

SCREENING-LEVEL HAZARD CHARACTERIZATION

Trixylenyl Phosphate (CASRN 25155-23-1)

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, EXTTOXNET, 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.

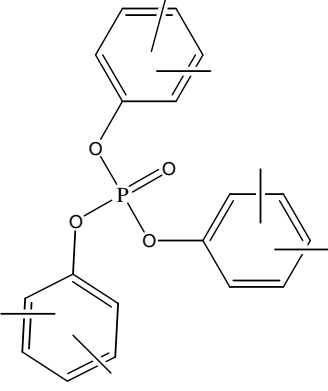
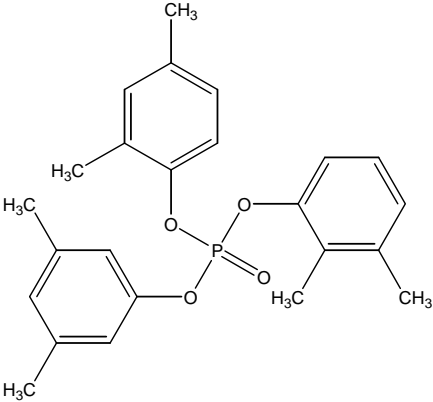
OPPT does not develop HCs for those HPV chemicals which have already been assessed internationally through the HPV program of the Organization for Economic Cooperation and Development (OECD) and for which Screening Initial Data Set (SIDS) Initial Assessment Reports (SIAR) and SIDS Initial Assessment 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>25155-23-1</p>
<p>Chemical Abstract Index Name</p>	<p>Phenol, dimethyl-, phosphate (3:1)</p>
<p>Structural Formula</p>	<div style="display: flex; justify-content: space-around; align-items: center;"> <div style="text-align: center;">  <p>General structure</p> </div> <div style="text-align: center;">  <p>Typical structure</p> </div> </div>

Summary

CASRN 25155-23-1 is a liquid with low water solubility and low vapor pressure at room temperature. It is expected to have low mobility in soil. Volatilization is considered low based on the Henry's Law constant of this substance. The rate of hydrolysis is considered negligible under environmental conditions. The rate of atmospheric photooxidation is considered moderate. CASRN 25155-23-1 is expected to be inherently biodegradable. CASRN 25155-23-1 is judged to have moderate persistence (P2) and low bioaccumulation potential (B1).

The acute oral and dermal toxicity of CASRN 25155-23-1 in rats and rabbits is low. In an oral combined repeated-dose/reproductive/developmental toxicity screening test in rats, toxicity in the liver and adrenals were observed in adult animals at 200 mg/kg-day and above; the NOAEL for systemic toxicity is 25 mg/kg-day. Degenerative changes in the testes and ovaries were also observed at 25 mg/kg-day, the lowest dose tested; the NOAEL for reproductive toxicity is not established. Observations in the offspring were essentially limited to a single dose (1000 mg/kg-day) in the recovery phase of the study due to near complete loss of pregnancies at 200 mg/kg-day and higher in the initial part of the study, however, no effects were noted for litter size, pup survival, or pup body weight at 1000 mg/kg-day; the NOAEL is not established. CASRN 25155-23-1 does not induce genetic mutation or chromosomal aberrations *in vitro*. CASRN 25155-23-1 is irritating to rabbit skin and eye. In a delayed neurotoxicity study in hens, CASRN 25155-23-1 showed possible evidence of neurotoxicity.

The 96-hour LC₅₀ for CASRN 25155-23-1 for fish is > 1.12 mg/L. The 48-hour EC₅₀ of CASRN 25155-23-1 for aquatic invertebrates is 0.06 mg/L. The 96-hour EC₅₀ of CASRN 25155-23-1 for

aquatic plants is > 1.01 mg/L for growth and biomass.

The sponsor, Akzo Nobel Chemicals Inc., submitted a Test Plan and Robust Summaries to EPA for trixylenyl phosphate (CASRN 25155-23-1; 9th CI Name: phenol, dimethyl-, phosphate (3:1)) on September 7, 2001. EPA posted the submission on the ChemRTK HPV Challenge website on October 10, 2001 (<http://www.epa.gov/chemrtk/pubs/summaries/trxpp/c13165tc.htm>). EPA comments on the original submission were posted to the website on May 16, 2002. Public comments were also received and posted to the website. The sponsor submitted updated/revise documents on May 15, 2002, June 7, 2002, June 7, 2002 and September 5, 2006 which were posted to the ChemRTK website on June 4, 2002, July 19, 2002, August 15, 2002 and August 29, 2007, respectively.

1. Chemical Identity

A summary of physical-chemical and environmental fate data submitted is provided in the Table 1 and Table 2. For the purpose of the screening-level hazard characterization, the review and summary of these data were limited to the octanol-water partition coefficient and biodegradation endpoints as indicators of bioaccumulation and persistence, respectively.

1.1 Identification and Purity

The following description is taken from the final Test Plan (2007):

CASRN 25155-23-1 (trixylenyl phosphate) is a liquid with 100% purity. As a commercial product this chemical is also known as Phosflex TXP, Fyrquel EHC, Fyrquel EHC-N and Kronitex.

1.2 Physical-Chemical Properties

The physical-chemical properties of CASRN 25155-23-1 are summarized in Table 1, while the environmental fate properties are provided in Table 2. Commercially this is not a single chemical. It is a mixture of over 50 different compounds, many of which are positional isomers. The number of isomers present in the mixture and their ratios are not reported. A representative structure is provided in the summary.

CASRN 25155-23-1 is a liquid with low water solubility and low vapor pressure at room temperature.

Property	Value
CASRN	25155-23-1
Molecular Weight	410
Physical State	Liquid
Melting Point	<25°C (liquid)
Boiling Point	243–265°C at 10 mm Hg (measured); 379–404°C at 760 mm Hg (estimated) ²
Vapor Pressure	5.2×10 ⁻⁸ mm Hg at 30°C (measured) ³
Water Solubility	0.0186 mg/L at 25°C (measured)
Dissociation Constant (pK _a)	Not applicable
Henry's Law Constant	7.2×10 ⁻⁸ atm-m ³ /mole (estimated) ⁴
Log K _{ow}	5.63 (measured)

¹Akzo Nobel Chemicals Inc. July 2, 2007. Revised Roust Summary and Test Plan for Trixylenyl Phosphate. Available online from:

<http://www.epa.gov/chemrtk/pubs/summaries/trxpp/c13165tc.htm> as of March 30, 2010.

²NOMO5. 1987. Programs to Enhance PC-Gems Estimates of Physical Properties for Organic Compounds. The Mitre Corp.

³SRC. The Physical Properties Database (PHYSPROP). SRC: Syracuse, NY. Available online from:

<http://www.syrres.com/what-we-do/free-demos.aspx> as of March 30, 2010.

⁴U.S. EPA. 2010. Estimation Programs Interface Suite™ for Microsoft® Windows, v4.00.

U.S. Environmental Protection Agency, Washington, DC, USA. Available online from:

<http://www.epa.gov/opptintr/exposure/pubs/episuitedi.htm> as of March 30, 2010.

2. General Information on Exposure

2.1 Production Volume and Use Pattern

According to the 2006 IUR submissions, CASRN 25155-23-1 had an aggregated production and/or import volume in the United States between 1 and 10 million pounds.

Industrial processing and uses as well as commercial and consumer uses for the chemical were claimed confidential.

2.2 Environmental Exposure and Fate

CASRN 25155-23-1 is expected to have low mobility in soil. A commercial mixture of CASRN 25155-23-1 known as Phosflex TXP was not readily biodegradable (0% degradation in 28 days) using a closed bottle test (OECD 301D) and a similar substance was not readily biodegradable using a modified MITI test (OECD 301C); however, it was shown to be inherently biodegradable using mixed microbial populations from activated sludge acclimated to the test substance in a semi-continuous activated sludge (SCAS) system and a die-away test in closed flasks. A modified Sturm test (OECD 301B) using acclimated bacterial seed from a 14-day die-away test indicated that although the test substance was not readily biodegradable, it did achieve 43.8% of

the theoretical CO₂ production with acclimated seed after 28 days. Volatilization of this chemical is considered low based on the Henry's Law constant. The rate of hydrolysis is considered negligible under environmental conditions; but may increase at elevated temperature. CASRN 25155-23-1 is expected to have moderate persistence (P2) and low bioaccumulation potential (B1).

The environmental fate properties are provided in Table 2.

Property	Value
Photodegradation Half-life	2.7 hours (estimated) ²
Hydrolysis Half-life	>1 year at pH 4 and 25°C (measured); >1 year at pH 7 and 25°C (measured); 219 days at pH 9 and 25°C (measured); Significant hydrolysis within 5 days at pH 7 and 9 at 50°C (measured)
Biodegradation	0% after 28 days (not readily biodegradable); 4.7, 43.8, and 65.2% at 7, 28, and 48 days, respectively (not readily biodegradable) ³ ; 65 and 13% after 24 hours at 3 and 13 mg/L/day addition rates (inherently biodegradable) ³ ; 0% after 28 days (not readily biodegradable) ⁴
Bioconcentration	BCF = 405.9 (measured in carp at 10 ppb) ⁴ ; BCF = 359.9 (measured in carp at 1 ppb) ⁴
Bioaccumulation Factor	BAF = 629.7 (estimated) ²
Log K _{oc}	5.3 (estimated) ²
Fugacity (Level III Model) ²	Air (%) <0.1 Water (%) 4.2 Soil (%) 46.4 Sediment (%) 49.4
Persistence ⁵	P2 (moderate)
Bioaccumulation ⁵	B1 (low)

¹Akzo Nobel Chemicals Inc. July 2, 2007. Revised Roust Summary and Test Plan for Trixylenyl Phosphate. Available online from:

<http://www.epa.gov/chemrtk/pubs/summaries/trxpp/c13165tc.htm> as of March 30, 2010.

²U.S. EPA. 2010. Estimation Programs Interface Suite™ for Microsoft® Windows, v4.00. U.S. Environmental Protection Agency, Washington, DC, USA. Available online from: <http://www.epa.gov/opptintr/exposure/pubs/episuitedi.htm> as of March 30, 2010.

³Saeger, V.W.; Hicks, O.; Kaley, R.G.; Michael, P.R.; Mieure, J.P.; Tucker, S.E. 1979. Environmental fate of selected phosphate esters. Environ. Sci. Technol. 13: 840–844.

⁴National Institute of Technology and Evaluation. 2002. Biodegradation and Bioaccumulation of the Existing Chemical Substances under the Chemical Substances Control Law. Available online from: http://www.safe.nite.go.jp/english/kizon/KIZON_start_hazkizon.html as of March 30, 2010.

⁵Federal Register. 1999. Category for Persistent, Bioaccumulative, and Toxic New Chemical Substances. *Federal Register* 64, Number 213 (November 4, 1999) pp. 60194–60204.

Conclusions: CASRN 25155-23-1 is a liquid with low water solubility and low vapor pressure at room temperature. It is expected to have low mobility in soil. Volatilization is considered low based on the Henry's Law constant of this substance. The rate of hydrolysis is considered negligible under environmental conditions. The rate of atmospheric photooxidation is considered moderate. This chemical is expected to be inherently biodegradable. CASRN 25155-23-1 is judged to have moderate persistence (P2) and low bioaccumulation potential (B1).

3. Human Health Hazard

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

Acute Oral Toxicity

(1) Sprague-Dawley rats (5/sex) were administered a single dose of trixylenyl phosphate (Phosflex TXP) via oral gavage at 20,000 mg/kg-bw and monitored for 14 days. No mortality was reported.

LD₅₀ > 20,000 mg/kg-bw

(2) Sprague-Dawley rats (10/sex) were administered a single dose of trixylenyl phosphate (Fyrquel EHC) via oral gavage at 5000 mg/kg-bw. The animals were fasted 24 hours after dosing and monitored for 14 days. No mortality was reported.

LD₅₀ > 5000 mg/kg-bw

Acute Dermal Toxicity

New Zealand White rabbits (5/sex) were administered trixylenyl phosphate (Fyrquel EHC) via the dermal route at a dose of 2000 mg/kg-bw to clipped, intact or abraded skin under semi-occlusive conditions for 24 hours. Following exposure, animals were monitored for 14 days. No mortality was reported. No treatment-related lesions were observed during necropsy.

LD₅₀ > 2000 mg/kg-bw

Repeated-Dose Toxicity

In a combined repeated-dose/reproductive/developmental toxicity screening study, Sprague-Dawley rats (11/sex/dose) were administered trixylenyl phosphate (Phosflex TXP) via oral gavage at 0, 25, 200 or 1000 mg/kg-day for 2 weeks prior to mating, during mating, and through gestation and lactation until sacrifice, for a total of approximately 33 days of dosing for males and 48 days for females. Parameters assessed in the study included parental body weights, food consumption, hematology, clinical chemistry, functional observational batteries (FOB), motor activity, organ weight and histopathology, number of successful matings, fertility, number of pregnancies, litter size, and fetal body weight and survival.

No relevant, treatment-related changes were observed for food consumption, body weight, or body weight gain. There were no differences in the FOB and motor activity measurements between treated and control groups. At the 200 and 1000 mg/kg-day dose groups, biologically relevant changes in serum chemistry, such as increases in blood urea nitrogen (BUN) and alanine aminotransferase (in one or both sexes), and decreases in plasma cholinesterase activity (in both sexes) were observed. Other clinical chemistry changes were noted, but given that the changes were either minimal or the direction of the change had uncertain clinical significance, the study authors did not consider them relevant. Absolute heart weight and heart-to-brain weight ratios were significantly decreased in both sexes at 1000 mg/kg-day. Absolute adrenal weights and adrenal weight ratios were increased in both sexes in the 200 and 1000 mg/kg-day dose groups; an increase was reported in females in the 25 mg/kg-day group. Absolute liver and liver weight ratios were increased in males and females at 1000 mg/kg-day, and in males at 200 and 1000 mg/kg-day. Changes in adrenal-to-brain weight ratios of males, liver in females, and in brain-to-body weight and heart-to-brain weight ratios in both sexes were still present after the recovery period. Gross necropsy findings were not significant. Histopathology revealed diffuse cytoplasmic vacuolation in the adrenals (all three dose levels of males and in the 200 and 1000 mg/kg-day females), and minimal to mild fatty vacuolation of individual hepatocytes in the 200 and 1000 mg/kg-day in females only. Microscopic examination of the heart showed no treatment-related changes. Following the recovery period, the incidence and severity of all changes were decreased in these tissues in the 1000 mg/kg-day group.

LOAEL (systemic/males and females) = 200 mg/kg-day (based on changes in clinical chemistry parameters, increases in organ weight, and histopathology, all consistent with toxicity of the liver and adrenal gland)

NOAEL (systemic/males and females) = 25 mg/kg-day

Reproductive/Developmental Toxicity

In a combined repeated-dose/reproductive/developmental toxicity screening study described above, reproductive and developmental parameters were also evaluated. As a result of the adverse effects on reproductive outcome (successful pregnancies) in the 200 and 1000 mg/kg-day dose groups (see below), the protocol was amended to include mating following a 4-week recovery period. Five males and five females from the control and high-dose groups were designated as recovery animals. The recovery phase of the study included the mating of the recovery animals as well as a cross-over mating. The recovery animals were mated within their respective groups (controls and high-dose) at the end of the four week recovery period. The cross-over mating consisted of cohabitation of high dose males with untreated females. The mating of recovery animals was added to the study in order to determine whether the effects on reproductive function were reversible; whereas the cross-over mating was added in order to determine if the observed effects were either male- or female-mediated. The recovery animals were mated within their respective groups at the end of the four week recovery period. Animals were mated four weeks (males) and three weeks (females) after exposure and then observed and sacrificed 7.5 weeks (males) or nine weeks (females) after successful mating/gestation. The cross-over mating consisted of cohabitation of the high dose males with untreated females and vice versa. Parameters assessed in the study included parental body weight and food

consumption, number of pregnancies, litter size, organ weights, and histopathology of the reproductive organs, as well as fetal body weight and survival.

Mating of the recovery animals showed complete reversal of the effects seen on reproductive performance. For the cross-over mating (untreated females x high dose males), all 5 untreated females become pregnant. For the within-group matings, successful parturition was seen for all sperm-positive dams resulting in 100% pregnancy in the control and high-dose recovery groups. The average length of gestation was similar among all recovery dams. The study authors concluded that the functional deficit in reproductive performance was reversed in males and females after recovery from dosing. Since the reversal was observed in both sexes, it was not possible to determine if the effects on reproductive performance were male- or female-mediated.

Limited treatment-related effects were reported on food consumption and body weights. Food consumption, body weight and body weight gain were slightly decreased in the 1000 mg/kg-day group during week 1 of treatment. During the later part of gestation, food consumption, body weight, and body weight gain were lower in the dams in the 200 and 1000 mg/kg-day groups, but this was thought to be due to the lower rate of successful pregnancies. The number of successful matings (sperm positive) was similar across all dose groups (11/11 mated in controls, and in the 25 and 200 mg/kg-day groups; 10/11 in the 1000 mg/kg-day group); however, reproductive outcome was significantly decreased in the 200 and 1000 mg/kg-day groups. Successful parturition was 100% in controls and 25 mg/kg-day dose groups, and 18% and 0% in the 200 and 1000 mg/kg-day groups. Of the dams that underwent successful parturition, the average length of gestation was unaffected by treatment. Although all animals underwent successful matings, changes in the reproductive organs were reported. Absolute testes and epididymides weights and their ratios were significantly reduced in the 1000 mg/kg-day group. Ovarian weights and ratios were significantly increased in the 200 and 1000 mg/kg-day group. Histopathology of the reproductive organs consisted of degeneration of the germinal epithelium of the testes with corollary findings of sloughed epithelial cells in the lumen of the epididymis (all three dose levels). Findings in the ovaries consisted of distinct mild diffuse hyperplasia of the interstitial cells (all dose levels). Evaluations of the litter survival and offspring body weight were limited to those dams that underwent successful parturition. The percentage of pups surviving to Day 4 were comparable to controls. No statistically significant differences in mean litter size or the number of stillborn pups were reported. No treatment-related effects were reported offspring body weight.

LOAEL (reproductive) = 25 mg/kg-day (based on degenerative changes in testes and ovaries)

NOAEL (reproductive) = Not established

LOAEL (maternal toxicity) = 200 mg/kg-day ((based on changes in clinical chemistry parameters and histopathology consistent with toxicity of the liver and adrenal gland as summarized in the repeated-dose section above)

NOAEL (maternal) = 25 mg/kg-day

LOAEL/NOAEL (developmental) = Not established (based on only a single dose tested and on limited evaluations in offspring during the recovery period of the study)

Genetic Toxicity – Gene Mutation

In vitro

(1) *Salmonella typhimurium* strains TA98, TA100, TA1535, TA1537 and TA1538, and *Escherichia coli* WP2 *uvrA* were exposed to trixylenyl phosphate (Phosflex TXP) diluted in dimethyl sulfoxide at concentrations of 0, 33.3, 100, 333, 1000, 3330 or 5000 µg/plate in the presence and absence of metabolic activation. Positive, negative and vehicle controls were tested concurrently and responded appropriately. Confirmatory assays were also conducted.

Trixylenyl phosphate was not mutagenic in this assay.

(2) In a National Toxicology Program (NTP) study with *S. typhimurium*, strains TA98, TA100, TA1535 and TA1537, were exposed to trixylenyl phosphate mixed isomers diluted in dimethyl sulfoxide at concentrations of 0, 100, 333, 1000, 3333 and 10000 µg/plate in the presence and absence of metabolic activation. All strains were negative for genetic mutations.

Trixylenyl phosphate was not mutagenic in this assay.

Genetic Toxicity – Chromosomal Aberrations

In vitro

Chinese hamster ovary (CHO) cells were exposed to trixylenyl phosphate (Phosflex TXP) diluted in dimethyl sulfoxide at concentrations of 0, 3.38, 4.84, 6.92, 9.89, 14.1, 20.2, 28.8, 41.2, 58.8, 84.0, 120, 172, 245, 350 and 500 µg/mL in the presence and absence of metabolic activation. Positive, negative and solvent controls were conducted concurrently and responded appropriately. A confirmatory assay was also performed.

Trixylenyl phosphate did not induce chromosomal aberrations in this assay.

Additional Information

Skin Irritation

New Zealand White rabbits (6 animals; sex not specified) were administered 0.5 mL of trixylenyl phosphate to shaved intact and abraded skin, under semi-occlusive conditions for 4 hours and observed for irritation at intervals of 4, 24, 48 and 72 hours after application and then observed for 14 days. All animals in the intact and abraded groups showed mild erythema through 24 hours. No edema was observed. At 72 hours, two animals continued to show very mild erythema. The Primary Irritation score was 0.70.

Trixylenyl phosphate was irritating to rabbit skin in this assay.

Eye Irritation

Nine New Zealand white rabbits (sex not specified) were administered 0.1 mL trixylenyl phosphate to the left eye. The right eye of each animal served as the control. Three animals had their eyes rinsed with water for 1 minute approximately 30 seconds after instillation, while the remaining six eyes remained unrinsed. Each treated eye was observed for irritation at 1, 24, 48 and 72 hours and 4 and 7 days after treatment. Mild to moderate irritation was observed at 1 hour in both washed and unwashed eyes of all nine animals. Irritation consisted of redness of the conjunctiva. There were no effects on the cornea or iris. The irritation resolved by the 24-hour observation period.

Trixylenyl phosphate was irritating to the rabbit eye in this assay.

Neurotoxicity

(1) White Leghorn hens (7 animals) were administered a single dose of trixylenyl phosphate via oral gavage at 11,400 mg/kg/day. Two control groups (4 animals/group) received either corn oil (negative control) or 250 mg/kg/day of tri-*ortho* cresyl phosphate (TOCP, positive control). Three of the hens exposed to trixylenyl phosphate were observed daily for 3 weeks, while the remaining four were sacrificed 24 hours after dosing for measurements of brain neurotoxic esterase and cholinesterase activities. The hens in the 3-week observation group developed motor incoordination at day 9 which increased in severity up to the time of sacrifice. One animal was not able to stand 17 days after treatment. The degree of ataxia was similar in intensity to the positive control. The four animals that were sacrificed 24 hours after dosing with trixylenyl phosphate resulted in 85% brain cholinesterase inhibition and 94% brain neurotoxic esterase (NTE). In the positive control, 73% brain cholinesterase inhibition and 89% brain neurotoxic esterase were observed.

Trixylenyl phosphate demonstrated potential neurotoxic activity in this assay.

(2) White Leghorn hens (4 animals/group) were administered trixylenyl phosphate via oral gavage at 114, 1140 and 11,400 mg/kg/day. A negative control (4 animals) group receiving corn oil and a positive control (8 animals) group receiving TOCP were included in the study. Twenty-four hours after dosing, animals were sacrificed for measurements of brain neurotoxic esterase and cholinesterase activities. The inhibition of neurotoxic esterase for 114, 1140, and 11,400 were 2.0, 13.4 and 55.8 percent, respectively. The inhibition in the positive control was 90.3 percent. Since inhibition of greater than 70% is thought to be necessary to elicit neurotoxic activity, all three doses were unlikely to induce neurotoxicity. Cholinesterase inhibition was inhibited 114 and 1140 mg/kg/day as well as in the positive control animals.

Trixylenyl phosphate demonstrated potential neurotoxic activity in this assay.

Conclusions: The acute oral and dermal toxicity of CASRN 25155-23-1 in rats and rabbits is low. In an oral combined repeated-dose/reproductive/developmental toxicity screening test in rats, toxicity in the liver and adrenals were observed in adult animals at 200 mg/kg-day and above; the NOAEL for systemic toxicity is 25 mg/kg-day. Degenerative changes in the testes and ovaries were also observed at 25 mg/kg-day, the lowest dose tested; the NOAEL for reproductive toxicity is not established. Observations in the offspring were essentially limited to a single dose (1000 mg/kg-day) in the recovery phase of the study due to near complete loss of pregnancies at 200 mg/kg-day and higher in the initial part of the study, however, no effects were

noted for litter size, pup survival, or pup body weight at 1000 mg/kg-day; the NOAEL is not established. CASRN 25155-23-1 does not induce genetic mutation or chromosomal aberrations *in vitro*. CASRN 25155-23-1 is irritating to rabbit skin and eye. In a delayed neurotoxicity study in hens, CASRN 25155-23-1 showed possible evidence of neurotoxicity.

Table 3. Summary Table of the Screening Information Data Set Submitted under the U.S. HPV Challenge Program: Human Health Data	
Endpoints	SPONSORED CHEMICAL Trixylenyl phosphate (25155-23-1)
Acute Oral Toxicity LD₅₀ (mg/kg)	> 5000
Acute Dermal Toxicity LD₅₀ (mg/kg-bw)	> 2000
Repeated-Dose Toxicity NOAEL/LOAEL Oral (mg/kg-day) Systemic (Rat)	NOAEL = 25 (33/48-d) LOAEL = 200 (33/48-d)
Reproductive/Developmental Toxicity NOAEL/LOAEL Oral (mg/kg-day) Reproductive Toxicity Maternal Toxicity Developmental Toxicity	NOAEL = Not established LOAEL = 25 NOAEL = 25 LOAEL = 200 LOAEL/NOAEL = Not established
Genetic Toxicity – Gene Mutation <i>In vitro</i>	Negative
Genetic Toxicity – Chromosomal Aberrations <i>In vitro</i>	Negative
Additional Information Skin Irritation (Rabbit) Eye Irritation (Rabbit) Neurotoxicity (Hen)	Irritating Irritating Positive

– indicates that endpoint was not addressed for this chemical.

4. Hazard to the Environment

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

Acute Toxicity to Fish

Fathead minnows (*Pimephales promelas*; 10/concentration) were exposed to CASRN 25155-23-1 at mean measured concentrations of 0.12, 0.27, 0.42, 0.78, and 1.111 mg/L under flow-through conditions for 96 hours. The solvent DMF was used. There were no signs of mortality. A 96-h $LC_{50} > 1.12$ was reported. There were no effects at water solubility limit.

96-h LC_{50} = NES

Acute Toxicity to Aquatic Invertebrates

Water fleas (*Daphnia magna*; 20/concentration) were exposed to CASRN 25155-23-1 at mean measured concentrations of 0.011, 0.02, 0.05, 0.11, 0.23, 0.42, 0.90, and 1.60 mg/L under static-renewal conditions for 48 hours. The solvent DMF was used. A 48-h EC_{50} of 0.06 mg/L was reported.

96-h EC_{50} = 0.06 mg/L

Toxicity to Aquatic Plants

Green algae (*Pseudokirchneriella subcapitata*) were exposed to measured concentrations of 0.05, 0.11, 0.23, 0.48 and 1.01 mg/L for 96 hours under static conditions. Three negative control replicates were also conducted. No effects at the water solubility limit were reported.

96-h EC_{50} (biomass) = NES

96-h EC_{50} (growth) = NES

Conclusions: The 96-hour LC_{50} for CASRN 25155-23-1 for fish is > 1.12 mg/L. The 48-hour EC_{50} of CASRN 25155-23-1 for aquatic invertebrates is 0.06 mg/L. The 96-hour EC_{50} of CASRN 25155-23-1 for aquatic plants is > 1.01 mg/L for growth and biomass.

Table 4. Summary Table of the Screening Information Data Set Submitted under the U.S. HPV Challenge Program: Aquatic Toxicity Data	
Endpoints	SPONSORED CHEMICAL Trixylenyl phosphate (25155-23-1)
Fish 96-h LC₅₀ (mg/L)	NES
Aquatic Invertebrates 48-h EC₅₀ (mg/L)	0.06
Aquatic Plants 72-h EC₅₀ (mg/L) (growth) or(biomass)	NES
Chronic Toxicity to Aquatic Invertebrates 21-d EC₅₀	0.003 (e)

– indicates the endpoint was not addressed for this chemical; NES, No effect at saturation; (e), estimated value.