food and Drug Administration (FDA) website (<http://www.fda.gov/Drugs/default.htm>).

2.2 Environmental Exposure and Fate

The environmental fate properties of CASRN 55-63-0 are provided in Table 2. CASRN 55-63-0 is expected to have moderate mobility in soil. In a modified Sturm test (OECD 301B), CASRN 55-63-0 was not readily biodegradable, achieving 0% of the theoretical CO₂ production after 29 days. However, in a modified shake flask method using microorganisms obtained from fresh activated sludge acclimated to the test substance, CASRN 55-63-0 was found to biodegrade, achieving 53.6% degradation in 5 days. In a continuous-culture multi-stage (chemostat) experiment, biodegradability of CASRN 55-63-0 after 5 days was 92.2%. These results indicate that while CASRN 55-63-0 is not readily biodegradable, it is not likely to be highly persistent in the environment. The rate of volatilization is considered low based on the Henry’s Law constant of this substance. The rate of hydrolysis is considered negligible. CASRN 55-63-0 is expected to have low persistence (P1) and low bioaccumulation potential (B1).

Table 2. Environmental Fate Characteristics of CASRN 55-63-0¹	
Property	Value
Photodegradation Half-life	9.7 days (estimated) ²
Hydrolysis Half-life	37 days (at pH 9 and 25°C) 134 days (at pH 5 and 80°C) 'years' (at pH 5 and 25°C) 5 days (at 1M HCl and 37°C) 10 days (at 1M HCl and 25°C) >100 years (at pH 3 and 37°C)
Biodegradation	0.0% after 29 days (not readily biodegradable) 53.6% after 5 days (shake flask test using activated sludge inoculum) 92.2% after 5 days (continuous-culture multi-stage experiment)
Bioaccumulation Factor	BAF = 4.5 (estimated) ²
Log K _{oc}	2.1 (estimated) ²
Fugacity (Level III Model) ²	
Air (%)	0.6
Water (%)	18.1
Soil (%)	81.2
Sediment (%)	0.1
Persistence ³	P1 (low)
Bioaccumulation ³	B1 (low)

¹U.S. Nitroglycerin Producers Consortium. 2004. Revised Test Plan and Robust Summary for Nitroglycerin. Available online at <http://www.epa.gov/chemrtk/pubs/summaries/nitroglnc13997tc.htm> as of June 25, 2010.

²U.S. EPA. 2010. Estimation Programs Interface Suite™ for Microsoft® Windows, v4.00. U.S. Environmental Protection Agency, Washington, DC, USA. Available online at <http://www.epa.gov/opptintr/exposure/pubs/episuitedi.htm> as of June 25, 2010.

³Federal 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

Acute Oral Toxicity

Wistar albino rats (5/sex/dose) were administered undiluted CASRN 55-63-0 via gavage at 159, 478, 1593 or 4779 mg/kg and observed for up for 14 days. Mortalities occurred at the three highest doses: 4779 mg/kg (5/5 males and 5/5 females on day-0); 1593 mg/kg (5/5 males and 4/5 females on day-0, and 1/5 females on day-1); and 478 mg/kg (2/5 males on day-0).

LD₅₀ = 685 mg/kg

Acute Dermal Toxicity

Wistar albino rats (10/sex/dose) were administered CASRN 55-63-0 via the dermal route at 9560 mg/kg to clipped, intact skin for 24 hours and were observed for 14 days. Mortality was not observed at any dose level.

LD₅₀ > 9560 mg/kg

Repeated-Dose Toxicity

(1) Albino rats (38/sex/dose) were administered CASRN 55-63-0 in the diet at 0, 0.01, 0.1 or 1.0% (~ 0, 3.04, 31.5 and 363 mg/kg/day for males and 0, 3.99, 38.1 and 434 mg/kg/day for females) for 2 years. No treatment-related effects on mortality were observed. The low-dose males, and mid- and high-dose animals of both sexes, had increased areas/foci of hepatocellular alteration. Dose-responsive increases in incidences of hepatocellular carcinoma were observed in both males and females in the mid- and high-dose groups, and incidences of areas/foci of hepatocellular alteration were approximately twice as high in males (all dose groups) compared to their corresponding controls. Incidences of areas/foci of hepatocellular alteration among females were approximately three times greater in the mid- and high-dose groups than the control group. Additionally, high-dose males and females had increased incidences of other liver histopathologic findings, including cholangiofibrosis, cystic bile duct hyperplasia, adenomatoid bile duct hyperplasia, and neoplastic nodules. Increases in liver weights (absolute and relative) in the male and female high-dose groups were consistent with the liver histopathology findings at that dose, the only dose for which findings were presented. Increased incidences of spleen hyperpigmentation were observed in both males and females of the mid- and high-dose groups. Testicular interstitial cell tumor incidences were increased in the mid-dose and high-dose males, and incidences of kidney epithelial hyperpigmentation was increased in males of all dose groups and females in the high-dose group. An increased incidence of mammary fibromas was observed in females of the mid-dose group. Other mammary histopathologic findings appeared to be normal (“background”) occurrences, in that they were comparable in type and incidence to those of controls. Percent methemoglobin was elevated and was statistically significant for high-dose males and females within the first 12 months and at 18 months.

LOAEL (males) ~ 3.04mg/kg/day, the lowest dose tested (based on histopathological findings in liver and kidney)

NOAEL (males) = Not established.

LOAEL (females) ~ 38.1 mg/kg/day (based on histopathological findings in the liver, spleen and mammary glands, including hepatocellular carcinoma)

NOAEL (females) ~ 3.99 mg/kg/day

(2) Albino Swiss mice (58/sex/dose) were administered CASRN 55-63-0 in the diet at 0, 0.01, 0.1 or 1.0% (~ 0, 11.1, 114.6 and 1022 mg/kg/day for males and 0, 9.72, 96.4 and 1058 mg/kg/day for females) for 2 years. Survival among females at 2 years (n=18), but not 2 years and 1 month (n=4) appeared decreased and related to dose, including the low dose. This discrepancy may be due to small numbers of animals evaluated at the longer time point. High dose females had lower body weights (statistically significant) than their corresponding controls at 12 and 24 months. Methemoglobin levels were elevated in the mid-dose males at 12 months, and in females at the high-dose, also at 12 months (statistically significant). Quantitative information for methemoglobin levels was not provided. Sequelae of methemoglobinemia (Heinz bodies, anemia, and/or pigment deposits) were elevated in the mid-dose and high-dose males and females at 1 year. This change was statistically significant for the high-dose groups, but not the mid-dose groups. During the first 12-13 months, the greatest incidence of pigment deposits (4 of 4 males that were examined at both 12 and 13 months) was observed in the liver of males in the high-dose group. The spleen was the primary organ observed with this deposit during the 2-year treatment period in the high-dose groups (5/5 males and 5/7 females). At 2

years, this elevation was observed in the mid-dose and high-dose females, and was statistically significant only in the high-dose females. Granular, brown pigment, which was not considered hemosiderin, was observed in various organs and discrete cells, as was excessive hepatocellular dysplasia. Among unscheduled deaths, hemosiderin was also elevated in the liver of male and female low dose groups. The frequencies and severity of hepatocellular dysplasia were stated to be comparable to those of the control group's, although no quantitative data were provided. Based on treatment related hematologic changes (methemoglobin elevation), the dose-related elevation in hemosiderin (a pigment that forms after hemorrhage or iron overload) may be an effect related to the methemoglobin elevation.

LOAEL (males) = 114.6 mg/kg/day (methemoglobinemia)

NOAEL (males) = 11.1 mg/kg/day

LOAEL (females) = 9.72 mg/kg/day (based on decreased survival)

NOAEL (females) = Not established

(3) Male and female beagle dogs (unspecified numbers), each of which were orally administered capsules containing CASRN 55-63-0 adsorbed on lactose via gelatin at 0, 1, 5 or 25 mg/kg/day for 1 year. Percent methemoglobin increased during months 6 and 9 in all groups, including that of the male and female control groups, and values at 12 months were suggestive of returning to baseline levels. However, increases were not suggestive of a dose-response effect, and values were comparable to those of controls. No historical methemoglobin data were provided. No meaningful treatment effects, including possible microscopic effects on tissue, were reported.

NOAEL (males and females) = 25 mg/kg/day (highest dose tested)

Reproductive Toxicity

In a three-generation study, albino rats (10 males and 20 females/dose, the F₀ group) were administered CASRN 55-63-0 in the diet at 0, 0.01, 0.1 or 1.0% (approximately 0, 3.6, 39 and 408 mg/kg/day for males and 0, 5.0, 46.0 and 452 mg/kg/day for females). Females were dosed during pregnancy and between matings, and males were dosed until successful delivery of each "b" generation. All litter parameters (litter size, liver-born index, birth weight, viability index, lactation index and weight at weaning) were reduced in F_{1a} offspring at the high-dose level. The offspring were discarded and the F₀ group remated to produce an F_{1b} generation. To produce the F₂ generation, 20-24 normal offspring (F_{1b}) were mated. The same procedure was followed with the F₂ group. Severe aspermatogenesis with resulting severe infertility was observed in the high-dose F_{2a} generation of males. The testes of these animals were ~ 25% of normal size and microscopic evaluation mild to moderate increases in the amount of interstitial tissues in the testes. Several litter parameters were reduced in both the high-dose F_{1b}, and F_{2a} litters, including lower birth and weaning weights. Reduced food intake (~65% that of corresponding controls) was also observed in F_{1b} dams (dose level not reported), and their gestational product (litter size x litter weight) was ~ 62% that of the corresponding control dams.

NOAEL (systemic toxicity, females) ~452 mg/kg/day (highest dose tested)

LOAEL (reproductive toxicity) ~ 408 mg/kg/day (based on severe aspermatogenesis, decreased testicular size)

NOAEL (reproductive toxicity) ~ 39 mg/kg/day

Developmental Toxicity

Pregnant albino rats (21/dose) were administered CASRN 55-63-0 in the diet at 0, 0.01, 0.1 or 1.0% w/w (~ 0, 0.06, 6 or 60 mg/kg/day, respectively) during gestation days 6-15. Late resorptions were increased in all treatment groups, but these were neither statistically significant nor dose-responsive. Significantly reduced body weights and increased liver weights were observed in high-dose dams. Diaphragmatic hernias occurred in 4/19 of the high-dose litters and were believed to be treatment related; however, their incidence was not significant. The incidences of unossified and incompletely ossified hyoid bones were significantly increased in the litters of the high-dose group.

LOAEL (maternal toxicity) ~ 60 mg/kg/day (based on decreased maternal body weight)

LOAEL (developmental toxicity) ~ 60 mg/kg/day (based on increased skeletal anomalies and incidence of diaphragmatic hernias)

NOAEL (maternal and developmental toxicity) ~ 6 mg/kg/day

Genetic Toxicity – Gene Mutation

In vitro

(1) *S. typhimurium* strains TA98, TA100, TA1535, TA1537 and TA1538 were incubated with CASRN 55-63-0 as a 10% mixture in lactose in dimethyl sulfoxide (DMSO) at concentrations of 10, 100, 300 or 1000 µg/plate in the presence and absence of metabolic activation. Significant increases in revertant ratios (test:control) were reported for the 1000 µg/plate dose in TA 1537 (without metabolic activation) and for the 1000 µg/plate dose in TA 1535 (with metabolic activation). Positive controls were tested, but their responses were not provided in the Robust Summary's data source.

CASRN 55-63-0 was mutagenic in this assay.

(2) *S. typhimurium* strains TA98, TA100, TA1535, TA1537 and TA1538 were incubated with CASRN 55-63-0 in DMSO at concentrations of 5, 16.6, 50, 166 or 500 µg/plate. CASRN 55-63-0 was also tested in a repeat assay with TA1535 with metabolic activation, and TA100 with and without metabolic activation, at 100, 200, 333, 500, 750, and 1000 µg/plate. CASRN 55-63-0 caused increases in revertants in both assays with TA1535. In the first assay positive findings in TA1535 were observed at 166 and 500 µg/plate, with and without metabolic activation. In the repeat assay with TA1535, and with metabolic activation, CASRN 55-63-0 elicited positive findings at 750 and 1000 µg/plate. Positive controls were tested concurrently in both assays, and responded appropriately.

CASRN 55-63-0 was mutagenic in this assay.

(3) *S. typhimurium* strains TA98, TA100, TA1535, TA1537 and TA1538 were incubated with CASRN 55-63-0 in ethanol at concentrations of 15, 50, 150, 500 or 1500 µg/plate in the presence and absence of metabolic activation. In a repeat assay, TA1535 was incubated with CASRN 55-63-0 at 50, 200, 500, 1500, or 2000 µg/plate, with metabolic activation. Cytotoxicity was observed at the highest dose. With metabolic activation, concentrations of 500, 1500 and 2000 µg/plate resulted in positive responses. Positive controls were tested concurrently and responded appropriately.

CASRN 55-63-0 was mutagenic in this assay.

Genetic Toxicity – Chromosomal Aberrations

In vitro

CHO cells were exposed to CASRN 55-63-0 at several concentrations, with or without metabolic activation (S-9). Concentrations, from 500-2270 µg/mL, although the robust summary was unclear on which concentrations were used for which test conditions (i.e., with or without metabolic activation, and the length of the treatment time (4 or 20 hours), or the time for recovery (0 or 16 hours). Positive controls were tested concurrently, but their responses were not provided. CASRN 55-63-0 induced aberrations at concentrations of 567.5 µg/mL in the absence of metabolic activation and 500 µg/mL in the presence of metabolic activation. The concentrations causing increases in chromosomal aberrations were also cytotoxic to the cells, with a 45 -52% growth inhibition of cells.

CASRN 55-63-0 induced chromosomal aberrations in this assay.

Additional Information

Skin Irritation

Undiluted CASRN 55-63-0 was applied to one intact and one abraded site on six female rabbits. Sites were occluded for 24 hours and assessed for up to 7 days.

CASRN 55-63-0 was a mild irritant to rabbit skin in this study.

Eye Irritation

Undiluted CASRN 55-63-0 was instilled in the conjunctival sac of the right eye of six male rabbits. Three eyes were washed at 1 minute post-treatment. Irritation was scored at 1, 24 and 72 hours and 7 days after administration.

CASRN 55-63-0 was not an irritant to rabbit eyes in this study.

Sensitization

In a guinea pig maximization test, CASRN 55-63-0 was applied to clipped, intact skin of 10 animals (sex not specified). The covering of the application sites, the number of application sites, the schedule of applications, and the application site grading were consistent with the Kligman Maximization reference. Ten controls received only skin applications of the vehicle: 65%peanut oil and 35 % lactose. Forty percent of the guinea pigs demonstrated positive sensitization responses at challenge.

CASRN 55-63-0 was a dermal sensitizer in guinea pigs in this study.

Table 3. Summary of the Screening Information Data Set under the U.S. HPV Challenge Program – Human Health Data	
Endpoints	SPONSORED CHEMICAL Nitroglycerin (55-63-0)
Acute Oral Toxicity LD₅₀ (mg/kg)	685
Acute Dermal Toxicity LD₅₀ (mg/kg)	> 9560
Repeated-Dose Toxicity NOAEL/LOAEL Oral (mg/kg/day)	(rat) NOAEL = not established (2-yr) LOAEL ~ 3.99 (2-yr) (mouse) NOAEL = not established (2-yr) LOAEL ~ 9.72 (2-yr) (dog) NOAEL = 25 mg/kg/day (highest dose tested) (1-yr)
Reproductive Toxicity NOAEL/LOAEL Oral (mg/kg/day)	Systemic Toxicity NOAEL ~ 452 (females, highest dose tested) Reproductive Toxicity NOAEL ~ 408 LOAEL ~ 39
Developmental Toxicity NOAEL/LOAEL Oral (mg/kg/day) Maternal and Developmental Toxicity	NOAEL ~ 6 LOAEL ~ 60
Genetic Toxicity – Gene Mutation <i>In vitro</i>	Positive
Genetic Toxicity – Chromosomal Aberrations <i>In vitro</i>	Positive
Additional Information Skin Irritation Eye Irritation Skin Sensitization	Mildly irritating Not irritating Sensitizing

Measured data in bold text

Conclusion: Acute oral toxicity of CASRN 55-63-0 to rats and mice and acute dermal toxicity to rabbits is low. Repeated oral exposure of rats to CASRN 55-63-0 for 2 years resulted in histopathologic findings as well as correlative liver weight findings. In females, histopathological changes in the liver (including liver cancer) were observed at 38.1 mg/kg/day and above. Histopathological changes among females were also noted in the spleen and mammary glands at this dose. In males, other histopathological changes in the liver, as well as histopathological findings of the kidney, were observed at doses as low as 3.04 mg/kg/day. The NOAEL for systemic toxicity was 3.99 mg/kg/day for females, and a NOAEL for males could not be established. Repeated oral exposures of CASRN 55-63-0 to mice for 2 years showed decreased survival among females at 9.72 mg/kg/day and above, and methemoglobinemia among males at 114.6 mg/kg/day, and above; the NOAEL for systemic toxicity could not be established for females, and was 11.1 mg/kg/day for males. Oral administration of CASRN 55-63-0 to dogs for 1 year did not show toxicity; the NOAEL for systemic toxicity was 25 mg/kg/day (highest dose tested). In a three-generation dietary reproductive toxicity study in rats, decreased food consumption was observed (at an unspecified dose), as well as signs of reproductive toxicity including severe aspermatogenesis, severe infertility and decreased testicular size, and decreased litter size and birth weights at ~408 mg/kg/day. The NOAEL for systemic toxicity of the females was ~452 mg/kg/day (highest dose tested); the NOAEL for reproductive toxicity was ~39 mg/kg/day. In a prenatal developmental toxicity study, CASRN 55-63-0 was orally administered to pregnant rats during gestation days 6-15. Signs of maternal toxicity included significantly reduced body weight and increased liver weights at ~60 mg/kg/day. Developmental toxicity included increased incidences of diaphragmatic hernias and increased incidences of skeletal anomalies in offspring at ~60 mg/kg/day. The NOAEL for maternal and developmental toxicity was ~6 mg/kg/day. CASRN 55-63-0 was mutagenic in several bacterial reverse mutation assays and induced chromosomal aberrations in mammalian cells *in vitro*. CASRN 55-63-0 is non-irritating to rabbit eyes, a mild irritant to rabbit skin and a dermal sensitizer in guinea pigs.

4. Hazard to the Environment

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

Acute Toxicity to Fish

(1) Rainbow trout (*Onchorhynchus mykiss*) were exposed to the test substance in ethanol at nominal concentrations of 0, 0 (solvent control), 1.0, 1.67, 2.78, 4.63 or 7.72 mg/L under flow-through conditions for 96 hours. Measured concentrations were 0, 0, 0.91, 1.46, 2.47, 3.89 and 6.25 mg/L. All fish exposed to 6.25 mg/L died within 24 hours and all fish exposed to 3.89 mg/L died within 72 hours. Mortality rates at 96 hours were 90 and 10% for the 2.47 and 1.46 mg/L concentrations, respectively. No mortality was noted at 0.91 mg/L or in the control groups.

96-h LC₅₀ = 1.90 mg/L

(2) Fathead minnow (*Pimephales promelas*) were exposed to the test substance at nominal concentrations of 0, 1.04, 1.76, 3.68, 4.80 or 8.0 mg/L under flow-through conditions for 96 hours. Measured concentrations were 0, 0.94, 1.63, 3.53, 4.51 and 7.71 mg/L. Mortality rates at

96 hours were 100, 60, 30, 15, 5 and 0% for the 7.71, 4.51, 3.53, 1.63 and 0.94 mg/L and control groups, respectively.

96-h LC₅₀ = 3.58 mg/L

Acute Toxicity to Aquatic Invertebrates

Water fleas (*Ceriodaphnia dubia*) were exposed to the test substance at nominal concentrations of 0, 5.8, 9.7, 16, 27 or 45 mg/L under static-renewal conditions for 48 hours. Measured concentrations were 0, 5.48, 9.45, 15.53, 26.98 and 44.8 mg/L. In the 44.8 mg/L concentrations, complete lethality was observed by 24 hours. At 26.98 mg/L, mortality rates were 30 and 50% for the two replicates and no effects were noted at ≤ 15.53 mg/L.

48-h EC₅₀ = 17.83 mg/L

Toxicity to Aquatic Plants

Green algae (*Pseudokirchneriella subcapitata*) were exposed to the test substance at nominal concentrations of 0, 0.22, 0.44, 0.72, 1.20 or 2.0 mg/L under static conditions for 96 hours. Measured concentrations were 0, 0.18, 0.37, 0.59, 1.14 and 1.89 mg/L.

96-h EC₅₀ (biomass) = 1.15 mg/L

Conclusion: The 96-hour LC₅₀ of CASRN 55-63-0 to fish is 1.90 mg/L, the 48-hour EC₅₀ for aquatic invertebrates is 17.83 mg/L, and the 96-hour EC₅₀ for aquatic plants is 1.15 mg/L (biomass).

Table 4. Summary of the Screening Information Data Set as Submitted under the U.S. HPV Challenge Program – Aquatic Toxicity Data	
Endpoints	SPONSORED CHEMICAL Nitroglycerin (55-63-0)
Fish 96-h LC₅₀ (mg/L)	1.90
Aquatic Invertebrates 48-h EC₅₀ (mg/L)	17.83
Aquatic Plants 96-h EC₅₀ (mg/L) (growth rate) (biomass)	– 1.15

bold = measured data (i.e., derived from testing); – indicates endpoint not addressed for this chemical