FINAL
8-11-2014

Provisional Peer-Reviewed Toxicity Values for

Styrene-Acrylonitrile (SAN) Trimer
(Various CASRNs)

Superfund Health Risk Technical Support Center
National Center for Environmental Assessment
Office of Research and Development
U.S. Environmental Protection Agency
Cincinnati, OH 45268

AUTHORS, CONTRIBUTORS, AND REVIEWERS

CHEMICAL MANAGERS

J. Phillip Kaiser, PhD, DABT
National Center for Environmental Assessment, Cincinnati, OH

Dan D. Petersen, PhD, DABT
National Center for Environmental Assessment, Cincinnati, OH

CONTRIBUTOR

Scott C. Wesselkamper, PhD
National Center for Environmental Assessment, Cincinnati, OH

PRIMARY INTERNAL REVIEWERS

Q. Jay Zhao, PhD, MPH, DABT
National Center for Environmental Assessment, Cincinnati, OH

Paul G. Reinhart, PhD, DABT
National Center for Environmental Assessment, Research Triangle Park, NC

This document was externally peer reviewed under contract to
Eastern Research Group, Inc.
110 Hartwell Avenue
Lexington, MA 02421-3136

Questions regarding the contents of this document may be directed to the U.S. EPA Office of Research and Development's National Center for Environmental Assessment, Superfund Health Risk Technical Support Center (513-569-7300).

TABLE OF CONTENTS

COMMONLY USED ABBREVIATIONS AND ACRONYMS

α2u-g	alpha 2u-globulin
ACGIH	American Conference of Governmental Industrial Hygienists
AIC	Akaike's information criterion
ALD	approximate lethal dosage
ALT	alanine aminotransferase
AST	aspartate aminotransferase
atm	atmosphere
ATSDR	Agency for Toxic Substances and Disease Registry
BMD	benchmark dose
BMDL	benchmark dose lower confidence limit
BMDS	Benchmark Dose Software
BMR	benchmark response
BUN	blood urea nitrogen
BW	body weight
CA	chromosomal aberration
CAS	Chemical Abstracts Service
CASRN	Chemical Abstracts Service Registry Number
CBI	covalent binding index
CHO	Chinese hamster ovary (cell line cells)
CL	confidence limit
CNS	central nervous system
CPN	chronic progressive nephropathy
CYP450	cytochrome P450
DAF	dosimetric adjustment factor
DEN	diethylnitrosamine
DMSO	dimethylsulfoxide
DNA	deoxyribonucleic acid
EPA	Environmental Protection Agency
FDA	Food and Drug Administration
FEV1	forced expiratory volume of 1 second
GD	gestation day
GDH	glutamate dehydrogenase
GGT	γ-glutamyl transferase
GSH	glutathione
GST	glutathione-S-transferase
Hb/g-A	animal blood-gas partition coefficient
Hb/g-H	human blood-gas partition coefficient
HEC	human equivalent concentration
HED	human equivalent dose
i.p.	intraperitoneal
IRIS	Integrated Risk Information System
IVF	in vitro fertilization
LC_{50}	median lethal concentration
LD_{50}	median lethal dose
LOAEL	lowest-observed-adverse-effect level
MN	micronuclei
MNPCE	micronucleated polychromatic erythrocyte
MOA	mode-of-action
MTD	maximum tolerated dose
NAG	N-acetyl-β-D-glucosaminidase
NCEA	National Center for Environmental Assessment
NCI	National Cancer Institute
NOAEL	no-observed-adverse-effect level
NTP	National Toxicology Program
NZW	New Zealand White (rabbit breed)
OCT	ornithine carbamoyl transferase
ORD	Office of Research and Development
PBPK	physiologically based pharmacokinetic
PCNA	proliferating cell nuclear antigen
PND	postnatal day
POD	point of departure
$POD_{[ADJ]}$	duration-adjusted POD
QSAR	quantitative structure-activity relationship
RBC	red blood cell
RDS	replicative DNA synthesis
RfC	inhalation reference concentration
RfD	oral reference dose
RGDR	regional gas dose ratio
RNA	ribonucleic acid
SAR	structure activity relationship
SCE	sister chromatid exchange
SD	standard deviation
SDH	sorbitol dehydrogenase
SE	standard error
SGOT	glutamic oxaloacetic transaminase, also known as AST
SGPT	glutamic pyruvic transaminase, also known as ALT
SSD	systemic scleroderma
TCA	trichloroacetic acid
TCE	trichloroethylene
TWA	time-weighted average
UF	uncertainty factor
UF_A	interspecies uncertainty factor
UF_H	intraspecies uncertainty factor
UF_S	subchronic-to-chronic uncertainty factor
UF_D	database uncertainty factor
U.S.	United States of America
WBC	white blood cell

PROVISIONAL PEER-REVIEWED TOXICITY VALUES FOR STYRENE ACRYLONITRILE TRIMER

BACKGROUND

A Provisional Peer-Reviewed Toxicity Value (PPRTV) is defined as a toxicity value derived for use in the Superfund Program. PPRTVs are derived after a review of the relevant scientific literature using established Agency guidance on human health toxicity value derivations. All PPRTV assessments receive internal review by a standing panel of National Center for Environment Assessment (NCEA) scientists and an independent external peer review by five scientific experts.

The purpose of this document is to provide support for the hazard and dose-response assessment pertaining to chronic and subchronic exposures to substances of concern, to present the major conclusions reached in the hazard identification and derivation of the PPRTVs, and to characterize the overall confidence in these conclusions and toxicity values. It is not intended to be a comprehensive treatise on the chemical or toxicological nature of this substance.

The PPRTV review process provides needed toxicity values in a quick turnaround timeframe while maintaining scientific quality. PPRTV assessments are updated approximately on a 5-year cycle for new data or methodologies that might impact the toxicity values or characterization of potential for adverse human health effects and are revised as appropriate. It is important to utilize the PPRTV database (http://hhpprtv.ornl.gov) to obtain the current information available. When a final Integrated Risk Information System (IRIS) assessment is made publicly available on the Internet (www.epa.gov/iris), the respective PPRTVs are removed from the database.

DISCLAIMERS

The PPRTV document provides toxicity values and information about the adverse effects of the chemical and the evidence on which the value is based, including the strengths and limitations of the data. All users are advised to review the information provided in this document to ensure that the PPRTV used is appropriate for the types of exposures and circumstances at the site in question and the risk management decision that would be supported by the risk assessment.

Other U.S. Environmental Protection Agency (EPA) programs or external parties who may choose to use PPRTVs are advised that Superfund resources will not generally be used to respond to challenges, if any, of PPRTVs used in a context outside of the Superfund program.

QUESTIONS REGARDING PPRTVs

Questions regarding the contents and appropriate use of this PPRTV assessment should be directed to the U.S. EPA Office of Research and Development's National Center for Environmental Assessment, Superfund Health Risk Technical Support Center (513-569-7300).

INTRODUCTION

Styrene-acrylonitrile Trimer (SAN Trimer) exists as a mixture of isomers of 4-cyano-1,2,3,4-tetrahydro-a-methyl-1-naphthaleneacetonitrile (THNA; CASRN 57964-39-3) and 4-cyano-1,2,3,4-tetrahydro-1-naphthalene-propionitrile (THNP; CASRN 57964-40-6) and is formed by the condensation of two moles of acrylonitrile and one mole of styrene. The SAN Trimer mixture includes the THNA form, consisting of four stereoisomers (*cis*-R-THNA, 30%; *cis*-S-THNA, 17%; *trans*-R-THNA, 25%; and *trans*-S-THNA, 14%), and the THNP form, consisting of two stereoisomers (*cis*-THNP, 8% and *trans*-THNP, 6%) (NTP, 2012) (see Figure 1). SAN Trimer is a by-product of specific manufacturing processes for polymers of styrene and acrylonitrile, but it is currently not considered commercially useful (NTP, 2012). The mixture exists as a viscous, light-brown, opaque liquid (at room temperature). SAN Trimer was selected for study by a United States Environmental Protection Agency (U.S. EPA) workgroup that was formed due to reports by the New Jersey Department of Health and Senior Services (NJ DHSS) regarding a potential link between childhood cancer incidence rates and previously unknown semivolatile contaminants (now known as SAN Trimer) in groundwater after dumping of spent process streams from styrene-acrylonitrile polymer manufacturing (NJ DHSS, 2003a, b, 1997). A table of physicochemical properties for SAN Trimer is provided (see Table 1).

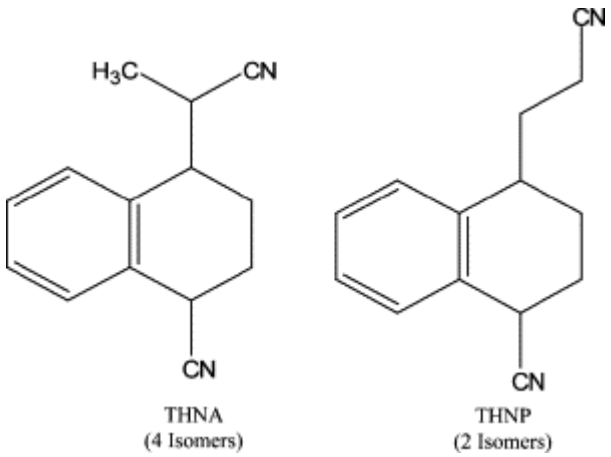

Figure 1. Structures of the SAN Trimers (Gargas et al., 2008)

Table 1. Physicochemical Properties of SAN Trimer (Various CASRNs)[a]

Property (unit)	Value
Boiling point (°C)	ND
Melting point (°C)	ND
Density (g/cm^3 at 20°C)	1.101
Vapor pressure (mm Hg at 235°C)	Estimated at 2.5
pH (unitless)	ND
Solubility in water (mg/L)	84.9
Specific gravity (at 20°C)	1.103
Molecular weight (g/mol)	210

[a]NTP (2012).

ND = no data.

No toxicity values were identified for SAN Trimer.

Literature searches were conducted on sources published from 1900 through February 2014 for studies relevant to the derivation of provisional toxicity values for SAN Trimer (various CASRNs). The following databases were searched by chemical name, synonyms, or CASRN: ACGIH, ANEUPL, ATSDR, BIOSIS, Cal/EPA, CCRIS, CDAT, ChemIDplus, CIS, CRISP, DART, EMIC, EPIDEM, ETICBACK, FEDRIP, GENE-TOX, HAPAB, HERO, HMTC, HSDB, IARC, INCHEM IPCS, IPA, ITER, IUCLID, LactMed, NIOSH, NTIS, NTP, OSHA, OPP/RED, PESTAB, PPBIB, PPRTV, PubMed (toxicology subset), RISKLINE, RTECS, TOXLINE, TRI, U.S. EPA IRIS, U.S. EPA HEAST, U.S. EPA HEEP, U.S. EPA OW, and U.S. EPA TSCATS/TSCATS2. The following databases were searched for relevant health information: ACGIH, ATSDR, Cal/EPA, U.S. EPA IRIS, U.S. EPA HEAST, U.S. EPA HEEP, U.S. EPA OW, U.S. EPA TSCATS/TSCATS2, NIOSH, NTP, OSHA, and RTECS.

REVIEW OF POTENTIALLY RELEVANT DATA (NONCANCER AND CANCER)

Tables 2A and 2B provides an overview of the relevant database for SAN Trimer and includes all potentially relevant repeat-dose short-term-, subchronic-, and chronic-duration studies. Principal studies are identified. The phrase "statistical significance," used throughout the document, indicates a *p*-value of <0.05 (i.e., the probability that differences in observed data are not due to chance alone).

Table 2A. Summary of Potentially Relevant Noncancer Data for SAN Trimer (Various CASRNs)									
Category[a]	**Number of Male/Female, Strain, Species, Study Type, Study Duration**	**Dosimetry[b]**	**Critical Effects**	**NOAEL**	**BMDL/ BMCL**	**LOAEL**	**Reference**	**Comments**	**Notes**
Human									
1. Oral									
Acute[c]	ND								
Short-term[d]	ND								
Long-term[e]	ND								
Chronic[f]	ND								
2. Inhalation									
Acute	ND								
Short-term	ND								
Long-term	ND								
Chronic	ND								
Animal									
1. Oral									
Acute	5/5, S-D, rat, gavage, single administration then observed for 14 days	0, 250, 500, or 1,000 mg/kg	Mortality: (sex not specified), 20% at 250 mg/kg, 40% at 500 mg/kg, 90% at 1,000 mg/kg.	NA	NC	NA	Huntingdon Life Sciences (1999a)	LD_{50} study	NPR

Table 2A. Summary of Potentially Relevant Noncancer Data for SAN Trimer (Various CASRNs)

Category[a]	Number of Male/Female, Strain, Species, Study Type, Study Duration	Dosimetry[b]	Critical Effects	NOAEL	BMDL/ BMCL	LOAEL	Reference	Comments	Notes
Short-term	6/6, S-D, rat, gavage, 14 days	0, 30, 75, 150, or 300 mg/kg-day	Increased absolute (150 mg/kg-day) and relative liver (≥75 mg/kg-day) weights in males; increased absolute and relative liver weights at ≥150 mg/kg-day in females; increased absolute and relative heart weights at ≥150 mg/kg-day in females; mortality in both males and females at 300 mg/kg-day.	30	NC	75	Huntingdon Life Sciences (1999b)		NPR
	0/7−8, F344/N, rat, dietary, GD 7−delivery (F0 dams)	0, 18, 35, 67, 130, or 197 mg/kg-day	Decreased body weight on GD 14.	130	NDr	197	NTP (2012)	Decreased body weight in dams could be due to decreased food consumption	PR; gestational component of the 2-week postweaning NTP (2012) study.

Table 2A. Summary of Potentially Relevant Noncancer Data for SAN Trimer (Various CASRNs)

Category[a]	Number of Male/Female, Strain, Species, Study Type, Study Duration	Dosimetry[b]	Critical Effects	NOAEL	BMDL/ BMCL	LOAEL	Reference	Comments	Notes
Short-term	0/6−8, F344/N, rat, dietary, PNDs 1−20 (F0 dams) 10/10 F1 pups	0, 40, 85, 166, 325, or 634 mg/kg-day	Decreased body weight in F0 dams on PNDs 1−20 and F1 pups on PND 20 (males and females).	F0 dams: 325 F1 pups (males and females): 166	NDr	634 325	NTP (2012)	Decreased body weight in dams could be due to decreased food consumption	PR; lactational component of the 2-week postweaning NTP (2012) study. Doses for F1 pups were calculated assuming that they received 100% of the dose given to dams.
	10/10, F344/N, rat, dietary, 2 weeks postweaning (F1 rats from dams exposed GD 7−PND 20)	0, 50, 90, 175, 270, or 410 mg/kg-day (F1 males) and 0, 45, 90, 185, 295, or 430 mg/kg-day (F1 females)	Decreased absolute and relative right testis and thymus weights at ≥90 mg/kg-day in males; increased relative liver weight at ≥175 mg/kg-day in males and at ≥185 mg/kg-day in females; other relative organ weight changes at doses ≥270 mg/kg-day in males and ≥295 mg/kg-day in females also observed; decreased body weight at ≥175 mg/kg-day in males and ≥295 mg/kg-day in females; increased brain cellularity in females at 430 mg/kg-day.	50	NC	90	NTP (2012)		PR

Table 2A. Summary of Potentially Relevant Noncancer Data for SAN Trimer (Various CASRNs)

Category[a]	Number of Male/Female, Strain, Species, Study Type, Study Duration	Dosimetry[b]	Critical Effects	NOAEL	BMDL/ BMCL	LOAEL	Reference	Comments	Notes
Subchronic	0/8−9, F344/N, rat, dietary, GD 7−delivery (F0 dams)	0, 6.9, 14, 28, 55, or 110 mg/kg-day	Decreased body weight on GD 20.	55	NDr	110	NTP (2012)		PR; gestational component of the 13-week postweaning NTP (2012) study.
	0/4−8, F344/N, rat, dietary, PNDs 1−20 (F0 dams) 10/10 F1 pups	0, 17, 33, 66, 132, or 264 mg/kg-day	No effects observed in F0 dams or F1 pups.	264	NDr	NDr	NTP (2012)		PR; lactational component of the 13-week postweaning NTP (2012) study.
	10/10, F344/N, rat, dietary, 13 weeks postweaning (F1 rats from dams exposed GD 7−PND 20)	**0, 10, 20, 40, 80, or 150 mg/kg-day (F1 males and females)**	**Increased absolute and relative liver weights in males at ≥40 mg/kg-day; increased absolute and relative heart weights in males at ≥10 mg/kg-day and relative heart weight in females at ≥80 mg/kg-day; other absolute and relative organ weight changes at doses ≥40 mg/kg-day in males and females also observed; decreased body weight at 150 mg/kg-day in males.**	**NDr**	**10 (based on increased absolute liver weight in males)**	**10**	**NTP (2012)**		**PR, PS**

Table 2A. Summary of Potentially Relevant Noncancer Data for SAN Trimer (Various CASRNs)

Category[a]	Number of Male/Female, Strain, Species, Study Type, Study Duration	Dosimetry[b]	Critical Effects	NOAEL	BMDL/ BMCL	LOAEL	Reference	Comments	Notes
Chronic	0/41−42, F344/N, rat, dietary, GD 7−delivery (F0 dams)	0, 28, 55, or 110 mg/kg-day	No effects observed.	110	NDr	NDr	NTP (2012)		PR; gestational component of the 2-year postweaning NTP (2012) study.
	0/24−27, F344/N, rat, dietary, PNDs 1−20 (F0 dams) 50/50 F1 pups	0, 66, 132, or 264 mg/kg-day	No effects observed in F0 dams or F1 pups.	264	NDr	NDr	NTP (2012)		PR; lactational component of the 2-year postweaning NTP (2012) study.

Table 2A. Summary of Potentially Relevant Noncancer Data for SAN Trimer (Various CASRNs)

Category[a]	Number of Male/Female, Strain, Species, Study Type, Study Duration	Dosimetry[b]	Critical Effects	NOAEL	BMDL/ BMCL	LOAEL	Reference	Comments	Notes
Chronic	**50/50, F344/N, rat, dietary, 2 years postweaning (F1 rats from dams exposed GD 7−PND 20)**	**0, 20, 40, or 75 mg/kg-day (F1 males) and 0, 20, 40, or 85 mg/kg-day (F1 females)**	**Increased incidences of mixed cell foci in liver and hyperplasia of the transitional epithelium of the bladder at 85 mg/kg-day in females; spinal root degeneration, chronic active liver inflammation, eosinophilic angiectasis, and bone marrow hyperplasia at 75 mg/kg-day in males; sciatic nerve degeneration, bone marrow hyperplasia, and granulomatosis inflammation at ≥40 mg/kg-day in females; mixed cell foci in liver at ≥20 mg/kg-day in males.**	**NA**	**3.2 (based on increased incidence of chronic active liver inflammation in male rats)**	**20**	**NTP (2012)**		**PR, PS**
Developmental	ND								
Reproductive	ND								

Table 2A. Summary of Potentially Relevant Noncancer Data for SAN Trimer (Various CASRNs)

Category[a]	Number of Male/Female, Strain, Species, Study Type, Study Duration	Dosimetry[b]	Critical Effects	NOAEL	BMDL/ BMCL	LOAEL	Reference	Comments	Notes
2. Inhalation									
Subchronic	ND								
Chronic	ND								
Developmental	ND								
Reproductive	ND								

[a]The duration classification for the studies from NTP (2012) is based on the time that F1 rats were directly treated with SAN Trimer in the diet.
[b]Dosimetry: NOAEL, BMDL/BMCL, and LOAEL values are converted to an adjusted daily dose (ADD in mg/kg-day) for oral noncancer effects and a human equivalent dose (HED in mg/kg-day) for oral carcinogenic effects. All long-term exposure values (4 weeks and longer) are converted from a discontinuous to a continuous exposure.
[c]Acute = exposure for ≤24 hours (U.S. EPA, 2002).
[d]Short-term = repeated exposure for >24 hours ≤30 days (U.S. EPA, 2002).
[e]Long-term = repeated exposure for >30 days ≤10% lifespan (based on 70-year typical life span) (U.S. EPA, 2002).
[f]Chronic = repeated exposure for >10% life span (U.S. EPA, 2002).

GD = Gestation Day; ND = no data; NA = not applicable; NDr = not determined; NC = not calculated; PR = peer-reviewed; NPR = non-peer-reviewed; PND = Postnatal Day; PS = principal study; S-D = Sprague-Dawley.

Table 2B. Summary of Potentially Relevant Cancer Data for SAN Trimer (Various CASRNs)									
Category[a]	**Number of Male/Female, Strain, Species, Study Type, Study Duration**	**Dosimetry[b]**	**Critical Effects**	**NOAEL**	**BMDL/ BMCL**	**LOAEL**	**Reference**	**Comments**	**Notes**
Human									
1. Oral									
Acute[c]	ND								
Short-term[d]	ND								
Long-term[e]	ND								
Chronic[f]	ND								

Table 2B. Summary of Potentially Relevant Cancer Data for SAN Trimer (Various CASRNs)

Category[a]	Number of Male/Female, Strain, Species, Study Type, Study Duration	Dosimetry[b]	Critical Effects	NOAEL	BMDL/ BMCL	LOAEL	Reference	Comments	Notes
Exposure duration cannot be determined	Evaluation of childhood cancer statistics from 1979 to 1995 for Ocean County, Dover Township, and the Toms River section of Dover Township, NJ	NDr	Ocean County: Increased incidences of sympathetic nervous system cancers (i.e., neuroblastomas) particularly in males <5 years old and brain/CNS astrocytomas (0−19 years of age and both sexes). Dover Township: Increased incidences of total childhood cancer (particularly in females <5 years old), leukemia, and acute lymphocytic leukemia (both in females at all ages). Toms River section of Dover Township: Increased incidences of total childhood cancer particularly in females <5 years old, brain/CNS cancer (both sexes), brain/CNS astrocytomas (both sexes), leukemia (females), and acute lymphocytic leukemia (females) in children <5 years old.	NDr	NC	NDr	NJ DHSS (1997); ATSDR (1997)	Epidemiologic study on small population to assess standardized incidence ratios for childhood cancers.	NPR; the drinking water supply was contaminated with SAN Trimer and other chemicals with known carcinogenic potential, including trichloroethylene and tetrachloroethylene, as discussed in NJ DHSS (2003a) and in a drinking water quality analysis (NJ DHSS, 2001).

Table 2B. Summary of Potentially Relevant Cancer Data for SAN Trimer (Various CASRNs)

Category[a]	Number of Male/Female, Strain, Species, Study Type, Study Duration	Dosimetry[b]	Critical Effects	NOAEL	BMDL/ BMCL	LOAEL	Reference	Comments	Notes
Exposure duration cannot be determined	Case-control design evaluating residents from Dover Township and the Toms River section of Dover Township, NJ from 1979 to 1996: interview (40 incident cases/159 controls); birth records (48 cases/480 controls)	NDr	Increased odds ratios (OR) for prenatal exposure to contaminated well water and leukemia in females ≤19 years old.	NDr	NC	NDr	NJ DHSS (2003a)	Epidemiologic study on small population; supports increased incidence of leukemia in children.	PR; the drinking water supply was contaminated with SAN Trimer and other chemicals with known carcinogenic potential, including trichloroethylene and tetrachloroethylene, as discussed in NJ DHSS (2003a) and in a drinking water quality analysis (NJ DHSS, 2001).

Table 2B. Summary of Potentially Relevant Cancer Data for SAN Trimer (Various CASRNs)

Category[a]	Number of Male/Female, Strain, Species, Study Type, Study Duration	Dosimetry[b]	Critical Effects	NOAEL	BMDL/ BMCL	LOAEL	Reference	Comments	Notes
Exposure duration cannot be determined	Evaluation of childhood cancer statistics from 1996 to 2000 for Dover Township and the Toms River section of Dover Township, NJ	NDr	No statistically significant increase in childhood cancer was observed.	NDr	NC	NDr	NJ DHSS (2003b)	Epidemiologic study on small population to assess standardized incidence ratios for childhood cancers	NPR; the drinking water supply was contaminated with SAN Trimer and other chemicals with known carcinogenic potential, including trichloroethylene and tetrachloroethylene, as discussed in NJ DHSS (2003a) and in a drinking water quality analysis (NJ DHSS, 2001).

Table 2B. Summary of Potentially Relevant Cancer Data for SAN Trimer (Various CASRNs)									
Category[a]	**Number of Male/Female, Strain, Species, Study Type, Study Duration**	**Dosimetry**[b]	**Critical Effects**	**NOAEL**	**BMDL/ BMCL**	**LOAEL**	**Reference**	**Comments**	**Notes**
Exposure duration cannot be determined	Evaluation of childhood cancer statistics from 2001 to 2005 for Toms River Township (formerly Dover Township) and the sub-Township area (formerly the Toms River section of Dover Township, NJ)	NDr	Increased incidence in soft tissue sarcomas in females ≤19 years old (2004−2005) in both the Toms River Township and sub-Township area.	NDr	NC	NDr	NJ DHSS (2008)	Epidemiologic study on small population; standardized incidence ratios (SIRs) for childhood cancers previously elevated at an earlier time interval (1979−1995) returned to background levels.	NPR; the drinking water supply was contaminated with SAN Trimer and other chemicals with known carcinogenic potential, including trichloroethylene and tetrachloroethylene, as discussed in NJ DHSS (2003a) and in a drinking water quality analysis (NJ DHSS, 2001).
2. Inhalation									
ND									
Animal									
1. Oral									
ND									
Carcinogenic	50/50, F344/N, rat, dietary, 2 year postweaning (F1 rats from dams exposed GD 7−PND 20)	HEDs: 0, 5.6, 11, or 20 mg/kg-day (F1 males) and 0, 5.0, 9.8, or 20 mg/kg-day (F1 females)	No statistically significant increases in tumor incidence observed.	NA	NC	NA	NTP (2012)		PR

Table 2B. Summary of Potentially Relevant Cancer Data for SAN Trimer (Various CASRNs)

Category[a]	Number of Male/Female, Strain, Species, Study Type, Study Duration	Dosimetry[b]	Critical Effects	NOAEL	BMDL/ BMCL	LOAEL	Reference	Comments	Notes
2. Inhalation									
Carcinogenic	ND								

[a]The duration classification for the studies from NTP (2012) is based on the time that F1 rats were directly treated with SAN Trimer in the diet.
[b]Dosimetry: NOAEL, BMDL/BMCL, and LOAEL values are converted to an adjusted daily dose (ADD in mg/kg-day) for oral noncancer effects and a human equivalent dose (HED in mg/kg-day) for oral carcinogenic effects. All long-term exposure values (4 weeks and longer) are converted from a discontinuous to a continuous exposure.
[c]Acute = exposure for ≤24 hours (U.S. EPA, 2002).
[d]Short-term = repeated exposure for >24 hours ≤30 days (U.S. EPA, 2002).
[e]Long-term = repeated exposure for >30 days ≤10% lifespan (based on 70-year typical life span) (U.S. EPA, 2002).
[f]Chronic = repeated exposure for >10% life span (U.S. EPA, 2002).

GD = Gestation Day; ND = no data; NA = not applicable; NDr = not determined; NC = not calculated; PR = peer-reviewed; PND = Postnatal Day.

HUMAN STUDIES

Oral Exposure

Four human studies on oral exposure to drinking water containing various chemicals, including SAN Trimer, are publicly available; one was a case-control study and the other three were ecological in nature because they examined town-level incidence and exposure data. These studies evaluated childhood cancer incidence in Ocean County, Dover Township, and the Toms River section of Dover Township, New Jersey, as discussed in NJ DHSS (2003a) and in a drinking water quality analysis (NJ DHSS, 2001). The three ecological studies were conducted covering the years 1979−1995 (NJ DHSS, 1997), and then extended to cover subsequent 5-year intervals from 1996−2000 (NJ DHSS, 2003b) and 2001−2005 (NJ DHSS, 2008). The case-control study analyzed the same population from 1979−1996 (NJ DHSS, 2003a). These studies are summarized chronologically below. It should be noted that due to the existence of other contaminants in drinking water from the well fields investigated, the conclusions that can be drawn from the epidemiologic studies regarding the toxicity of SAN Trimer are relatively limited. Some of the chemicals that were present in the drinking water (e.g., trichloroethylene and tetrachloroethylene) are known to have carcinogenic potential. Finally, even if SAN Trimer were determined to have carcinogenic potential, the dosimetry necessary to establish a dose-response relationship is absent because SAN Trimer concentrations in the drinking water were not measured as part of these studies.

NJ DHSS (1997)

Due to residential concern about cancer levels in the Toms River section of Dover Township, New Jersey, the NJ DHSS in collaboration with the ATSDR reviewed cancer incidence data from the New Jersey State Cancer Registry for children living in Ocean County, Dover Township, and the Toms River section of Dover Township in 1997 (see Figure A-1) in a non-peer-reviewed study (NJ DHSS, 1997). The purpose of the report was to verify results and extend the analysis of the time period investigated in a previous cancer analysis done by the New Jersey Department of Health (NJDOH) and the ATSDR (NJDOH, 1996) (not publicly available) and to determine which cancer types were increased. Residents under the age of 20 who were diagnosed with selected childhood cancer types (i.e., leukemia, brain and central nervous system, lymphomas, soft tissue sarcomas, sympathetic nervous system, bone, and kidney cancer) during a 17-year period (1979−1995) were included in the analysis. To quantitatively analyze childhood cancer incidence, the study authors calculated standardized incidence ratios (SIR) for each area by dividing the reported number of new cancers by the expected number using the State of New Jersey as a comparison population. An SIR greater than one indicates that more cancer cases than expected were observed. The study authors conducted statistical evaluations using a 95% confidence interval (CI) to determine whether differences in an SIR were due to chance alone. A 95% CI that includes the null value of 1 indicates results that are not statistically significant at a *p*-value of 0.05.

In Ocean County, there was no increase in total childhood cancer incidence compared to the expected number. However, there was a statistically significant increase in sympathetic nervous system cancers (i.e., neuroblastomas), particularly in males (SIR = 2.21, CI = 1.29–3.54) under the age of 5 (see Table A-1) (NJ DHSS, 1997). There was also a statistically significant increase in brain/CNS astrocytomas when all age, race, and sex groups were pooled (SIR = 1.46, CI = 1.02–2.03). The study authors also reported a slight increase (SIR = 1.77, CI = 0.88−3.17) in astrocytomas between the observed and expected incidences in both sexes for children under age 5 (not statistically significant). Similar findings for astrocytomas were also found in Dover

Township (SIR = 1.91, CI = 0.21−6.91) in both sexes for children under age 5. Additional findings in Dover Township included: (1) a statistically significant increase for total childhood cancer incidence, especially in females under the age of 5 (SIR = 1.90, CI = 1.09–3.09; see Table A-2), (2) a statistically significant increase in the incidence of leukemia in females from ages 0−19 (SIR = 1.99, CI = 1.06–3.40) and 0−4 (SIR = 2.65, CI = 1.06–5.45), and (3) a statistically significant increase in the incidence of acute lymphocytic leukemia in females from ages 0−19 (SIR = 2.59, CI = 1.34–4.53) and ages 0−4 (SIR = 3.27, CI = 1.31–6.73). In the Toms River section of Dover Township, the total childhood cancer incidence was statistically significantly elevated, most notably in females under the age of 5 (SIR = 6.17, CI = 2.95–11.34; see Table A-3). Additionally, the incidence of brain and central nervous system cancer was statistically significantly higher in children of both sexes (SIR = 7.04, CI = 1.89–18.03) and females only (SIR = 11.6, CI = 2.33–33.88), both under the age of 5. Astrocytomas were increased in children of both sexes (SIR = 9.47, CI = 1.06–34.19) below the age of 5, and elevated leukemia incidence (SIR = 7.84, CI = 2.11–20.06) and acute lymphocytic leukemia (SIR = 9.68, CI = 2.60–24.78) were both observed in females. A time-trend analysis for the State of New Jersey, Ocean County, Dover Township, and the Toms River section of Dover Township provided limited evidence of differences in total cancer rates between the State and Ocean County throughout the period of analysis. During the mid to late 1980s, increased incidences for all combined cancers, leukemia, and brain/central nervous system cancers were reported for Dover Township. From 1988−1990, there was a notable increase in cancer incidence for the Toms River section of Dover Township.

The study authors concluded that these data support the findings of increased incidence of childhood brain cancer in the Dover Township and the Toms River section of Dover Township previously reported by an earlier analysis (NJDOH, 1996) (not publicly available). Most notably, statistically significantly increased incidences of leukemia and brain/central nervous system cancer were observed, particularly in females under the age of 5.

NJ DHSS (2003a)

In response to their previous findings (NJ DHSS, 1997), the NJ DHSS (2003a) conducted a case-control epidemiologic analysis of childhood cancers in Dover Township and the Toms River section of Dover Township on individuals exposed during the period between 1979 and 1996. During the study, water distributions and air plumes originating from the Ciba-Geigy Superfund site and the Reich Farm Superfund site were modeled. Individuals were potentially exposed to contaminants through air and by drinking water provided to the community from a well system. The United Water Toms River (UWTR) Parkway and the UWTR Holly Street well fields served this community, and there was also potential contamination of private wells due to releases from both Superfund sites. Early in the course of evaluating sources of contamination attributable to the Reich Farm site, Richardson et al. (1999) performed a chromatographic analysis of drinking water samples from the UWTR Parkway well field in 1996. The study authors determined that three to four of the highest chromatographic peaks were unknown compounds later determined to be isomers of SAN Trimer (ATSDR, 1998).

There were two components to the case-control epidemiologic study. First, four controls were identified for each case, matched by age, sex, and residence yielding 40 cases and 159 controls in the Interview Study. Spatial location and other information were gathered at diagnosis ± 1 year. In the second part of the study, 10 controls were identified per case, yielding 48 cases and 480 controls in the Birth Records Study.

The relative risk (odds ratio) of childhood cancers was calculated using conditional logistic regression to evaluate the degree to which exposure factors were associated with disease. An odds ratio (OR) greater than one means that the exposure factor was more common in cases than controls. ORs were calculated for two groups: children diagnosed before age 5 and children diagnosed before age 20. For the Interview Study, ORs were calculated for leukemia and nervous system cancers, leukemia alone, all nervous system cancers, brain and central nervous systems cancers, and all other cancers. The Birth Records Study was meant to address people who were exposed to contaminants in drinking water from the UWTR well fields but had since moved away. Cancer incidences in children that moved out of the study area prior to diagnosis were identified using cancer registries in ten states, with roughly 70% of these children moving to states with cancer registries. A source of confounding in this study is that the groundwater contained several contaminants in addition to SAN Trimer, including trichloroethylene and tetrachloroethylene that are both known to demonstrate carcinogenic potential. Specifically, trichloroethylene and tetrachloroethylene have been classified by IRIS as *carcinogenic to humans* and *likely to be carcinogenic in humans*, respectively (IRIS, 2012, 2011), and both chemicals have been classified by the International Agency for Research on Cancer (IARC) as *probably carcinogenic to humans* (IARC, 1997). There were also additional sources of air emissions and lifestyle factors that potentially confound the analysis; these factors were only partly addressed through matching of controls. Furthermore, the results from the case-control study should be interpreted with caution based on the study authors' inference that "Due to the relatively small number of study subjects, the analyses are sensitive to random fluctuations in numbers, which can result in substantial imprecision in the odds ratios (reflected by wide confidence intervals)."

Three exposure categories (high, medium, and low) were created based on the average percent of public water delivered from each of the aforementioned well fields. When prenatal, time-specific, UWTR Parkway well field, high-level exposures were analyzed separately by sex, the study authors found an association (OR = 5.0, 95% CI = 0.8−31) for prenatal exposures and leukemia in females 0−19 years old. For females with leukemia, the prenatal, time-specific, UWTR Parkway well field, high-exposure category was also significantly associated (OR = 6.0, 95% CI = 1.1−32). Because of uncertainty as to when the contaminants reached the wells, the exposure period was broken into several windows. The new windows were 1978−1996, 1979−1996, 1981−1996, 1983−1996, 1984−1996, 1985−1996, and 1986−1996. For the period of 1984−1996, the OR for exposure to UWTR Parkway well water and the development of leukemia in females was 15 (95% CI = 0.8−274). The Birth Records Study showed no pattern or consistency of association with childhood cancers. Exposure to unadjusted UWTR Holly Street well field water or various time-specific periods of exposure to Holly Street well field water did not appear to be associated with any childhood cancers in either the Birth Records or the Interview Studies. Very few children lived in residences with private groundwater wells in any of the groundwater regions, and no association was seen between leukemia and brain and central nervous system cancer groupings in either the Interview or the Birth Record Studies.

There was also an association with Ciba-Geigy air emissions that appeared only with leukemia in females from ages 0−4 in high- (75th percentile), medium- (50th percentile), or low-exposure categories. The OR = 19 for the high-exposure category (95% CI = 0.9−397); for the medium-exposure category the OR = 5.2 (95% CI = 0.5−57) relative to the lowest exposure category. No associations were found in males for overall leukemia or central nervous system cancers. There were also no associations with air emissions from the Oyster Creek Nuclear plant. Associations were seen for proximity to the Ciba-Geigy pipeline and leukemia for both sexes combined and females only during the prenatal and postnatal period (OR range = 2.3−14). For the Birth Records Study, only the prenatal period was evaluated, and it was assumed that the residence at birth was also the residence throughout pregnancy. As in the Interview Study, elevated ORs were found in the Birth Records Study for the high- and medium-exposure categories of Ciba-Geigy ambient air emissions for prenatal exposure in females diagnosed with leukemia prior to age 5 (high-exposure category OR = 7.8, 95% CI = 0.8−77; medium-exposure category OR = 2.0, 95% CI = 0.1−35).

Exposure to UWTR Parkway well field water containing SAN Trimer was associated with a statistically significant increase in childhood leukemia. No association was seen with other public water supply fields. There was also an association with Ciba-Geigy air emissions that appeared only with leukemia in females. Because of the small number of cases, the NJ DHSS (2003a) study lacks the power to detect statistically significant findings (i.e., most 95% CIs overlap 1.0).

A groundwater treatment system (i.e., air stripper) had been installed at the Parkway well field in the mid-1980s to remove the known contaminants but not SAN Trimer; the treated water was used for drinking. In November 1996, wells contaminated with SAN Trimer were removed as sources of drinking water. Beginning in May 1997, filtration systems began to be installed on drinking water wells to specifically remove SAN Trimer contamination.

NJ DHSS (2003b)

In response to a public comment draft of the report titled *Case-control Study of Childhood Cancers in Dover Township (Ocean County), New Jersey* (NJ DHSS, 2003a), the NJ DHSS and ATSDR analyzed five further years (1996−2000) of childhood cancer incidence data in Dover Township and the Toms River section of Dover Township to identify any differences in incidence and time trends of childhood cancer (NJ DHSS, 2003b). In this non-peer-reviewed report, no statistically significant increases in the incidences of childhood cancers in either Dover Township or the Toms River section of Dover Township were observed during the 5-year period. Furthermore, the study authors stated that SIRs for childhood cancer incidences in Dover Township and the Toms River section of Dover Township appeared to be decreasing. A time-trend analysis for the 22-year period (1979−2000) determined that the incidence of all combined childhood cancers for the State of New Jersey increased slightly through the period but then dropped in the late 1990s. The incidence rates for Dover Township increased compared to those of the State from 1985−1995 and in the latter part of the 1990s. The childhood cancer incidence rates for the Toms River section of Dover Township were the highest and the most variable during the late 1980s. The study authors concluded that these results should be interpreted with caution due to the small number of cases that were diagnosed during the most recent 5-year period (1996−2000) (NJ DHSS, 2003b).

NJ DHSS (2008)

In an additional non-peer-reviewed analysis (NJ DHSS, 2008), the cancer registry data from the (NJ DHSS, 1997) study were updated for the period from 2001−2005, thus extending the period of analysis for childhood cancers from 1979−2005. During the 5-year interval from 2001−2005, there were 26 additional cancers among children of Toms River Township (formerly Dover Township) and five in the sub-Township (formerly the Toms River section of Dover Township). The most frequently diagnosed cancers were brain/CNS, leukemias, and soft-tissue sarcomas (which were statistically significantly increased from 2004–2005 in females in both the Toms River Township [SIR = 5.5; 95% CI = 1.5−14] and the sub-Township area [SIR = 21; 95% CI = 4.2−62]). Compared to the expected incidences based on State levels, childhood cancer incidence in females was not significantly increased (as measured by SIRs), while leukemia incidence was lower than expected. The total cancer incidence in males was similar to background levels. Trend analysis for the 1979−2005 period showed increased total childhood cancer, brain/CNS cancer, and leukemia incidence from the mid 1980s to mid 1990s for female children in Toms River Township.

In conclusion, the initial epidemiologic study reported statistically significant increases in childhood cancer incidences from 1979−1995 (NJ DHSS, 1997). In the case-control study (NJ DHSS, 2003a), the study authors observed a statistically significant association for leukemia in females (0−19 years of age) and exposure to drinking water from well fields containing multiple chemicals, including SAN Trimer, some of which are known to demonstrate carcinogenic potential (e.g., trichloroethylene and tetrachloroethylene). The results from the case-control study should be interpreted with caution because of the small number of study subjects as described above. Following cleanup and removal of SAN Trimer along with the other chemicals (e.g., trichloroethylene and tetrachloroethylene) from the drinking water, total childhood cancer and brain/CNS cancer incidence have returned to background levels, and fewer than expected cases of leukemia have been observed (see Figures 2 and 3 and Tables A-2 and A-3). However, due to the existence of other contaminants in drinking water from the well fields investigated, the conclusions that can be drawn from the epidemiologic studies are relatively limited.

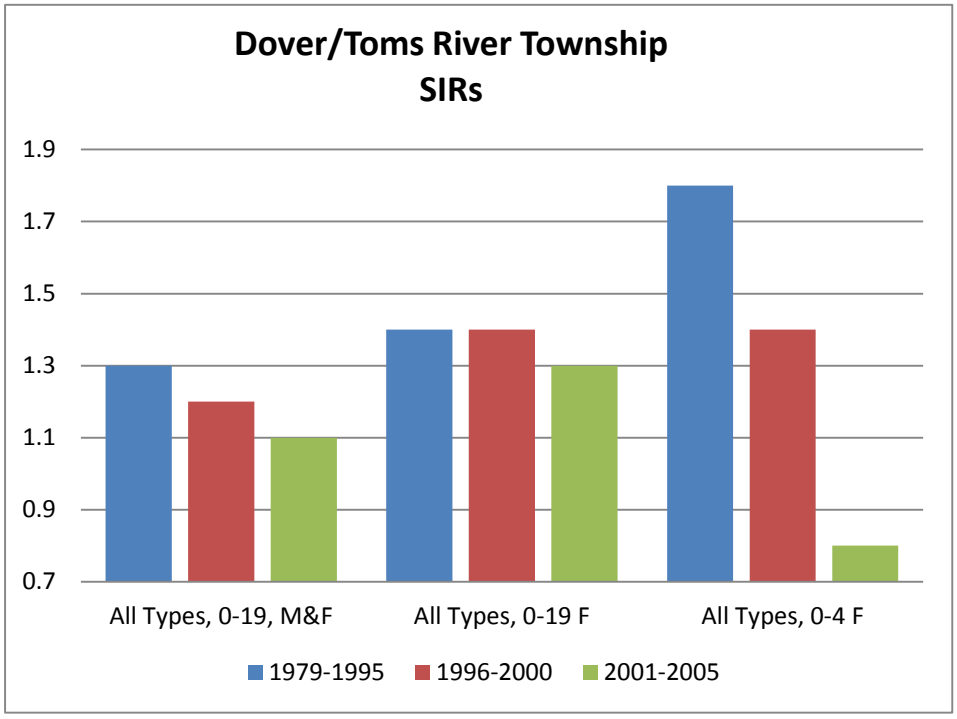

Figure 2. Standardized Incidence Ratios (SIRs) of Cancer (All Types) in Three Populations during Three Time Intervals from 1979−2005

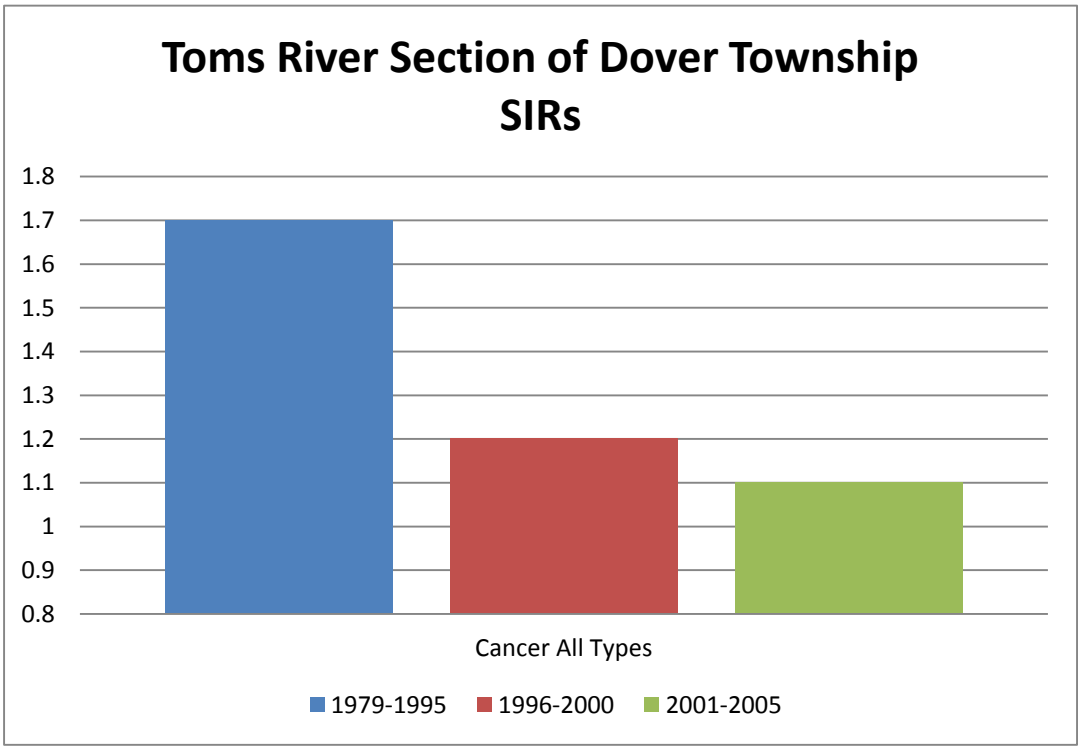

Figure 3. Standardized Incidence Ratios (SIRs) of Cancer (All Types) in Males and Females (0−19 years) during Three Time Intervals from 1979−2005

Inhalation Exposures

No studies have been identified.

ANIMAL STUDIES

Oral Exposures

The effects of oral exposure of rats to SAN Trimer have been evaluated in one acute study (Huntingdon Life Sciences, 1999a), two short-term-duration studies Huntingdon Life Sciences (1999b); NTP (2012), one subchronic-duration study (NTP, 2012), and one chronic-duration study (NTP, 2012). No developmental or reproductive toxicity studies were identified.

Acute Studies

Huntingdon Life Sciences (1999a)

In a non-peer-reviewed Good Laboratory Practice (GLP) study conducted by Huntingdon Life Sciences (1999a) for the Union Carbide Corporation that was submitted to the U.S. EPA, male and female S-D rats (5/sex/dose) were treated with SAN Trimer (98% purity) in corn oil by a single gavage administration at doses of 0, 250, 500, or 1,000 mg/kg. In determination of the LD_{50}, there were 2/10 deaths at the 250 mg/kg dose, 4/10 deaths at the 500 mg/kg dose, and 9/10 deaths at the 1,000 mg/kg dose. All animals that died did so within 72 hours, and the surviving rats were observed for 14 days. The study authors concluded that the LD_{50} was 440 mg/kg for males and 590 mg/kg for females at both 72 hours and 14 days.

Short-Term-Duration Studies

Huntingdon Life Sciences (1999b)

In a non-peer-reviewed report, Huntingdon Life Sciences (1999b) conducted a 14-day study for the Union Carbide Corporation in which Sprague-Dawley (S-D) rats (6/sex/dose) were treated daily with 0, 30, 75, 150, or 300 mg/kg-day SAN Trimer (purity 98%) by gavage in corn oil. At 300 mg/kg-day all the males and 5/6 females died. Health effects evaluated included standard hematology (hemoglobin concentration, hematocrit, erythrocyte count, platelet count, mean corpuscular volume, mean corpuscular hemoglobin, total leukocyte count, differential leukocyte count, absolute lymphocyte count, absolute segmented neutrophil count, prothrombin time, activated partial thromboplastin time, and reticulocyte count), standard clinical chemistry (aspartate aminotransferase, alanine aminotransferase, alkaline phosphatase, lactate dehydrogenase, sorbitol dehydrogenase, blood urea nitrogen, creatinine, glucose, creatine kinase, total protein, albumin, globulin, albumin/globulin ratio, total bilirubin, direct bilirubin, indirect bilirubin, sodium, potassium, chloride, calcium, inorganic phosphorus, gamma-glutamyl transferase), urinalysis (specific gravity, appearance, osmolality, protein, glucose, ketones, occult blood, pH, bilirubin, urobilinogen, creatinine, and *N*-acetyl-B-D-glucosaminidase), body weights, food consumption, and organ weights (testes, ovaries, heart, liver, and kidney). In addition, adrenal glands, aorta (thoracic), bone (sternum/femur with articular surface), bone marrow (sternum/femur), brain (medulla/pons, cerebrum, and cerebellum), esophagus, eyes (with optic nerve), heart, kidneys, large intestine (cecum, colon, rectum), lacrimal glands, liver, lungs (with mainstem bronchi), lymph nodes (mesenteric), mammary glands, muscle (Biceps femoris), nerve (sciatic/tibial/sural), ovaries, pancreas, pituitary, prostate, salivary gland (submandibular), seminal vesicles, skin, small intestine (duodenum, ileum, and jejunum), spinal cord (cervical, thoracic, lumbar), spleen, stomach, testes with epididymides, thymic region, thyroid (with parathyroid), trachea urinary bladder, uterus (body/horns with cervix), and Zymbal's gland. Macroscopic lesions were preserved and examined microscopically.

Food consumption was statistically significantly decreased at 150 mg/kg-day in males after one week of treatment. Among the hematologic endpoints there were statistically significant (approximately 10%) decreases in hemoglobin, hematocrit, and red blood cells at 150 mg/kg-day in females, while in the one surviving female treated with 300 mg/kg-day, these same parameters showed a decrease of 20−30%. There was a statistically significant decrease (approximately 20%) in mean activated partial thromboplastin time (a measurement of the time it takes for blood to clot) in the 150 mg/kg-day males compared to control values. The mean activated partial thromboplastin time for the single surviving female treated with 300 mg/kg-day was also approximately 20% lower than the control mean. Globulin was statistically significantly decreased at 30, 75, and 150 mg/kg-day in females, but the study authors did not consider this biologically significant due to the "absence of any evidence of systemic toxic changes at the lower doses." No significant findings were observed in the urinalysis. Relative liver weight was statistically significantly increased in males at 150 mg/kg-day. Absolute and relative liver weights were statistically significantly increased at 150 mg/kg-day in females. Mean absolute and relative heart weights were also statistically significantly increased at 150 mg/kg-day in females. Other organ weights were similar to those of the control rats. The histopathological analysis showed that 4/6 males had vacuolation of periacinar hepatocytes and 3/6 females had periacinar hypertrophy of the liver in the 300 mg/kg-day-dose group. In addition, 4/6 males in the 300 mg/kg-day-dose group had vacuolation of cortical tubular epithelial cells in the kidney, and the study authors indicated there was no evidence of alpha 2u-globulin involvement (although it is unclear if immunostaining was performed).

In summary, a number of effects of SAN Trimer treatment were observed in this short-term-duration study. The study authors reported dose-related effects indicative of toxicity including decreased hematocrit and erythrocyte counts, decreased activated partial thromboplastin time, and elevated liver and heart weights. A LOAEL of 75 mg/kg-day with a corresponding NOAEL of 30 mg/kg-day is identified for this study based on increased relative liver weight (>10% change compared to control values) in male rats.

NTP Studies

In a study conducted by Batelle Columbus Operations (Columbus, OH) for the NTP (2012), male and female F344/N rats were treated with SAN Trimer in feed in perinatal and postnatal studies for 7 weeks, 18 weeks, or 2 years. The results of these studies are also published in a peer-reviewed manuscript by Behl et al. (2013). SAN Trimer (Batch 3, 96% purity) was used in all of the NTP studies. As indicated in the NTP (2012) study, "Animals in the 7-week, 18-week, and 2-year studies were exposed to the test chemical for 2 weeks during their in utero development, for 3 weeks through their mother's milk plus consumption of dosed feed, and for 2 weeks, 13 weeks, and 2 years after weaning through dosed feed." The durations of these studies are classified for this PPRTV assessment based on the time that F1 rats were directly treated with SAN Trimer in the diet (i.e., 2 weeks, 13 weeks, and 2 years) as depicted in Figure 4.

For the 2-week, 13-week, and 2-year studies, male and female F344/N rats (30 males and 60 females for the 2- and 13-week studies, 100 males and 200 females for the 2-year study) were mated during a 7-day breeding period (9 days for the 2-year study). During breeding, Gestation Day (GD) 1 was designated upon the presence of a vaginal plug or the identification of sperm in vaginal lavage fluid. Beginning on GD 7, the study authors administered dietary doses of SAN Trimer in NIH-07 feed to groups of pregnant females (F0) through delivery (dosimetry

calculations for the gestational component are presented in Appendix C). Groups of females that birthed litters were treated with the same concentrations in the diet from Postnatal Day (PND) 1 to PND 20 of the last litter delivered (dosimetry calculations for the lactational component are presented in Appendix C). Water and food were provided ad libitum. Dams were monitored twice daily and observed for labor beginning at GD 18 up to 24. PND 0 was designated as the day that litters were delivered. The study authors reported body weights and clinical findings observed in dams on GDs and PNDs 1, 7, 14, and 20 (GD 18 for 2-week study). Pups (F1) were evaluated (number, weight, and sex) on PNDs 1, 4 (except for the 2-year study), 7, 14, and 20. Pups from mated pairs were culled to eight pups (maximum) for the 2- and 13-week studies and 10 pups for the 2-year study with equal numbers of males and females per pair; culled weanlings were removed and not examined further. Remaining pups were reevaluated on PND 20, and two males and females per litter were chosen to be treated for the remainder of the study (2- and 13-week studies); the dams of these selected pups were necropsied at this time. For the 2-year study, three pups/sex/litter were randomly chosen for the core study (2-year treatment) and two pups/sex/litter were selected for special-study groups for hematology, clinical chemistry, or urinalysis evaluation (treated up to 78 weeks) on PND 18. Necropsy results of the dams were not provided by the study authors. Weaning of the F1 rats occurred on PND 21. F1 rats were treated with the same concentrations of SAN Trimer in the diet (NTP-2000) as their respective dams beginning on PND 21 until study termination. It is assumed that pups received 100% of the dose given to their respective dams during lactation.

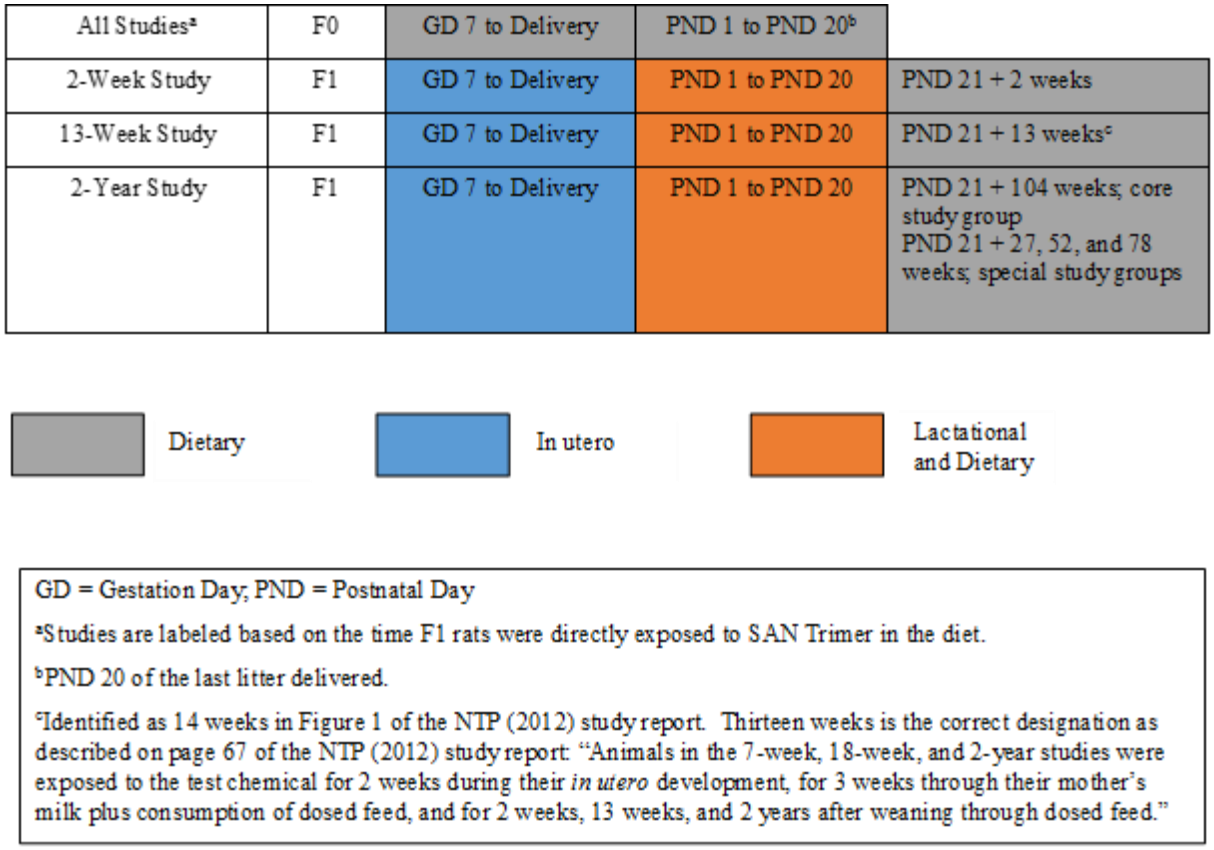

Figure 4. Study Design for the 2-Week, 13-Week, and 2-Year Perinatal and Postnatal Feed Studies of SAN Trimer (NTP, 2012)

NTP (2012) 2-Week Study

In the 2-week study, groups of 7 or 8 pregnant female F344/N rats (F0) were fed diets containing 0, 250, 500, 1,000, 2,000, or 4,000 ppm SAN Trimer (equivalent to 0, 18, 35, 67, 130, or 197 mg/kg-day determined for this PPRTV assessment by calculating the time-weighted average doses provided by the study authors) from GD 7 to delivery of pups. Of the dams with litters, groups of 6−8 females were further treated with the same concentrations in the diet as gestation from PND 1 to PND 20 (equivalent to 0, 40, 85, 166, 325, or 634 mg/kg-day as calculated for this PPRTV assessment). Doses for F1 pups were calculated with the assumption that they received 100% of the dose given to their respective dams. Of the 60 F0 female rats that were mated during the 7-day breeding period, 46 were identified to be sperm positive and 44 gave birth to live pups on GD 23 or GD 24. The two dams that did not give birth were determined to not be pregnant or had unsuccessful pregnancies. Of the 44 dams that gave birth, 95% produced litters of 3 or more pups. Animals were monitored and data collected as described above. No apparent effects of SAN Trimer were observed on gestation length, fertility, litter size, live birth/implantation ratio, or sex number. At the highest dose tested (197 mg/kg-day during gestation and 634 mg/kg-day during lactation), dams at GD 14 and 18 and PNDs 1, 7, 14, and 20 showed statistically significant decreases in body weight (see Table A-4). Food consumption was decreased in dams at the two highest doses (130 and 197 mg/kg-day) from GDs 7−14. From PNDs 8−15 and 15−20, food consumption was reduced

(30% lower compared to controls) in dams at the high dose (634 mg/kg-day). Statistical evaluation was not performed for food consumption data by the study authors. For F1 male and female pups, body weight was statistically significantly reduced at the highest dose (634 mg/kg-day) from PNDs 1 to 14 and at 325 mg/kg-day on PND 20 (see Table A-5). From this study, a maternal LOAEL of 197 mg/kg-day is identified during gestation with a corresponding NOAEL of 130 mg/kg-day for statistically significantly decreased body weight in F0 dams on GD 14. Also, a maternal LOAEL of 634 mg/kg-day is established during lactation with a NOAEL of 325 mg/kg-day based on statistically significantly decreased body weight in dams. For F1 pups, a LOAEL of 325 mg/kg-day (NOAEL of 166 mg/kg-day) is identified for statistically significantly decreased body weight as measured at PND 20.

Groups of F1 rats (10/sex/dose) from the treated dams were fed diets containing 0, 250, 500, 1,000, 2,000, or 4,000 ppm SAN Trimer (equivalent to 0, 50, 90, 175, 270, or 410 mg/kg-day in males and 0, 45, 90, 185, 295, or 430 mg/kg-day in females, calculated by the study authors) for 2 weeks postweaning (PND 21 to PND 35). Food consumption was recorded on Days 1, 4, 8, 11, and at the end of the study. The heart, kidney, liver, lung, spleen, thymus, and uterus were weighed at study termination. No clinical chemistry or hematologic analyses were conducted. Histopathology was conducted on brain, kidney, liver, spleen, epididymis, prostate, seminal vesicle, testis, ovary, and uterus where applicable. Body weights were statistically significantly reduced in 175-, 270-, and 410-mg/kg-day males and 295- and 430-mg/kg-day females at study termination. At PND 35, body weight reductions in females exceeded 10% at both 295 and 430 mg/kg-day. However, food consumption was also reduced (not statistically significant) in the aforementioned dose groups. There was a dose-related decrease in food consumption that was less than half of the control at the high dose. Observed organ weight differences (see Table A-6) included: (1) significant decreases in absolute heart (≥175 mg/kg-day [M]; ≥295 mg/kg-day [F]), right kidney (≥270 mg/kg-day [M]; ≥295 mg/kg-day [F]), liver (≥270 mg/kg-day [M]; ≥295 mg/kg-day [F]), lung (≥270 mg/kg-day [M]; ≥295 mg/kg-day [F]), and spleen (≥410 mg/kg-day [M]; ≥430 mg/kg-day [F]); (2) significant decreases in absolute and relative thymus weight in males (≥90 mg/kg-day) and (≥295 mg/kg-day) females, right testis weight in males (≥90 mg/kg-day), and uterus weight in (≥295 mg/kg-day) females; and (3) significant increases in relative heart (≥410 mg/kg-day [M]; ≥430 mg/kg-day [F]), right kidney (≥410 mg/kg-day [M]; ≥430 mg/kg-day [F]), liver (≥175 mg/kg-day [M]; ≥185 mg/kg-day [F]), lung (≥410 mg/kg-day [M]; ≥430 mg/kg-day [F]), and spleen (≥270 mg/kg-day [M]; ≥295 mg/kg-day [F]). Nonneoplastic lesions were observed in the brain, thymus, spleen, liver, kidney, and reproductive organs of both sexes, but the study authors characterized these lesions as not toxicologically relevant and considered them secondary to severe calorie reduction; the dose levels at which these lesions occurred were not specifically identified by the study authors. There was, however, increased cellularity in the cerebrum at the level of the septal nuclei (incidences of 0/10, 0/9, 0/6, 0/3, 0/5, and 4/6 in males and 0/5, 0/3, 0/2, 0/3, 1/5, and 10/10 in females). The increased cellularity could be due to a developmental delay caused by inadequate caloric intake, but other mechanisms cannot be ruled out.

A LOAEL of 90 mg/kg-day is established for statistically significantly decreased relative right testis and thymus weights in the male F1 offspring with a corresponding NOAEL of 50 mg/kg-day.

Subchronic-Duration Studies

NTP (2012) 13-Week Study

Beginning on presumed GD 7, the study authors administered dietary (NIH-07) doses of 0, 100, 200, 400, 800, and 1,600 ppm (equivalent to 0, 6.9, 14, 28, 55, or 110 mg/kg-day as calculated for this PPRTV assessment)[1] of SAN Trimer to groups of 8 or 9 sperm-positive females (F0). Groups of 4−8 females that birthed litters were treated with the same concentrations (equivalent to 0, 17, 33, 66, 132, or 264 mg/kg-day as calculated for this PPRTV assessment)[1] up to PND 20 of the last litter delivered. Doses for F1 pups were calculated with the assumption that they received 100% of the dose given to their respective dams. Of the 60 F0 female rats that were mated during the 7-day breeding period, 50 were sperm positive and 39 gave birth to live pups on GD 23 or GD 24. For the females that did not give birth, it was unclear whether they were not impregnated or the lack of delivery was due to resorption. Of the dams that produced litters, only one (treated with 110 mg/kg-day) delivered less than five pups. One pup died in the control group prior to PND 4; no cause of death was provided by the study authors. A litter of 8 pups in the low-dose group was sacrificed moribund after the dam died due to chylothorax (leakage of lymphatic fluid of intestinal origin into the pleural space). Besides culling that occurred on PND 4, all remaining pups survived through weaning. The study authors reported an effect of SAN Trimer on fertility index, number of litters, and litter size in dams treated with 110 mg/kg-day of the chemical. The data for fertility index was determined not to be statistically significant based on Fisher's Exact test, and no variance information was provided for number of litters and litter size to perform statistical analysis.

The study authors reported statistically significant decreased body weight on GD 14 in dams treated with 28, 55, and 110 mg/kg-day of SAN Trimer (see Table A-7). Body weight was statistically significantly reduced in dams in the high-dose group (110 mg/kg-day) on GD 20. No body-weight changes were observed in male pups during lactation. Body weight in female pups was statistically significantly increased on PND 20 at 17 mg/kg-day (see Table A-8); the biological relevance of increased body weight is unknown. A maternal LOAEL of 110 mg/kg-day is identified with a corresponding NOAEL of 55 mg/kg-day for statistically significantly decreased body weight in dams observed during gestation. A NOAEL of 264 mg/kg-day is identified during lactation based on lack of effects observed in dams or pups. Because 264 mg/kg-day is the highest dose tested, a LOAEL for lactational effects cannot be determined.

Postweaning, F1 rats (10/sex/dose) were treated for 13 weeks with the same dietary (NTP-2000) concentrations of SAN Trimer (equivalent to 0, 10, 20, 40, 80, or 150 mg/kg-day as calculated by the study authors) as the F0 generation. Water and food were provided ad libitum. Rats were monitored twice daily. The study authors reported body weights and clinical findings at the beginning of treatment, weekly, and at study termination. Food consumption was reported biweekly until the end of the study. At the end of the study, urine samples were collected and the following parameters were included in the urinalysis: creatinine, glucose, protein, alkaline phosphatase, aspartate aminotransferase, *N*-acetyl-β-D-glucosaminidase, volume, and specific gravity. The study authors collected blood samples from the retro-orbital sinus for measurement of the following clinical chemistry parameters: urea nitrogen, creatinine, glucose, sodium,

[1]Dose = Feed Concentration × Food Consumption per Day × (1 ÷ Body Weight) × (Days Dosed ÷ Total Days), where the food factor was calculated from the body weight and food consumption data reported in dams from the 2-week NTP (2012) study.

potassium, chloride, calcium, phosphorus, total protein, albumin, total bilirubin, cholesterol, triglycerides, alanine aminotransferase, alkaline phosphatase, aspartate aminotransferase, creatine kinase, lactate dehydrogenase, sorbitol dehydrogenase, γ-glutamyl transferase, and bile acids. The following parameters were evaluated for hematology: hematocrit, hemoglobin concentration, erythrocyte, reticulocyte, platelet counts, Howell-Jolly bodies, mean cell volume, mean cell hemoglobin, mean cell hemoglobin concentration, leukocyte count and differentials, and activated partial thromboplastin time. To assess sperm motility, sperm samples were taken from male rats treated with 0, 40, 80, and 150 mg/kg-day of SAN Trimer. The following parameters were measured to evaluate sperm motility: spermatid heads per testis and per gram testis, spermatid counts, and epididymal spermatozoa motility and concentration. The left cauda, epididymis, and testis were weighed. To evaluate vaginal cytology, samples were collected for up to 12 days before study termination from females in the 0, 40, 80, and 150 mg/kg-day dose groups. Estrous cycles were evaluated to determine the probability of extended estrus, diestrus, and metestrus. After 13 weeks of direct exposure to SAN Trimer in the diet, F1 rats were necropsied. The study authors collected and weighed the following organs: brain, heart, right kidney, liver, lung, spleen, right testis, thymus, and uterus. Complete histopathological examinations for the 0 and 150 mg/kg-day-dose groups were performed on the following organs: adrenal gland, bone with marrow, brain, clitoral gland, esophagus, eye, Harderian gland, heart and aorta, large intestine (cecum, colon, rectum), small intestine (duodenum, jejunum, ileum), kidney, liver, lung, lymph node (mandibular and mesenteric), mammary gland, nose, ovary, pancreas, parathyroid gland, pituitary gland, preputial gland, prostate gland, salivary gland, skin, spleen, stomach (forestomach and glandular), testes with epididymis and seminal vesicle, thymus, thyroid gland, trachea, urinary bladder, and uterus. Tissues were fixed and preserved, embedded in paraffin, sectioned, and stained with hematoxylin and eosin.

Survival was 100% for F1 rats that were directly exposed to SAN Trimer in the diet for 13 weeks. Body and organ weight data are presented in Table A-9. Terminal body weight was statistically significantly decreased in male rats at the high dose. The study authors reported statistically significantly decreased body weight in female rats at ≥20 mg/kg-day. Increased absolute and relative liver weights were statistically significant at all dose levels in male rats. Increased relative liver weight was statistically significant at ≥40 mg/kg-day in female rats. Absolute spleen weight in male rats was statistically significantly increased at the two highest doses and at the high dose in females. Relative spleen weight was statistically significantly increased at ≥40 mg/kg-day in male rats and at the two highest doses in females. Increased relative brain weight was statistically significant at the high dose in males and at doses ≥20 mg/kg-day in females. Statistically significantly increased relative right testis weight was reported in males at the high dose. Absolute right testis and left testis weights were statistically significantly decreased in males at the high dose. Absolute heart weight was statistically significantly increased at 10, 40, and 80 mg/kg-day in male rats. Increased relative heart weight in male rats was statistically significant at all doses and at the two highest doses in females. Increased relative kidney weight in male and female rats was reported to be statistically significant in the two highest dose groups. The study authors reported that no histopathologic lesions due to SAN Trimer were observed in the organs examined for both male and female rats. Exposure to SAN Trimer had no statistically significant effect on sperm parameters in males or on the estrous cyclicity in female rats.

The hematology, clinical chemistry, and urinalysis findings are summarized in Table A-10. In high-dose male rats, erythrocytes and hemoglobin were statistically significantly decreased (both 3−4% below control) while mean cell volume was statistically significantly increased (2% above control). There were no statistically significant changes in hematology in female rats at any dose level. Serum albumin was statistically significantly increased in males in the two highest dose groups. Serum total protein was statistically significantly increased in males in the 80 mg/kg-day-dose group, but not in males of the highest dose group. There was statistically significantly reduced serum cholesterol in male rats at the high dose. Serum triglycerides were statistically significantly decreased at 40, 80, and 150 mg/kg-day in male rats and 80 and 150 mg/kg-day in females. In males, alanine aminotransferase was statistically significantly decreased starting at 20 mg/kg-day. Aspartate aminotransferase was statistically significantly reduced at all dose levels in males and at 80 and 150 mg/kg-day in females. There was a statistically significant decrease in bile acids at 40 mg/kg-day in males and at 10 and 80 mg/kg-day in females. In female rats, the following statistically significant changes were also observed in clinical chemistry parameters: increased creatinine (20−150 mg/kg-day) and elevated urea nitrogen (150 mg/kg-day). Urinalysis revealed the following statistically significant changes in male rats: increased glucose (80 mg/kg-day), elevated glucose/creatinine ratio (80 and 150 mg/kg-day), increased protein (80 and 150 mg/kg-day), increased protein/creatinine ratio (150 mg/kg-day), aspartate aminotransferase (decreased at 80 mg/kg-day and increased at 150 mg/kg-day), and aspartate aminotransferase/creatinine ratio (decreased at 80 mg/kg-day and elevated at 150 mg/kg-day). Urinalysis in female rats revealed that the alkaline phosphatase/creatinine ratio was statistically significantly decreased at the high dose and *N*-acetyl-β-creatinine-D-glucosaminidase/creatinine ratio was statistically significantly decreased in the two highest dose groups.

Based on statistically significantly increased absolute and relative heart weight in F1 male rats, a LOAEL of 10 mg/kg-day is identified. Because 10 mg/kg-day is the lowest dose tested, a NOAEL cannot be determined.

Chronic-Duration Studies

NTP (2012) 2-Year Study

In the chronic-duration study by NTP (2012), 41 or 42 pregnant females per dose group were treated with 0, 400, 800, or 1,600 ppm (equivalent to 0, 28, 55, or 110 mg/kg-day as calculated for this PPRTV assessment)[1] via the diet from GD 7 until delivery. Of the female rats that were mated during the 7-day breeding period, 83% were determined to be sperm positive and 52% gave birth to at least one live pup; those pups were treated with the same dietary concentrations (equivalent to 0, 66, 132, or 264 mg/kg-day as calculated for this PPRTV assessment)[1] through PND 20. Doses for F1 pups were calculated with the assumption that they received 100% of the dose given to their respective dams. There were no effects of SAN Trimer on the fertility index, gestational index, or number of litters during gestation and lactation. Four dams, whose offspring either died or were fostered to other litters, were sacrificed during the postnatal phase. The study authors reported that 45 pups died (contributions from all dose groups) from PND 1 to PND 20. However, the survival rate was greater than 92% for pups of all dose groups. The study authors provided no explanation as to why pups died during postnatal exposure. The study authors reported no changes in body weight in dams or F1 pups exposed from GD 7 to PND 20 (data not shown). Based on no effects observed in dams or pups,

NOAELs of 110 mg/kg-day (gestation) and 264 mg/kg-day (lactation) are identified. Because these are the highest doses tested, LOAELs for gestational and lactational effects cannot be determined.

Groups of F1 rats (50/sex/dose) were directly treated with SAN Trimer in the diet at doses of a 0, 20, 40, and 75/85 (males/females, respectively) mg/kg-day as calculated by the study authors for 2 years beginning at PND 21; these rats were referred to as the core study groups by the study authors. Groups of rats (20/sex/dose) were exposed to the same concentrations in the feed as the core study groups but were sacrificed at various time points up to 78 weeks in special study groups to identify effects on urinalysis, clinical chemistry, hematology parameters, and plasma concentrations of SAN Trimer. Rats were monitored twice daily for mortality and morbidity. The study authors recorded body weight and food consumption weekly for the first 13 weeks then monthly afterward; body weight was also documented at the conclusion of the study. The study authors also reported clinical observations at PND 29 and monthly, thereafter. At 23, 48, and 74 weeks postweaning, 3 rats/sex/dose along with 6 controls (both sexes) from the special study groups were switched to a SAN Trimer-free diet and blood was collected periodically up to 360 minutes after switching to SAN Trimer-free feed. During the Week 23 collection, male rats scheduled to be evaluated at 240 and 360 minutes were actually bled at 180 and 300 minutes, respectively. Plasma from these animals was analyzed by gas chromatography/mass spectrometry using 4-cyano-1,2,3,4-tetrahydro-a-methyl-1-naphthaleneacetonitrile (THNA) and 4-cyano-1,2,3,4-tetrahydro-1-naphthalene-propionitrile (THNP) as markers to determine concentrations of SAN Trimer. After blood was collected from these animals, they were further exposed to SAN Trimer in the diet. Urine was collected from 10 rats/sex/dose in the special study at postweaning weeks 26, 51, and 77 weeks. Urine samples were evaluated for the same parameters (except γ-glutamyltransferase) as the 13-week study described above. One week after urine collection (27, 52, and 78 weeks postweaning), the study authors collected blood from the retro-orbital sinus of 10 rats/sex/dose in the special study. These blood samples were analyzed for similar hematology and clinical chemistry parameters as described in the 13-week study. All core study rats and special study rats bled at Week 78 underwent complete necropsies. Complete histopathological examinations were performed on various organs for all F1 rats treated with SAN Trimer in the diet for 2 years. Organ weights were not examined in the 2-year study.

None of the plasma samples that were analyzed by gas chromatography/mass spectrometry contained SAN Trimer at concentrations above the limit of detection (0.400 µg/mL). Further experiments with 55 samples from the low- and high-dose groups and one control from the 74-week time point using limits of detection of 0.004 µg/mL for THNA isomers and 0.01 µg/mL for the THNP isomers determined that 14 of the 55 samples exceeded the limit for THNA. THNP was not above the limit of detection in any of the samples. Of the samples that contained THNA, the levels were below 0.0500 µg/mL in 13 of 14 samples; 9 of the 14 samples were less than 0.0052 µg/mL. The study authors stated that no correlation was seen between THNA concentration and exposure time.

After 2 years of treatment, there were no treatment-related effects on survival in F1 rats. Statistical analysis of these body-weight data was not provided by the study authors and cannot be performed due to the lack of variance information. There were no differences in food consumption among the treated groups compared to controls. The study authors reported

statistically significant changes in hematology, clinical chemistry, and urinalysis parameters (see Table A-11). These changes were not consistent and were transient; therefore, the biological significance of these effects is unclear.

In the histopathology evaluations, a number of nonneoplastic lesions and degenerative changes were observed and are shown in Table A-12. There was a statistically significant increase in the incidence of spinal nerve root degeneration in males in the high-dose group with an accompanying dose-related increase in severity. In female rats, there was a statistically significant increased incidence of sciatic nerve fiber degeneration at the two highest dose levels. In the livers of males, there was a statistically significant increased incidence of angiectasis, eosinic foci, and chronic active inflammation in the high-dose group. The incidence of mixed cell foci in the liver was statistically significantly increased in males in the low- and high-dose groups and in females at the high dose. There was a statistically significant increased incidence of bone marrow hyperplasia in both sexes (at 75 mg/kg-day for males and at 40 and 85 mg/kg-day for females). In the urinary bladder, there was a statistically significant increased incidence of hyperplasia of the transitional epithelium in females at the highest dose tested.

A LOAEL of 20 mg/kg-day is identified for this study based on a statistically significant increased incidence of mixed cell foci in the livers of male rats. Because 20 mg/kg-day is the lowest dose tested, a corresponding NOAEL cannot be identified.

Developmental Portion of the NTP (2012) Study

The NTP (2012) study did not examine any endpoints identified by the U.S. EPA's *Guidelines for Developmental Toxicity Risk Assessment* (U.S. EPA, 1991) to assess developmental toxicity.

Reproductive Portion of the NTP (2012) Study

The NTP (2012) study examined a limited number of reproductive parameters (e.g., fertility index) identified by the U.S. EPA's *Guidelines for Reproductive Toxicity Risk Assessment* (U.S. EPA, 1996) to assess reproductive toxicity; therefore, this study is not considered as a comprehensive reproductive study.

Carcinogenicity Studies

NTP (2012) 2-Year Cancer Results

The 2-year study by the NTP (2012) reported statistically significant changes in tumor incidences in F1 male and female F344/N rats. Tumor data for male and female rats are listed in Table A-13. In male rats, the incidences of total testicular interstitial cell adenomas (combined unilateral and bilateral) and bilateral interstitial cell adenomas were statistically significantly increased at the low (20 mg/kg-day) and high dose (75 mg/kg-day). An increase in combined brain and spinal cord astrocytomas also occurred in male rats, but this increase was not statistically significant at any dose level. The incidence of combined brain and spinal cord granular cell tumors were also increased in male rats (not statistically significant). There was a statistically significant decreased trend for pituitary gland adenomas, mononuclear cell leukemia in all organs, and malignant neoplasms in all organs examined in male rats. Pituitary gland adenomas were statistically significantly decreased at 75 mg/kg-day in male rats. There was a statistically significant decrease in mononuclear cell leukemia in all organs at all dose groups. Statistically significant decreased incidences of malignant neoplasms occurred at the mid (40 mg/kg-day) and high dose (75 mg/kg-day) in male rats. For female rats, the following

tumors occurred with a statistically significant negative trend: mammary gland tumors (classified as fibroadenomas; fibroadenomas or adenoma; fibroadenomas, adenoma, or carcinoma), pituitary gland tumors (i.e., adenomas and adenomas or carcinomas), and mononuclear cell leukemia in all organs, and neoplasms (benign and benign or malignant). Mammary gland tumor incidences were statistically significantly decreased at 40 and 85 mg/kg-day. Pituitary gland tumors in female rats were statistically significantly decreased in the low- and high-dose groups (20 and 85 mg/kg-day, respectively). The incidence of mononuclear cell leukemia in all organs was statistically significantly decreased at all dose levels. Malignant neoplasms (all organs) were statistically significantly decreased at 20 and 40 mg/kg-day in female rats. Benign and benign or malignant neoplasms incidences were statistically significantly decreased in female rats at the high dose. Based on the results of this study, the NTP (2012) report concludes that there is *no evidence of carcinogenic activity* for SAN Trimer.

Inhalation Exposure

No studies have been identified.

OTHER DATA

Metabolism Studies

Gargas et al. (2008)

Gargas et al. (2008) performed disposition studies to evaluate the pharmacokinetic behavior of SAN Trimer in rats. Nonpregnant F344/N female rats were given a single dose of [^{3}H]SAN Trimer via intravenous injection (26 mg/kg) or gavage (25, 75, or 200 mg/kg in corn oil), and pregnant and lactating rats were also administered a single gavage dose (200 mg/kg in corn oil); the use of a control group was not reported by the study authors. For nonpregnant rats, blood was taken at various time points ranging from 0.25 to 48 hours after SAN Trimer administration, and urine and feces were collected between 24 and 168 hours after dosing. For pregnant rats, the study authors sampled blood, placenta, fetus, and milk (collected from lactating rats) at a single time point 2 hours after gavage. Liquid scintillation spectroscopy and HPLC were performed to analyze the collected samples. The study authors determined that in nonpregnant rats, the elimination half-lives for [^{3}H]SAN Trimer following intravenous and gavage dosing were approximately 1 hour and 3.5 hours, respectively. For nonpregnant rats, C_{max} was achieved from 0.25 to 2 hours and was dose dependent following gavage administration of SAN Trimer, and was determined to be achieved 0.25 hours following an intravenous dose. SAN Trimer was rapidly excreted in the urine and feces after intravenous and gavage exposure. The study authors characterized the excreted radiolabel and determined that less than 1% of the radioactivity in urine and less than 3% in feces was from unchanged [^{3}H]SAN Trimer. For pregnant rats 2 hours post gavage, concentrations of radioactivity and SAN Trimer were highest in the blood, but the chemical was also detected in the placenta and fetus. In lactating rats, there was marginally more radioactivity and SAN Trimer detected in milk than in blood. The study authors concluded that SAN Trimer is rapidly absorbed, metabolized, and excreted in nonpregnant female rats, and not likely to accumulate in blood or tissues. They also concluded that fetal exposure to SAN Trimer can occur in utero through the ability of the compound to pass the placental barrier during gestation. It was determined that exposure can also occur via lactation.

Genotoxicity

MA Bioservices (1998a), (1998c), (1998b), (1998d)

Table 3 summarizes the studies examining genotoxicity (e.g., clastogenicity, mutagenicity) of SAN Trimer. One group of studies was conducted with SAN Trimer Batch 2, which was made by distillation of the product to 98% purity. In MA Bioservices (1998a) a bacterial reverse mutation assay was conducted in *Salmonella typhimurium* strains TA98, TA100, TA1535, TA1537, and TA102 with and without S9 extracts. A positive response was observed in strains TA98, TA100, and TA1537 in the absence of S9 activation. In a Chinese hamster ovary (CHO) cell in vitro mutation assay by MA Bioservices (1998b), SAN trimer did not induce mutations in the *hgprt* gene with and without S9 activation. MA Bioservices (1998c) also conducted an in vitro mammalian chromosome aberration assay in CHO cells that was concluded to be positive with a statistically significant increase in the percentage of cells with chromosomal aberrations with and without S9 activation after a 4-hour treatment with SAN Trimer, but not following 20 hours of treatment. In MA Bioservices (1998d), a mammalian bone marrow chromosome aberration and micronucleus test was conducted in S-D rats in vivo. There were no statistically significant increases in abnormal erythrocytes at 18 or 42 hours, and the study authors concluded both tests to be negative.

Hobbs et al. (2012), NTP (2012), Vacek et al. (2005)

The following tests used SAN Trimer Batch 3 (96% purity), which is prepared by extraction rather than distillation. It is postulated that distillation, which subjects the mixture to high temperatures, produces a different mixture of compounds than Batch 2. Hobbs et al. (2012) treated F344/N juvenile male and female rats (four to five rats per treatment group) with SAN Trimer (0, 37.5, 75, 150, or 300 mg/kg-day) via gavage in corn oil, once daily for 4 days. Using a comet assay, the study authors determined that SAN Trimer caused a statistically significant increase in DNA damage in total brain, cerebrum, cerebellum (not statistically significant in males), liver, and blood leukocytes (not statistically significant in females) in male and female rats as determined by quantitative staining. Hobbs et al. (2012) also reported statistically significant increased frequencies of micronucleated reticulocytes in both male (300 mg/kg-day) and female (≥150 mg/kg-day) rats (Hobbs et al., 2012). In a study conducted by SITEK Research Laboratories that was included in the NTP (2012) study, SAN Trimer was not mutagenic in *Salmonella typhimurium* strains TA98 or TA100 or in *Escherichia coli* strain WP2 *uvrA*/pKM101 in tests conducted with and without S9 activation. Vacek et al. (2005) compared the mutation frequencies of the hypoxanthine phosphoribosyl transferase (*HPRT*) gene in nonexposed, healthy children to children living in Dover Township, New Jersey whose siblings were included in a childhood cancer incidence study (NJ DHSS, 1997). The study authors observed no change in mutation frequency between the two groups (Vacek et al., 2005).

Table 3. Summary of Studies Evaluating Genotoxicity and Mutagenicity

Endpoint	Test System	Dose Concentration	Results[a]		Comments	References
			Without Activation	With Activation		
Genotoxicity studies in prokaryotic organisms						
Reverse mutation	*S. typhimurium* strains TA98, TA100, TA1535, TA1537, TA102	25–1,000 µg/plate without S9 activation; 25–5,000 µg/plate with S9 activation	+ (TA98,TA100, TA1537) - (TA1535, TA102)	-	SAN Trimer Batch 2	MA Bioservices (1998a)
Reverse mutation	*S. typhimurium* strains TA98, TA100	0−7,500 µg/plate	-	-	SAN Trimer Batch 3	NTP (2012)
Reverse mutation	*Escherichia coli* strain WP2 *uvrA*/pKM101	0−10,000 µg/plate	-	-	SAN Trimer Batch 3	NTP (2012)
Genotoxicity studies in mammals—in vitro						
Gene mutation	CHO cells/*hgprt*	50−400 µg/mL without S9 activation; 150−600 µg/mL with S9 activation	-	-	SAN Trimer Batch 2	MA Bioservices (1998b)
Chromosome aberration	CHO cells	50−450 µg/mL for 4 hours 25−400 µg/mL for 20 hours	+ -	+	SAN Trimer Batch 2 Marginally positive results obtained at 4 hours (approx. 1.8% above control at 440 µg/mL without S9)	MA Bioservices (1998c)
Genotoxicity studies in mammals—in vivo						
Chromosome aberration	S-D, rats, bone marrow (single dose gavage)	0, 125, 250, or 500 (males)and 0, 163, 325, or 650 (females) mg/kg	-	ND	SAN Trimer Batch 2	MA Bioservices (1998d)
Gene mutation	Human peripheral lymphocytes/*HPRT*	NDr	-	ND	SAN Trimer Batch 3	Vacek et al. (2005)

Table 3. Summary of Studies Evaluating Genotoxicity and Mutagenicity

Endpoint	Test System	Dose Concentration	Results[a]		Comments	References
			Without Activation	With Activation		
DNA damage	F344/N juvenile male and female rats (gavage, four to five rats per treatment group, 4 days)	0, 37.5, 75, 150, or 300 mg/kg-day	(+) Total brain (+) Cerebrum (+) Cerebellum (females) (+) Liver (at p <0.05) (+) Blood Leukocytes (males)	ND	SAN Trimer Batch 3	Hobbs et al. (2012); also reported in NTP (2012)
Micronucleus test	F344/N juvenile male and female rats (gavage, 4 days)	0, 37.5, 75, 150, or 300 mg/kg-day	(+) Reticulocytes (-) Erythrocytes	ND	SAN Trimer Batch 3 Lowest dose was not analyzed	Hobbs et al. (2012); also reported in NTP (2012)
Micronucleus test	S-D, rats, bone marrow (single dose gavage)	0, 125, 250, or 500 (males)and 0, 163, 325, or 650 (females) mg/kg	-	ND	SAN Trimer Batch 2	MA Bioservices (1998d)

[a]- = negative; NA = not applicable; ND = no data; NDr = not determined.

In summary, SAN Trimer was shown to be both positive and negative for mutagenicity in *S. typhimurium* strains and was not mutagenic in *E. coli* strains. SAN Trimer was not shown to induce *hgprt* mutations in CHO cells and *HPRT* mutations in human lymphocytes. In in vivo rat studies, SAN Trimer caused micronuclei formation in reticulocytes but not in bone marrow erythrocytes following gavage. SAN Trimer induced chromosomal aberrations in CHO cells in vitro but was negative in bone marrow erythrocytes from treated rats. SAN Trimer also caused DNA damage in brain, liver, and blood following gavage treatment in rats.

SYNTHESIS OF RESULTS FROM NONCANCER AND CANCER STUDIES

Four human studies on oral exposure to drinking water containing various chemicals, including SAN Trimer, are publicly available; one was a case-control study and the other three were ecological in nature because they examined town-level incidence and exposure data. The experimental database for animals is limited and includes a single acute study, two short-term-duration studies, a single subchronic-duration study, and a single chronic-duration study, all conducted in rats.

During the period from 1979−1995, the following childhood cancers were determined to be statistically significantly increased in children exposed to drinking water containing multiple chemicals, including SAN Trimer, in Ocean County, Dover Township, and the Toms River section of Dover Township, New Jersey: sympathetic nervous system, leukemia, brain/CNS (including other tumor types and astrocytomas), acute lymphocytic leukemia, and brain/CNS astrocytomas (NJ DHSS, 1997). In the case-control study on individuals exposed during the period between 1979 and 1996 in Dover Township and the Toms River section of Dover Township (NJ DHSS, 2003a), the study authors observed a statistically significant association for leukemia in females (0−19 years of age) and exposure to drinking water The results from the case-control study should be interpreted with caution because of the small number of study subjects. In addition, some of the other chemicals that were present in the drinking water, including trichloroethylene and tetrachloroethylene, are known to demonstrate carcinogenic potential. Following cleanup and removal of SAN Trimer along with the other chemicals from the drinking water, total childhood cancer and brain/CNS cancer incidence have returned to background levels, and fewer than expected cases of leukemia have been observed (see Figures 2 and 3 and Tables A-2 and A-3) (NJ DHSS 2008, 2003b). Due to the existence of other contaminants in drinking water from the well fields investigated, the conclusions that can be drawn from the epidemiologic studies are relatively limited.

In a 2-year cancer bioassay conducted by the NTP, the study authors reported increased incidences of testicular tumors, brain and spinal cord astrocytomas, and granular cell tumors (see Table 12) in male F344/N rats (F1) treated perinatally and postnatally with SAN Trimer for 2 years (NTP, 2012). Although the testicular tumors were statistically significantly increased, the high background incidence in the control group suggests that these tumors may not be related to SAN Trimer treatment. The incidences of brain and spinal cord astrocytomas and granular cell tumors in males were not statistically significantly increased. Noncancer nervous system effects in rats consisting of sciatic nerve degeneration (at ≥40 mg/kg-day in female rats) and spinal root degeneration (at 75 mg/kg-day in male rats) were reported in the 2-year NTP (2012) study. There was also increased incidence of cellularity in the brain cerebrum in male and female rats (statistically significant at 430 mg/kg-day in females) treated with SAN Trimer in the 2-week NTP study. These effects indicate the nervous system as a potential target organ for SAN Trimer toxicity.

There were also consistent liver effects in rats treated with SAN Trimer at various time points. At 2 weeks of treatment, SAN Trimer increased absolute (150 mg/kg-day) and relative liver weights (≥75 mg/kg-day) in males and females at ≥150 mg/kg-day as well as liver histopathological lesions at 300 mg/kg-day, including vacuolation of periacinar hepatocytes in males and periacinar hypertrophy in females (Huntingdon Life Sciences, 1999b). The NTP studies reported increased relative liver weight at ≥175 mg/kg-day in male and female rats at 2 weeks and at ≥40 mg/kg-day in male rats at 13 weeks (NTP, 2012). Following 2 years of treatment, the study authors reported the following histopathological noncancer changes in the liver: chronic active inflammation, eosinophilic foci, angiectasis (all at 75 mg/kg-day in males), and mixed cell foci at ≥20 mg/kg-day in males and at 85 mg/kg-day in females (NTP, 2012). These observed effects identify the liver as a potential target organ for SAN Trimer toxicity.

Finally, there were effects on reproductive parameters (e.g., testis weight, pup body weight) and other organs including the heart, bone marrow, and bladder, but these effects were not as consistently observed as the nervous system or hepatic effects. The potential points of departure (PODs) for subchronic- and chronic-duration effects are shown in Tables 6 and 9, respectively.

DERIVATION OF PROVISIONAL VALUES

Tables 4 and 5 present a summary of provisional noncancer reference values and cancer values, respectively.

Table 4. Summary of Provisional Noncancer Reference Values for SAN Trimer (Various CASRNs)

Toxicity Type	Species/Sex	Critical Effect	p-Reference Value	POD Method	POD_{HED}	UF_C	Principal Study
Subchronic p-RfD	F344/N Rat/M	Increased absolute liver weight	8×10^{-3} mg/kg-day	$BMDL_{10}$	2.4 mg/kg-day	300	NTP (2012)
Chronic p-RfD	F344/N Rat/Both	Increased incidences of chronic active inflammation in the liver (males) and sciatic nerve degeneration (females)	3×10^{-3} mg/kg-day	$BMDL_{10}$	0.77 mg/kg-day	300	NTP (2012)
Subchronic p-RfC	ND						
Chronic p-RfC	ND						

ND = not determined.

Table 5. Summary of Provisional Cancer Values for SAN Trimer (Various CASRNs)

Toxicity type	Species/Sex	Tumor Type	Cancer Value	Principal Study
p-OSF	ND			
p-IUR	ND			

ND = not determined.

DERIVATION OF PROVISIONAL ORAL REFERENCE DOSES

Derivation of Subchronic Provisional RfD (Subchronic p-RfD)

The subchronic-duration portion of the NTP (2012) study in F344/N rats is selected as the principal study for derivation of the subchronic p-RfD. In this study, groups of F1 rats (10/sex/dose) from SAN Trimer-treated dams were fed diets containing SAN Trimer for 13 weeks postweaning. This study is a peer-reviewed technical report and was conducted according to GLP standards. It is well conducted and meets the standard of study design and performance, with numbers of animals, examination of potential toxicity endpoints, and presentation of information (details are provided in the *Review of Potentially Relevant Data* section), and is the only available study that reports effects following subchronic-duration SAN Trimer treatment.

The study authors (NTP, 2012) reported statistically significant changes in body weight and organ weights that were amenable in some cases to modeling using the U.S. EPA's Benchmark Dose Software (BMDS version 2.2 as described in Appendix B) for consideration as potential PODs (see Table 6). Increased absolute organ weight changes in female F1 rats observed in the 13-week NTP (2012) study are not considered as potential critical effects because there were concomitant statistically significant decreases in body weight at all doses expect the lowest dose (10 mg/kg-day) that could have contributed to the increased absolute organ weights. For absolute organ weight changes in male F1 rats, data were modeled without the high dose because there was a statistically significant decrease in body weight at the highest dose (150 mg/kg-day).

The most sensitive effects following subchronic-duration treatment with SAN Trimer appear to be increased heart weight (absolute and relative) in male rats with a LOAEL of 10 mg/kg-day, and increased absolute liver weight in male rats with a $BMDL_{10}$ of 10 mg/kg-day (BMR of 10% relative risk). However, there is more support for increased absolute liver weight being selected as the critical effect based on the available data for SAN Trimer. When excluding the high dose as described above, the data for increased absolute liver weight in male rats display a clearer dose-response trend than the data for increased absolute and relative heart weights in male rats (see Table A-9). Increased heart and liver weights were both observed in multiple duration studies (NTP, 2012; Huntingdon Life Sciences, 1999b). However, increased incidences of pathological indices of heart toxicity were not observed in F1 rats that received dietary treatment of SAN Trimer for 2 weeks, 13 weeks, or 2 years (NTP, 2012). Heart lesions were also not observed in S-D rats that received gavage treatment of SAN Trimer for 2 weeks. The absence of pathological heart effects following SAN Trimer treatment suggests that increased absolute and relative heart weights may not be biologically significant; therefore, data for increased absolute and relative heart weights are not selected as a potential POD. The selection of increased absolute liver weight as the critical effect is supported by the observation that the liver appears to be a target organ of SAN Trimer toxicity. Liver effects (i.e., liver lesions and increased absolute and relative liver weights) were reported in both sexes of two strains of rats (F344/N and S-D) in short-term-duration studies (NTP, 2012; Huntingdon Life Sciences, 1999b), and F344/N rats in a chronic-duration study of dietary SAN Trimer treatment (NTP, 2012). **Therefore, the $BMDL_{10}$ of 10 mg/kg-day based on increased absolute liver weight in male F1 rats (NTP, 2012) is chosen as the POD to derive a subchronic p-RfD.**

Table 6. Candidate Subchronic PODs for SAN Trimer

Effect	Sex	NOAEL (mg/kg-day)	LOAEL (mg/kg-day)	BMD	BMDL	Comment
Decreased body weight in dams (F0) during gestation	Females	55	110[a]	Not Run	Not Run	No data variability available
Decreased body weight in F1 rats[d]	Males	80	150[a]	94[b] 141[c]	83[b] 127[c]	
Increased relative heart weight	Males	NDr	10	NF	NF	
Increased absolute heart weight	Males	NDr	10	NF	NF	
Increased relative right kidney weight[d]	Males	80	150[a]	55[b] 111[c]	44[b] 91[c]	
Increased relative liver weight[d]	Males	20	40[a]	12[b] 35[c]	8.3[b] 24[c]	
Increased absolute liver weight[d]	**Males**	**20**	**40[a]**	**13[b]** **25[c]**	**5[b]** **10[c]**	
Increased relative spleen weight	Males	20	40	46[b]	38[b]	
Increased absolute spleen weight	Males	40	80	82[b]	48[b]	
Increased relative right testis weight	Males	80	150	NF	NF	
Decreased body weight in F1 rats	Females	150	NDr	NF	NF	
Increased relative heart weight	Females	40	80	46[b]	15[b]	
Increased relative right kidney weight	Females	150	NDr	NF	NF	
Increased relative liver weight[d]	Females	80	150[a]	24[b] 91[c]	13[b] 56[c]	
Increased relative spleen weight	Females	40	80	NF	NF	

[a]Change was >10% compared to control values.
[b]BMR of 1 standard deviation relative risk.
[c]BMR of 10% relative risk.
[d]Based on language from U.S. EPA's *Benchmark Dose Technical Guidance Document* (U.S. EPA, 2012) that states, *"For consistency in reporting, the BMD corresponding to a one control SD shift in the control mean should always be presented along with the BMDs and BMDL for whatever BMR is being used for the POD,"* the BMD(L)s with BMRs of 1SD and 10% are both shown.

NDr = not determined; NF = no fit.

Dosimetric Adjustments:

Because the NTP (2012) study is a continuous feed study, dosimetric adjustments to convert to an adjusted daily dose were not needed.

In the U.S. EPA's *Recommended Use of Body Weight$^{3/4}$ as the Default Method in Derivation of the Oral Reference Dose* (U.S. EPA, 2011), the U.S. EPA endorses a hierarchy of approaches to derive human equivalent oral exposures from data from laboratory animal species, with the preferred approach being physiologically based pharmacokinetic modeling. Other approaches may include using some chemical-specific information without a complete physiologically based pharmacokinetic model. In lieu of chemical-specific models or data to inform the derivation of human equivalent oral exposures, the U.S. EPA endorses body-weight scaling to the 3/4 power (i.e., $BW^{3/4}$) as a standard method to extrapolate toxicologically equivalent doses of orally administered agents from all laboratory animals to humans for the purpose of deriving an RfD under certain exposure conditions. More specifically, the use of $BW^{3/4}$ scaling for deriving an RfD is recommended when the observed effects are associated with the parent compound or a stable metabolite, but not for portal-of-entry effects or developmental toxicity endpoints. A validated human physiologically based pharmacokinetic model for SAN Trimer is not available for use in extrapolating doses from animals to humans. The selected critical effect of increased absolute liver weight is associated with the parent compound or a stable metabolite. Furthermore, this liver effect is not a portal-of-entry or developmental toxicity effect. Therefore, scaling by $BW^{3/4}$ is relevant for deriving human equivalent doses (HEDs) for this effect.

Following U.S. EPA (2011) guidance, the POD for increased absolute liver weight in adult animals is converted to a HED through application of a dosimetric adjustment factor (DAF)[2] derived as follows:

$$DAF = (BW_a^{1/4} \div BW_h^{1/4})$$

where

DAF = dosimetric adjustment factor
BW_a = animal body weight
BW_h = human body weight

Using a BW_a of 0.25 kg for rats and a BW_h of 70 kg for humans (U.S. EPA, 1988), the resulting DAF is 0.24. Applying this DAF to the $BMDL_{10}$ of 10 mg/kg-day identified for the critical effect in rats yields a $BMDL_{10HED}$ as follows:

$$\begin{aligned} BMDL_{10HED} &= 10 \text{ mg/kg-day} \times DAF \\ &= 10 \text{ mg/kg-day} \times 0.24 \\ &= 2.4 \text{ mg/kg-day} \end{aligned}$$

The subchronic p-RfD for SAN Trimer is derived as follows:

$$\begin{aligned} \textbf{Subchronic p-RfD} &= BMDL_{10HED} \div UF_C \\ &= 2.4 \text{ mg/kg-day} \div 300 \\ &= \mathbf{8 \times 10^{-3} \text{ mg/kg-day}} \end{aligned}$$

[2]As described in detail in *Recommended Use of Body Weight$^{3/4}$ as the Default Method in Derivation of the Oral Reference Dose* U.S. EPA (2011), rate-related processes scale across species in a manner related to both the direct ($BW^{1/1}$) and allometric scaling ($BW^{3/4}$) aspects such that $BW^{3/4} \div BW^{1/1} = BW^{-1/4}$, converted to a $DAF = BW_a^{1/4} \div BW_h^{1/4}$.

The composite uncertainty (UF_C) for the subchronic p-RfD for SAN Trimer is 300, as summarized in Table 7. Table 8 provides the confidence descriptors for the subchronic p-RfD.

Table 7. UFs for Subchronic p-RfD of SAN Trimer

UF	Value	Justification
UF_A	3	A UF_A of 3 ($10^{0.5}$) has been applied to account for uncertainty in characterizing the toxicokinetic or toxicodynamic differences between rats and humans following oral SAN Trimer exposure. The toxicokinetic uncertainty has been accounted for by calculation of a human equivalent dose (HED) through application of a dosimetric adjustment factor (DAF) as outlined in the U.S. EPA's *Recommended Use of Body Weight$^{3/4}$ as the Default Method in Derivation of the Oral Reference Dose* (U.S. EPA, 2011).
UF_D	10	A UF_D of 10 has been applied for deficiencies in the available database because there are no acceptable two-generation reproductive or developmental toxicity studies for SAN Trimer via the oral route. The NTP (2012) study design did not satisfy the requirements for either of these study types.
UF_H	10	A UF_H of 10 has been applied for inter-individual variability to account for human-to-human variability in susceptibility in the absence of quantitative information to assess the toxicokinetics and toxicodynamics of SAN Trimer in humans.
UF_L	1	A UF_L of 1 has been applied for LOAEL-to-NOAEL extrapolation because the POD is a $BMDL_{10}$.
UF_S	1	A UF_S of 1 has been applied because a subchronic-duration study was selected as the principal study.
UF_C	300	Composite Uncertainty Factor = $UF_A \times UF_D \times UF_H \times UF_L \times UF_S$

Table 8. Confidence Descriptors for Subchronic p-RfD for SAN Trimer

Confidence Categories	Designation[a]	Discussion
Confidence in study	H	The study by the NTP (2012) is a well-conducted, peer-reviewed, GLP-compliant, and comprehensive study with a sufficient number of animals that examined a variety of endpoints.
Confidence in database	M	The database for SAN Trimer includes comprehensive acute, short-term-, subchronic-, and chronic-duration toxicity studies but is lacking acceptable two-generation reproductive and developmental toxicity studies.
Confidence in subchronic p-RfD[b]	M	The overall confidence in the subchronic p-RfD is medium.

[a]L = low, M = medium, H = high.
[b]The overall confidence cannot be greater than lowest entry in table.

Derivation of a Chronic Provisional RfD (Chronic p-RfD)

The chronic-duration portion of the NTP (2012) study in F344/N rats is selected as the principal study for derivation of the chronic p-RfD. In this study, groups of F1 rats (50/sex/dose) from SAN Trimer-treated dams were fed diets containing SAN Trimer for 2 years (104 weeks) postweaning. This study is a peer-reviewed technical report and was conducted according to GLP standards. It is well conducted and meets the standard of study design and

performance with numbers of animals, examination of potential toxicity endpoints, and presentation of information (details are provided in the *Review of Potentially Relevant Data* section), and is the only available study that reports effects following chronic-duration SAN Trimer treatment.

As discussed in the *Review of Potentially Relevant Data* section, pathological effects were reported in the following organs in F1 rats following chronic-duration SAN Trimer treatment (see Table A-12): liver (≥20 mg/kg-day), bone marrow (≥40 mg/kg-day), bladder (85 mg/kg-day), and the peripheral nervous system (≥40 mg/kg-day) (NTP, 2012). An attempt was made to model these data using the U.S. EPA's BMDS version 2.2 (as described Appendix B) for consideration as potential PODs (see Table 9). The most sensitive of these effects is increased incidence of chronic active inflammation in the liver of males with a $BMDL_{10}$ of 3.2 mg/kg-day (see Table 9). Although liver weight was not examined in the NTP (2012) 2-year study, support for chronic active inflammation in the liver as the critical effect for derivation of the chronic p-RfD is provided by the consistent observation of liver effects in male and female F344/N rats in the 2-week, 13-week, and 104-week studies performed by the NTP (NTP, 2012), as well as male and female S-D rats in the 2-week study by (NTP, 2012; Huntingdon Life Sciences, 1999b).

There were also effects in the nervous system (i.e., spinal root degeneration in males and sciatic nerve degeneration in females) that were statistically significantly increased in rats in the 2-year study NTP (2012). These effects were also modeled and the PODs are very similar to the $BMDL_{10}$ of 3.2 mg/kg-day for increased incidence of chronic active hepatic inflammation in male rats. As shown in Table 9, the $BMDL_{10}$ for spinal root degeneration in males is 4.9 mg/kg-day and the $BMDL_{10}$ for sciatic nerve degeneration in females is 3.5 mg/kg-day. Therefore, sciatic nerve degeneration in female rats and chronic active inflammation in the liver of male rats can be considered as co-critical effects due to the similarity in their BMDLs. In addition, the majority recommendation of the external peer reviewers was that these effects should be considered co-critical effects. **Thus, the increased incidences of chronic active inflammation in the liver of male rats and sciatic nerve degeneration in females are chosen as co-critical effects, and the $BMDL_{10}$ of 3.2 mg/kg-day is chosen as the POD.** The selection of the $BMDL_{10}$ of 3.2 mg/kg-day as the POD will protect against both liver and nervous system effects and also other chronic-duration effects that occurred in the bladder and bone marrow of rats (see Table 9).

Table 9. Candidate Chronic PODs: Effect Levels From Histopathological Endpoints in F344/N Rats Treated with SAN Trimer by Diet for 2 Years[a]

Lesion	NOAEL	LOAEL	BMD_{10}	$BMDL_{10}$
Peripheral Nervous System				
Sciatic nerve degeneration, females	**20**	**40**	**4.7**	**3.5**
Spinal root degeneration, males	40	75	67	4.9
Liver				
Chronic active inflammation, males	**40**	**75**	**8.0**	**3.2**
Eosinophilic foci, males	40	75	17	13
Mixed cell foci, females	40	85	41	23
Mixed cell foci, males	None	20	8.5[b]	3.7[b]
Angiectasis, males	40	75	NF	NF
Bone Marrow				
Bone marrow hyperplasia, females	20	40	13	3.6
Inflammation, granulomatous, females	20	40	40	34
Bone marrow hyperplasia, males	40	75	54	18
Bladder				
Hyperplasia of transitional epithelium, females	40	85	80	62

[a]NTP (2012).
[b]Data for incidence of mixed cell foci in male rats were modeled with BMDS and the mid- and high-dose groups were removed from the analysis because of a plateau effect observed when the data were modeled with all doses.

NF = no fit (none of the BMD models adequately fit the data).

Dosimetric Adjustments:

Because the NTP (2012) study is a continuous feed study, dosimetric adjustments to convert to an adjusted daily dose were not needed. As discussed in the subchronic p-RfD derivation section above, the U.S. EPA endorses body-weight scaling to the 3/4 power (i.e., $BW^{3/4}$) as a standard methodology to extrapolate toxicologically equivalent doses of orally administered agents from all laboratory animals to humans for the purpose of deriving an RfD under certain exposure conditions (U.S. EPA, 2011). The POD for chronic active inflammation in the liver of male rats is converted to a HED through application of a DAF derived as follows:

$$DAF = (BW_a^{1/4} \div BW_h^{1/4})$$

where

DAF = dosimetric adjustment factor
BW_a = animal body weight
BW_h = human body weight

Using a BW_a of 0.25 kg for rats and a BW_h of 70 kg for humans (U.S. EPA, 1988), the resulting DAF is 0.24. Applying this DAF to the $BMDL_{10}$ of 3.2 mg/kg-day identified for the co-critical effect of chronic active inflammation in the liver of male rats yields a $BMDL_{10HED}$ as follows:

$$\begin{aligned} BMDL_{10HED} &= 3.2 \text{ mg/kg-day} \times DAF \\ &= 3.2 \text{ mg/kg-day} \times 0.24 \\ &= 0.77 \text{ mg/kg-day} \end{aligned}$$

The chronic p-RfD for SAN Trimer is derived as follows:

$$\begin{aligned} \textbf{Chronic p-RfD} &= BMDL_{10HED} \div UF_c \\ &= 0.77 \text{ mg/kg-day} \div 300 \\ &= \mathbf{3 \times 10^{-3} \text{ mg/kg-day}} \end{aligned}$$

The UF_C for the chronic p-RfD for SAN Trimer is 300, as summarized in Table 10. Table 11 provides the confidence descriptors for the chronic p-RfD.

Table 10. UFs for Chronic p-RfD of SAN Trimer

UF	Value	Justification
UF_A	3	A UF_A of 3 ($10^{0.5}$) has been applied to account for uncertainty in characterizing the toxicokinetic or toxicodynamic differences between rats and humans following oral SAN Trimer exposure. The toxicokinetic uncertainty has been accounted for by calculation of a human equivalent dose (HED) through application of a dosimetric adjustment factor (DAF) as outlined in the U.S. EPA's *Recommended Use of Body Weight$^{3/4}$ as the Default Method in Derivation of the Oral Reference Dose* (U.S. EPA, 2011).
UF_D	10	A UF_D of 10 has been applied for deficiencies in the available database because there are no acceptable two-generation reproductive or developmental toxicity studies for SAN Trimer via the oral route. The NTP (2012) study design did not satisfy the requirements for either of these study types.
UF_H	10	A UF_H of 10 has been applied for inter-individual variability to account for human-to-human variability in susceptibility in the absence of quantitative information to assess the toxicokinetics and toxicodynamics of SAN Trimer in humans.
UF_L	1	A UF_L of 1 has been applied for LOAEL-to-NOAEL extrapolation because the POD is a $BMDL_{10}$.
UF_S	1	A UF_S of 1 has been applied because a chronic-duration study was selected as the principal study.
UF_C	300	Composite Uncertainty Factor = $UF_A \times UF_D \times UF_H \times UF_L \times UF_S$

Table 11. Confidence Descriptors for Chronic p-RfD for SAN Trimer

Confidence Categories	Designation[a]	Discussion
Confidence in study	H	The study by the NTP (2012) is a well-conducted, peer-reviewed, GLP-compliant, and comprehensive study with a sufficient number of animals that examined a variety of endpoints.
Confidence in database	M	The database for SAN Trimer includes comprehensive acute, short-term-, subchronic-, and chronic-duration toxicity studies but is lacking acceptable two-generation reproductive and developmental toxicity studies.
Confidence in chronic p-RfD[b]	M	The overall confidence in the chronic p-RfD is medium.

[a]L = low, M = medium, H = high.
[b]The overall confidence cannot be greater than lowest entry in table.

DERIVATION OF PROVISIONAL INHALATION REFERENCE CONCENTRATIONS

No information that could be used to derive subchronic or chronic p-RfCs for SAN Trimer was identified.

CANCER WEIGHT-OF-EVIDENCE (WOE) DESCRIPTOR

The *Guidelines for Carcinogen Risk Assessment* (Cancer Guidelines) (U.S. EPA, 2005) emphasize the importance of applying a weight-of-evidence (WOE) approach in reaching conclusions about the carcinogenic potential of chemicals in humans. Each cancer descriptor may be applicable to a variety of potential data sets and represent points along a continuum of evidence. The available carcinogenic evidence for SAN Trimer could be considered a borderline case between two potential descriptors— *suggestive evidence of carcinogenic potential* and *inadequate information to assess carcinogenic potential.* For example, oral treatment with SAN Trimer caused a non-statistically significant increase in the incidences of brain and spinal cord astrocytomas and granular cell tumors in male rats, which is consistent with one of the examples provided in the Cancer Guidelines (U.S. EPA, 2005) for the descriptor *suggestive evidence of carcinogenic potential*. The example states that supporting data for this descriptor may include "*a small, and possibly not statistically significant, increase in tumor incidence observed in a single animal or human study that does not reach the weight of evidence for the descriptor 'Likely to Be Carcinogenic to Humans.'*" However, because there was not a statistically significant increase in any relevant tumor type in rats treated with SAN Trimer for 2 years (NTP, 2012), some tumor types (e.g., pituitary gland adenomas, mammary gland tumors, etc.) were statistically significantly decreased in male and/or female rats, and the carcinogenic potential of SAN Trimer has only been evaluated in one animal species in one study (NTP, 2012), the descriptor indicating that there is "*inadequate information to assess the carcinogenic potential* is also relevant. Finally, whereas children exposed to drinking water containing SAN Trimer exhibited a statistically significant elevation in various tumor types (NJ DHSS, 1997), it is not possible to determine if SAN Trimer solely contributed to these cancer effects in children because other chemicals that have been shown to demonstrate carcinogenic potential (e.g., trichloroethylene, tetrachloroethylene) were also identified in the drinking water (Richardson et al., 1999).

The evidence from the only available carcinogenicity study in laboratory animals shows increased incidences of testicular tumors, brain and spinal cord astrocytomas and granular cell tumors (see Table 12) in male F344/N rats (F1) treated perinatally and postnatally with SAN Trimer for 2 years (NTP, 2012). There was a statistically significant increase in the incidence of total testicular tumors; however, the high background incidence in the control group (41/50) indicates that these tumors may not be treatment-related. This tumor has been identified as the most frequently observed spontaneous tumor in the male F344/N rat (Haseman et al., 1990; Takaki et al., 1989). Therefore, the incidence of total testicular tumors is not considered in the WOE for the carcinogenic potential of SAN Trimer. In addition, the incidences of brain and spinal cord astrocytomas and granular cell tumors in male rats were not statistically significantly increased at any dose level compared to concurrent controls. Furthermore, as pointed out by two of the five external peer reviewers of this PPRTV assessment, data presented by Sills et al. (1999) have shown brain astrocytoma rates in F334/N control rats fed the NIH-07 diet in several NTP studies to be up to 4% in males, whereas brain granular cell tumor rates have been up to 2% in males. In the NTP (2012) study, brain astrocytomas and granular cell tumors occurred in 2% of male rats at the mid- and high-dose levels (see Table 12). These data indicate that, although rare, the brain tumors observed in male rats in the NTP (2012) study are within the range of historical controls as reported by Sills et al. (1999). Based on these results from the NTP (2012) study, the NTP concluded that there was *no evidence of carcinogenic activity* for SAN Trimer.

In evaluating this borderline case, the U.S. EPA considered Section 2.5 of the Cancer Guidelines which states that the descriptor *suggestive evidence of carcinogenic potential* is appropriate when "*the weight of evidence is suggestive of carcinogenicity, a concern for potential carcinogenic effects is raised, but the data are not judged sufficient for a stronger conclusion*." The Cancer Guidelines further state that the descriptor *inadequate information to assess carcinogenic potential* is appropriate when "*available data are judged inadequate for applying one of the other descriptors*." Although either descriptor could be considered plausible, this PPRTV assessment attaches greater weight to the fact there is only one cancer study in one species (NTP, 2012) that reported mostly negative tumor findings and no statistically significant increase in any relevant tumor type in male and female rats. Although there may be a similar observation of the occurrence of brain/CNS astrocytomas between humans exposed to drinking water containing SAN Trimer among many other chemicals (NJ DHSS, 1997) and male rats following oral exposure to SAN Trimer (NTP, 2012), the incidence of brain and spinal cord astrocytomas was not statistically significantly increased in male rats compared to concurrent controls, and the observed incidences of these brain tumors are within the historical range of data presented in a previous publication (Sills et al., 1999). Thus under the U.S. EPA's Cancer Guidelines (U.S. EPA, 2005) and in concurrence with the majority recommendation of the external peer reviewers for this PPRTV assessment, there is *inadequate information to assess carcinogenic potential* for SAN Trimer.

Table 12. Incidences of Selected Cancer Endpoints in Male F344/N Rats Administered SAN Trimer by Diet for 2 Years[a]

Exposure Group (Human Equivalency Dose, mg/kg-day)[b]				
Parameter	**0 (0)**	**20 (5.6)**	**40 (11)**	**75 (20)**
Sample size	**50**	**50**	**50**	**50**
Brain				
Astrocytomas[c]	0	0	1 (2)	1 (2)
Granular cell tumors[c]	0	0	1 (2)	1 (2)
Spinal Cord				
Astrocytomas[c]	0	0	0	1 (2)
Granular cell tumors[c]	0	1	0	0
Testes				
Total, interstitial cell, adenoma (bilateral + unilateral, combined)[c,e]	41 (82)	49 (98)[d]	44 (88)	49 (98)[d]

[a]NTP (2012).
[b]Doses were converted from adjusted daily doses to human equivalency doses using the following formula: $Dose_{HED} = Dose_{ADJ} \times (Body\ Weight\ Animal \div Body\ Weight\ Human)^{(0.25)}$.
[c]Incidence, (corresponding percentage).
[d]Significantly different from control ($p \leq 0.05$) using the Poly-3 test, as reported by the study authors.
[e]Statistically significant trend using Poly-3 test for dose-response relationship in male rats, as reported by the study authors.

DERIVATION OF PROVISIONAL CANCER POTENCY VALUES

Derivation of Provisional Oral Slope Factor (p-OSF)

Because there are inadequate data to determine human carcinogenic potential, the derivation of an oral slope factor is precluded. The decision to not develop a quantitative estimate of cancer risk from oral exposure to SAN Trimer was supported by the majority of the external reviewers.

Derivation of Provisional Inhalation Unit Risk (p-IUR)

No human or animal studies examining the carcinogenicity of SAN Trimer following inhalation exposure have been identified. Therefore, derivation of a p-IUR is precluded.

APPENDIX A. FIGURES AND DATA TABLES

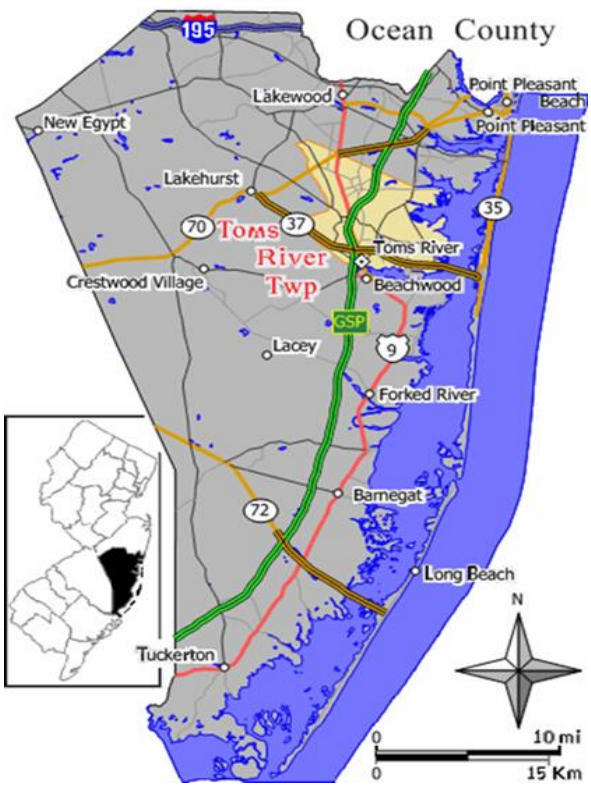

Figure A-1. Map of Ocean County, Dover Township (currently Toms River Township) and the Toms River section of Dover Township (currently the sub-Township area) New Jersey

Table A-1. Summary of Statistically Significantly Elevated SIRs for Ocean County[a,b]							
1979−1995							
Cancer Type	**Age Group**	**Sex**	**Race**	**Number Observed**	**Number Expected**	**SIR**	**95% CI Lower-Upper**
Brain/CNS astrocytoma	0−19	Both	All	35	23.9	1.46*	1.02−2.03
PNS neuroblastoma	0−19	Both	All	27	15.9	1.70*	1.12−2.47
PNS neuroblastoma	0−4	Both	All	24	13.3	1.81*	1.16−2.69
PNS neuroblastoma	0−19	Male	All	20	9.1	2.20*	1.34−3.40
PNS neuroblastoma	0−4	Male	All	17	7.7	2.21*	1.29−3.54

[a]NJ DHSS (1997).
[b]Incidence data for Ocean County were not reported in the later studies (NJ DHSS, 2008, 2003b).

*Statistically elevated, $p < 0.05$.

CNS = Central Nervous System; PNS = Peripheral Nervous System; SIR = Standardized Incidence Ratio.

Table A-2. Summary of Statistically Significantly Elevated SIRs for Dover/Toms River Township

Cancer Type	Age Group	Sex	Race	Number Observed	Number Expected	SIR	95% CI Lower-Upper
1979−1995							
All types[a]	0−19	Both	All	90	67.0	1.34* (1.3*)	1.08−1.65 (1.04−1.6)
All types[a]	0−19	Female	All	47	33.3	1.41* (1.4*)	1.04−1.88 (1.01−1.85)
All types[a]	0−4	Female	All	16	8.4	1.90* (1.8)	1.09−3.09 (0.99−3.04)
Acute lymphocytic leukemia[a]	0−19	Female	All	12	4.6	2.59* (2.6*)	1.34−4.53 (1.34−4.53)
Acute lymphocytic leukemia[a]	0−4	Female	All	7	2.2	3.27* (3.3*)	1.31−6.73 (1.30−6.70)
Leukemia	0−19	Female	All	13	6.5	1.99*	1.06−3.40
1996−2000							
All types	0−19	Both	All	25	20.6	1.2	0.78−1.79
All types	0−19	Female	All	13	9.4	1.4	0.74−2.37
All types	0−4	Female	All	3	2.2	1.4	0.28
Acute lymphocytic leukemia	0−19	Female	All	3	1.5	2.0	0.40−5.83
Acute lymphocytic leukemia	0−4	Female	All	2	0.7	2.9	0.32−10.4
Leukemia	0−19	Female	All	4	2.0	2.0	0.53−5.06
2001−2005							
All types	0−19	Both	All	26	22.8	1.1	0.75−1.7
All types	0−19	Female	All	14	10.4	1.3	0.73−2.3
All types	0−4	Female	All	2	2.6	0.8	0.09−2.8
Acute lymphocytic leukemia	0−19	Female	All	1	1.6	NC	NC
Acute lymphocytic leukemia	0−4	Female	All	0	0.6	NC	NDr
Leukemia	0−19	Female	All	1	1.6	NC	NC

[a]Numbers in parentheses are updated from NJ DHSS (2003b).

*Statistically elevated, $p < 0.05$

NC= Not calculated by the study authors; NDr = Not determined.

Table A-3. Summary of Statistically Significantly Elevated SIRs for Toms River Section/Sub-Township

Cancer Type	Age Group	Sex	Race	Number Observed	Number Expected	SIR	95% CI Lower-Upper
1979−1995							
All types	0−19	Both	All	24 (24)	14.1 (14.4)	1.70* (1.7*)	1.09−2.53 (1.07−2.49)
All types	0−4	Both	All	13 (12)	3.5 (3.4)	3.73* (3.6*)	1.99−6.38 (1.84−6.22)
All types	0−4	Female	All	10 (10)	1.6 (1.5)	6.17* (6.5*)	2.95−11.34 (3.13−12.0)
Brain/CNS	0−4	Both	All	4 (4)	0.6 (0.6)	7.04* (7.0*)	1.89−18.03 (1.87−17.8)
Brain/CNS	0−4	Female	All	3 (3)	0.3 (0.3)	11.60* (11.3*)	2.33−33.88 (2.27−33.0)
Brain/CNS astrocytoma	0−4	Both	All	2	0.2 (0.2)	9.47* (8.9*)	1.06−34.19 (1.00−32.1)
Acute lymphocytic leukemia	0−4	Female	All	4	0.4 (0.4)	9.68* (9.4*)	2.60−24.78 (2.52−24.0)
Leukemia	0−4	Female	All	4	0.5	7.84*	2.11−20.06
1996−2000							
All types	0−19	Both	All	5	4.2	1.2	0.39−2.81
All types	0−4	Both	All	0	1.1	NC	NDr
All types	0−4	Female	All	0	0.4	NC	NDr
Brain/CNS	0−4	Both	All	0	0.2	NC	NDr
Brain/CNS	0−4	Female	All	0	0.1	NC	NDr
Brain/CNS astrocytoma	0−4	Both	All	0	0.1	NDr	NDr
Acute lymphocytic leukemia	0−4	Female	All	0	0.1	NDr	NDr
Leukemia	0−4	Female	All	0	0.2	NDr	NDr
2001−2005							
All types	0−19	Both	All	5	4.6	1.1	0.35−2.5
All types	0−4	Both	All	2	1.3	1.6	0.18−5.8
All types	0−4	Female	All	1	0.5	NC	NC
Brain/CNS	0−4	Both	All	1	0.2	NC	NC
Brain/CNS	0−4	Female	All	0	0.1	NC	NDr
Brain/CNS astrocytoma	0−4	Both	All	0	0.1	NC	NDr

Table A-3. Summary of Statistically Significantly Elevated SIRs for Toms River Section/Sub-Township

Cancer Type	Age Group	Sex	Race	Number Observed	Number Expected	SIR	95% CI Lower-Upper
2001−2005							
Acute lymphocytic leukemia	0−4	Female	All	0	0.1	NC	NDr
Leukemia	0−4	Female	All	0	0.2	NC	NDr

[a]Numbers in parentheses are updated from NJ DHSS (2003b).

*Statistically elevated, $p < 0.05$.

CNS = Central Nervous System; NC= Not calculated by study authors; NDr= Not determined; SIR = Standardized Incidence Ratio.

Table A-4. Mean Body Weights in Dams from 2-Week Toxicity Study of SAN Trimer[a]

Endpoint	Exposure Concentration in ppm (mg/kg-day)					
	0	**250 (18)**	**500 (35)**	**1,000 (67)**	**2,000 (130)**	**4,000 (197)**
Gestation						
No. of animals[b]	8	8	8	7	8	7
GD 1 body weight (g) (%)[d]	163	169 (↑4)	161 (↓1)	169 (↑4)	166 (↑2)	170 (↑4)
GD 7 body weight (g) (%)[d]	180	185 (↑3)	176 (↓2)	187 (↑4)	181 (↑1)	187 (↑4)
GD 14 body weight (g) (%)[d]	203	207 (↑2)	195 (↓4)	207 (↑2)	199 (↓2)	181** (↓11)
GD 18 body weight (g) (%)[d]	223	231 (↑4)	217 (↓3)	233 (↑4)	226 (↑1)	204* (↓8)
Endpoint	**Exposure Concentration in ppm (mg/kg-day)**					
	0	**250 (40)**	**500 (85)**	**1,000 (166)**	**2,000 (325)**	**4,000 (634)**
Postnatal						
No. of animals[b]	7	7[c]	7	7	8	6
PND 1 body weight (g) (%)[d]	199	206 (↑4)	193 (↓3)	202 (↑2)	195 (↓2)	177** (↓11)
PND 7 body weight (g) (%)[d]	210	221 (↑5)	206 (↓2)	219 (↑4)	209 (0)	184** (↓12)
PND 14 body weight (g) (%)[d]	234	239 (↑2)	225 (↓4)	236 (↑1)	223 (↓5)	177** (↓24)
PND 20 body weight (g) (%)[d]	235	237 (↑1)	227 (↓3)	241 (↑3)	222 (↓6)	174** (↓26)

[a]NTP (2012).
[b]No. of animals weighed on each day.
[c]Eight dams were weighed on PND 1.
[d]Mean (% change from control).

*Dunnett's test $p \leq 0.05$.
**Dunnett's test $p \leq 0.01$.

GD = Gestation Day; PND = Postnatal Day.

Table A-5. Mean Body Weight in F1 Pups from 2-Week Toxicity Study of SAN Trimer[a]

Endpoint	Exposure Concentration in ppm (mg/kg-day)					
	0	250 (40)	500 (85)	1,000 (166)	2,000 (325)	4,000 (634)
Male						
No. of animals[b]	39	30	33	31	38	27
PND 1 body weight (g) (%)	5.8	5.9 (↑2)	6.0 (↑3)	5.8 (0)	5.8 (0)	5.3** (↓9)
No. of animals[c]	10	10	10	10	10	10
PND 4 body weight (g) (%)[d]	8.8	9.0 (↑2)	8.6 (↓2)	8.4 (↓5)	8.8 (0)	7.5** (↓15)
PND 7 body weight (g) (%)[d]	13.5	13.6 (↑1)	13.0 (↓4)	13.0 (↓4)	12.9 (↓4)	10.4** (↓23)
PND 14 body weight (g) (%)[d]	25.7	26.2 (↑2)	25.1 (↓2)	24.9 (↓3)	24.6 (↓4)	16.8** (↓35)
PND 20 body weight (g) (%)[d]	35.0	35.8 (↑2)	34.7 (↓1)	34.5 (↓1)	31.6** (↓10)	19.8** (↓43)
Female						
No. of animals[b]	23	34	32	40	49	31
PND 1 body weight (g) (%)[d]	5.4	5.6 (↑4)	5.4 (0)	5.5 (↑2)	5.3 (↓2)	5.0** (↓7)
No. of animals[c]	10	10	10	10	10	10
PND 4 body weight (g) (%)[d]	8.2	8.5 (↑4)	8.2 (0)	8.0 (↓2)	8.2 (0)	7.3* (↓11)
PND 7 body weight (g) (%)[d]	12.7	12.9 (↑2)	12.7 (0)	12.4 (↓2)	12.4 (↓2)	9.9** (↓22)
PND 14 body weight (g) (%)[d]	24.7	24.7 (0)	25.0 (↑1)	24.2 (↓2)	23.7 (↓4)	16.1** (↓35)
PND 20 body weight (g) (%)[d]	33.6	33.9 (↑1)	33.6 (0)	32.7 (↓3)	30.3** (↓10)	18.8** (↓44)

[a]NTP (2012).
[b]No. of animals weighed on PND 1.
[c]No. of animals weighed on PND 4, 7, 14, and 20.
[d]Mean (% change from control).

*Dunnett's test $p \leq 0.05$.
**Dunnett's test $p \leq 0.01$.

PND = Postnatal Day.

Table A-6. Absolute and Relative Organ Weights in Male and Female F1 Rats from 2-Week Toxicity Study of SAN Trimer[a]

Endpoint	Exposure Concentration in ppm					
	0	**250**	**500**	**1,000**	**2,000**	**4,000**
Males						
mg/kg-d	0	50	90	175	270	410
Number of animals	10	10	10	10	10	9
Necropsy body weight (g) (%)[b]	118 ± 6	114 ± 4 (↓3)	109 ± 4 (↓8)	106* ± 2 (↓10)	74** ± 3 (↓37)	34** ± 2 (↓71)
Absolute Heart (g) (%)[b]	0.52 ± 0.02	0.51 ± 0.02 (↓2)	0.48 ± 0.02 (↓8)	0.47* ± 0.01 (↓10)	0.34** ± 0.01 (↓35)	0.34** ± 0.02 (↓35)
Relative Heart Ratio (%)[b]	4.395 ± 0.062	4.478 ± 0.061 (↑2)	4.392 ± 0.083 (0)	4.424 ± 0.060 (↑1)	4.564 ± 0.069 (↑4)	10.297** ± 1.002 (↑134)
Absolute right kidney (g) (%)[b]	0.60 ± 0.03	0.58 ± 0.02 (↓3)	0.56 ± 0.02 (↓7)	0.55* ± 0.01 (↓8)	0.39** ± 0.01 (↓35)	0.25** ± 0.01 (↓58)
Relative right kidney ratio (%)[b]	5.098 ± 0.082	5.063 ± 0.067 (↓1)	5.134 ± 0.073 (↑1)	5.158 ± 0.112 (↑1)	5.346 ± 0.092 (↑5)	7.380** ± 0.233 (↑45)
Absolute liver (g) (%)[b]	5.91 ± 0.34	6.21 ± 0.35 (↑5)	5.69 ± 0.27 (↓4)	6.02 ± 0.13 (↑2)	3.89** ± 0.13 (↓34)	2.34** ± 0.10 (↓60)
Relative liver ratio (%)[b]	49.876 ± 0.756	54.119* ± 1.370 (↑9)	52.179* ± 0.592 (↑5)	56.806** ± 1.009 (↑14)	52.935** ± 0.825 (↑6)	68.468** ± 1.307 (↑37)
Absolute lung (g) (%)[b]	0.92 ± 0.03	0.88 ± 0.04 (↓4)	0.85 ± 0.03 (↓8)	0.86 ± 0.03 (↓7)	0.60** ± 0.02 (↓35)	0.45** ± 0.05 (↓51)
Relative lung ratio (%)[b]	7.896 ± 0.289	7.762 ± 0.250 (↓2)	7.817 ± 0.259 (↓1)	8.064 ± 0.245 (↑2)	8.208 ± 0.214 (↑4)	12.827** ± 0.892 (↑162)
Absolute spleen (g) (%)[b]	0.385 ± 0.013	0.364 ± 0.015 (↓5)	0.348 ± 0.014 (↓10)	0.346 ± 0.006 (↓10)	0.334 ± 0.033 (↓13)	0.255** ± 0.008 (↓34)
Relative spleen ratio (%)[b]	3.281 ± 0.069	3.186 ± 0.065 (↓3)	3.211 ± 0.070 (↓2)	3.269 ± 0.067 (0)	4.461** ± 0.327 (↑36)	7.587** ± 0.532 (↑231)
Absolute right testis (g) (%)[b]	0.561 ± 0.037	0.535 ± 0.043 (↓5)	0.451* ± 0.037 (↓20)	0.440** ± 0.019 (↓22)	0.237** ± 0.017 (↓58)	0.069** ± 0.003 (↓88)
Relative right testis ratio (%)[b]	4.724 ± 0.159	4.644 ± 0.260 (↓2)	4.121* ± 0.234 (↓13)	4.145* ± 0.150 (↓12)	3.207** ± 0.175 (↓32)	2.037** ± 0.067 (↓57)
Absolute thymus (g)	0.378 ± 0.018	0.348 ± 0.019 (↓8)	0.306** ± 0.011 (↓19)	0.298** ± 0.009 (↓21)	0.174** ± 0.007 (↓54)	0.045** ± 0.004 (↓88)

Table A-6. Absolute and Relative Organ Weights in Male and Female F1 Rats from 2-Week Toxicity Study of SAN Trimer[a]

Endpoint	Exposure Concentration in ppm					
	0	**250**	**500**	**1,000**	**2,000**	**4,000**
Relative thymus ratio (%)[b]	3.212 ± 0.085	3.035 ± 0.064 (↓6)	2.832** ± 0.092 (↓12)	2.810** ± 0.063 (↓13)	2.371** ± 0.060 (↓26)	1.304** ± 0.086 (↓59)
Females						
mg/kg-d	0	45	90	185	295	430
Number of animals	10	10	10	10	10	10
Necropsy body weight (g) (%)[b]	106 ± 5	103 ± 3 (↓3)	101 ± 3 (↓5)	102 ± 2 (↓4)	72** ± 3 (↓32)	32** ± 1 (↓70)
Absolute heart (g) (%)[b]	0.46 ± 0.02	0.47 ± 0.01 (↑2)	0.45 ± 0.01 (↓2)	0.46 ± 0.01 (0)	0.32** ± 0.01 (↓30)	0.28** ± 0.01 (↓39)
Relative heart ratio (%)[b]	4.333 ± 0.106	4.609 ± 0.103 (↑6)	4.465 ± 0.093 (↑3)	4.521 ± 0.057 (↑4)	4.493 ± 0.052 (↑4)	8.822** ± 0.601 (↑91)
Absolute right kidney (g) (%)[b]	0.55 ± 0.03	0.54 ± 0.02 (↓2)	0.52 ± 0.02 (↓5)	0.55 ± 0.01 (0)	0.38** ± 0.02 (↓31)	0.23** ± 0.01 (↓58)
Relative right kidney Ratio (%)[b]	5.171 ± 0.086	5.232 ± 0.060 (↑1)	5.207 ± 0.082 (↑1)	5.449 ± 0.073 (↑5)	5.267 ± 0.052 (↑2)	7.200** ± 0.166 (↑39)
Absolute liver (g) (%)[b]	5.12 ± 0.28	5.38 ± 0.20 (↑5)	5.13 ± 0.14 (0)	5.84 ± 0.14 (↑14)	3.83** ± 0.20 (↓25)	2.07** ± 0.10 (↓60)
Relative liver ratio (%)[b]	48.351 ± 0.891	52.056* ± 0.832 (↑8)	50.916* ± 0.478 (↑5)	57.438** ± 0.602 (↑19)	52.956** ± 1.362 (↑10)	64.269** ± 1.124 (↑33)
Absolute lung (g) (%)[b]	0.79 ± 0.04	0.90 ± 0.04 (↑14)	0.83 ± 0.04 (↑5)	0.87 ± 0.04 (↑10)	0.57** ± 0.02 (↓28)	0.38** ± 0.01 (↓52)
Relative lung ratio (%)[b]	7.534 ± 0.279	8.801 ± 0.418 (↑17)	8.208 ± 0.285 (↑9)	8.509 ± 0.336 (↑13)	7.971 ± 0.303 (↑6)	11.886** ± 0.581 (↑58)
Absolute spleen (g) (%)[b]	0.334 ± 0.016	0.334 ± 0.013 (0)	0.317 ± 0.006 (↓5)	0.338 ± 0.006 (↑1)	0.300 ± 0.028 (↓10)	0.223** ± 0.007 (↓33)
Relative spleen ratio (%)[b]	3.170 ± 0.073	3.233 ± 0.081 (↑2)	3.154 ± 0.064 (↓1)	3.331 ± 0.031 (↑5)	4.122 ± 0.253** (↑30)	7.072 ± 0.386** (↑123)
Absolute thymus (g) (%)[b]	0.360 ± 0.015	0.336 ± 0.010 (↓7)	0.332 ± 0.012 (↓8)	0.330 ± 0.013 (↓8)	0.188** ± 0.011 (↓48)	0.043** ± 0.005 (↓88)
Relative thymus ratio (%)[b]	3.438 ± 0.117	3.270 ± 0.081 (↓5)	3.300 ± 0.093 (↓4)	3.236 ± 0.085 (↓6)	2.627** ± 0.150 (↓24)	1.310** ± 0.100 (↓62)

Table A-6. Absolute and Relative Organ Weights in Male and Female F1 Rats from 2-Week Toxicity Study of SAN Trimer[a]

Endpoint	Exposure Concentration in ppm					
	0	**250**	**500**	**1,000**	**2,000**	**4,000**
Absolute uterus (g) (%)[b]	0.165 ± 0.020	0.202 ± 0.029 (↑22)	0.176 ± 0.017 (↑7)	0.161 ± 0.022 (↓2)	0.052** ± 0.005 (↓68)	0.023** ± 0.002 (↓86)
Relative uterus ratio (%)[b]	1.573 ± 0.201	1.932 ± 0.255 (↑23)	1.746 ± 0.162 (↑11)	1.600 ± 0.226 (↑2)	0.722** ± 0.072 (↓54)	0.729** ± 0.066 (↓54)

[a]NTP (2012).
[b]Mean ± standard error (% change from control).

*Williams' test $p \leq 0.05$.
**Williams' test $p \leq 0.01$.

Table A-7. Mean Body Weights in Dams from 13-Week Toxicity Study of SAN Trimer[a]

Endpoint	Dose Group ppm (mg/kg-day)					
	0	**100 (6.9)**	**200 (14)**	**400 (28)**	**800 (55)**	**1,600 (110)**
Gestation						
No. of animals[b]	9	9	8[c]	8	8	8
GD 1 body weight (g) (%)[d]	187	178 (↓5)	179 (↓4)	180 (↓4)	181 (↓3)	183 (↓2)
GD 7 body weight (g) (%)[d]	201	192 (↓4)	194 (↓3)	191 (↓5)	193 (↓4)	195 (↓3)
GD 14 body weight (g) (%)[d]	224	212 (↓5)	216 (↓4)	209* (↓7)	207* (↓8)	205* (↓8)
GD 20 body weight (g) (%)[d]	268	254 (↓5)	259 (↓3)	245 (↓9)	243 (↓9)	230* (↓14)
Endpoint	**Dose Group ppm (mg/kg-day)**					
	0	**100 (17)**	**200 (33)**	**400 (66)**	**800 (132)**	**1,600 (264)**
Postnatal						
No. of animals[b]	8	8	7	6	6	4[e]
PND 1 body weight (g) (%)[d]	217	209 (↓4)	211 (↓3)	209 (↓4)	210 (↓3)	209 (↓4)
PND 7 body weight (g) (%)[d]	225	219 (↓3)	224 (0)	221 (↓2)	218 (↓3)	223 (↓1)
PND 14 body weight (g) (%)[d]	252	243 (↓4)	247 (↓2)	240 (↓5)	243 (↓4)	243 (↓4)
PND 20 body weight (g) (%)[d]	244	241 (↓1)	246 (↑1)	239 (↓2)	245 (0)	240 (↓2)

[a]NTP (2012).
[b]No. of animals weighed on each day.
[c]Nine dams were weighed on PND 20.
[d]Mean (% change from control).
[e]Three dams were weighted on PND 20.

*Dunnett's test $p \leq 0.05$.

GD = Gestation Day; PND = Postnatal Day.

Table A-8. Mean Body Weights of F1 Pups from 13-Week Toxicity Study of SAN Trimer[a]

Endpoint (*n* = 10)	Dose Group ppm (mg/kg-day)					
	0	**100 (17)**	**200 (33)**	**400 (66)**	**800 (132)**	**1,600 (264)**
Males						
PND 4 BW (g) (%)[b]	8.4	8.8 (↑5)	8.6 (↑2)	8.8 (↑5)	8.9 (↑6)	8.5 (↑1)
PND 7 BW (g) (%)[b]	12.8	12.7 (↓1)	13.2 (↑3)	13.3 (↑4)	12.9 (↑1)	12.7 (↓1)
PND 14 BW (g) (%)[b]	21.9	23.2 (1↑6)	23.1 (↑5)	23.0 (↑5)	22.5 (↑3)	22.5 (↑3)
PND 20 BW (g) (%)[b]	28.7	31.3 (↑9)	30.7 (↑7)	31.1 (↑8)	30.0 (↑5)	29.9 (↑4)
Females						
PND 4 BW (g) (%)[b]	8.2	8.5 (↑4)	8.0 (↓2)	8.3 (↑1)	8.8 (↑7)	8.3 (↑1)
PND 7 BW (g) (%)[b]	12.4	12.4 (0)	12.1 (↓2)	12.6 (↑2)	12.8 (↑3)	12.0 (↓3)
PND 14 BW (g) (%)[b]	21.3	22.7 (↑7)	21.7 (↑2)	21.9 (↑3)	22.1 (↑4)	21.4 (0)
PND 20 BW (g) (%)[b]	27.9	30.9* (↑11)	28.9 (↑4)	29.7 (↑6)	29.4 (↑5)	28.8 (↑3)

[a]NTP (2012).
[b]Mean (% change from control).

BW = Body Weight; PND = Postnatal Day.

Table A-9. Absolute and Relative Organ Weights in Male and Female F1 Pups in the 13-Week Toxicity Study of SAN Trimer[a]

Endpoint	Dose Group ppm (mg/kg-day)					
	0	**100 (10)**	**200 (20)**	**400 (40)**	**800 (80)**	**1,600 (150)**
Males						
Number of animals	10	10	10	10	10	10
Necropsy body weight (g) (%)[b]	338 ± 7	344 ± 3 (↑2)	342 ± 4 (↑1)	337 ± 4 (0)	335 ± 6 (↓1)	302** ± 5 (↓11)
Absolute brain (g) (%)[b]	1.951 ± 0.017	1.964 ± 0.017 (↑1)	1.990 ± 0.020 (↑2)	1.968 ± 0.015 (↑1)	1.988 ± 0.019 (↑2)	1.987 ± 0.010 (↑2)
Relative brain ratio (%)[b]	5.785 ± 0.097	5.715 ± 0.083 (↓1)	5.823 ± 0.071 (↑1)	5.846 ± 0.037 (↑1)	5.947 ± 0.107 (↑3)	6.588** ± 0.098 (↑14)
Absolute heart (g) (%)[b]	0.91 ± 0.02	1.00** ± 0.01 (↑10)	0.97 ± 0.02 (↑7)	1.01** ± 0.02 (↑11)	0.98* ± 0.02 (↑8)	0.92 ± 0.01 (↑1)
Relative heart ratio (%)[b]	2.701 ± 0.036	2.894** ± 0.044 (↑7)	2.830** ± 0.042 (↑5)	3.000** ± 0.054 (↑11)	2.935** ± 0.038 (↑9)	3.049** ± 0.034 (↑13)
Absolute R. kidney (g) (%)[b]	1.00 ± 0.02	1.00 ± 0.02 (0)	1.03 ± 0.02 (↑3)	1.04 ± 0.02 (↑4)	1.04 ± 0.02 (↑4)	1.01 ± 0.03 (↑1)
Relative R. kidney ratio (%)[b]	2.956 ± 0.041	2.897 ± 0.058 (↓2)	3.004 ± 0.033 (↑2)	3.088 ± 0.040 (↑4)	3.099* ± 0.052 (↑5)	3.348** ± 0.043 (↑13)
Absolute liver (g) (%)[b]	11.25 ± 0.21	11.96* ± 0.17 (↑6)	12.31** ± 0.21 (↑9)	12.53** ± 0.25 (↑11)	13.13** ± 0.34 (↑17)	12.68** ± 0.24 (↑13)
Relative liver ratio (%)[b]	33.282 ± 0.367	34.754* ± 0.327 (↑4)	35.998** ± 0.446 (↑8)	37.195** ± 0.633 (↑12)	39.154** ± 0.535 (↑18)	41.972** ± 0.395 (↑26)
Absolute lung (g) (%)[b]	1.99 ± 0.07	1.92 ± 0.07 (↓4)	1.92 ± 0.05 (↓4)	2.02 ± 0.09 (↑2)	2.11 ± 0.09 (↑6)	1.74 ± 0.05 (↓13)
Relative lung ratio (%)[b]	5.879 ± 0.173	5.597 ± 0.215 (↓5)	5.631 ± 0.175 (↓4)	6.006 ± 0.256 (↑2)	6.281 ± 0.213 (↑7)	5.722 ± 0.141 (↓3)
Absolute spleen (g) (%)[b]	0.733 ± 0.014	0.780 ± 016 (↑6)	0.764 ± 0.023 (↑4)	0.786 ± 0.015 (↑7)	0.799* ± 0.017 (↑9)	0.799* ± 0.013 (↑9)
Relative spleen ratio (%)[b]	2.172 ± 0.037	2.267 ± 0.042 (↑4)	2.232 ± 0.053 (↑3)	2.333** ± 0.041 (↑7)	2.383** ± 0.024 (↑10)	2.646** ± 0.042 (↑22)
Absolute right testis (g) (%)[b]	1.460 ± 0.019	1.455 ± 0.017 (0)	1.448 ± 0.030 (↓1)	1.463 ± 0.014 (0)	1.457 ± 0.026 (0)	1.381* ± 0.014 (↓5)
Relative right testis ratio (%)[b]	4.326 ± 0.064	4.232 ± 0.059 (↓2)	4.240 ± 0.106 (↓2)	4.350 ± 0.073 (↑1)	4.350 ± 0.030 (↑1)	4.575* ± 0.048 (↑6)
Absolute left testis (g)[b]	1.5770 ± 0.0200	Not Measured	Not Measured	1.5301 ± 0.0167 (↓3)	1.5373 ± 0.0220 (↓3)	1.4476 ± 0.0141** (↓8)

Table A-9. Absolute and Relative Organ Weights in Male and Female F1 Pups in the 13-Week Toxicity Study of SAN Trimer[a]

Endpoint	Dose Group ppm (mg/kg-day)					
	0	**100 (10)**	**200 (20)**	**400 (40)**	**800 (80)**	**1,600 (150)**
Absolute thymus (g) (%)[b]	0.316 ± 0.009	0.337 ± 0.014 (↑7)	0.298 ± 0.021 (↓6)	0.299 ± 0.008 (↓5)	0.302 ± 0.011 (↓4)	0.267 ± 0.015 (↓16)
Relative thymus (%)[b]	0.938 ± 0.034	0.980 ± 0.038 (↑4)	0.873 ± 0.060 (↓7)	0.888 ± 0.025 (↓5)	0.900 ± 0.030 (↓4)	0.882 ± 0.041 (↓6)
Females						
Number of animals	10	10	10	10	10	10
Necropsy body weight (g) (%)[b]	203 ± 4	202 ± 3 (0)	192* ± 2 (↓5)	196* ± 2 (↓3)	190** ± 1 (↓6)	184** ± 2 (↓9)
Absolute brain (g) (%)[b]	1.823 ± 0.013	1.813 ± 0.017 (↓1)	1.798 ± 0.018 (↓1)	1.845 ± 0.015 (↑1)	1.844 ± 0.010 (↑1)	1.841 ± 0.007 (↑1)
Relative brain ratio (%)[b]	9.022 ± 0.179	9.000 ± 0.136 (0)	9.372* ± 0.109 (↑4)	9.410* ± 0.070 (↑4)	9.705** ± 0.078 (↑8)	10.016** ± 0.109 (↑11)
Absolute heart (g) (%)[b]	0.64 ± 0.01	0.64 ± 0.02 (0)	0.62 ± 0.01 (↓3)	0.65 ± 0.01 (↑2)	0.64 ± 0.01 (0)	0.63 ± 0.01 (↓2)
Relative heart ratio (%)[b]	3.146 ± 0.074	3.190 ± 0.063 (↑1)	3.238 ± 0.045 (↑3)	3.291 ± 0.046 (↑5)	3.363 ± 0.046** (↑7)	3.412 ± 0.046** (↑8)
Absolute R. kidney (g) (%)[b]	0.68 ± 0.01	0.67 ± 0.01 (↓1)	0.66 ± 0.01 (↓3)	0.69 ± 0.02 (↑1)	0.67 ± 0.01 (↓1)	0.66 ± 0.01 (↓3)
Relative R. kidney ratio (%)[b]	3.362 ± 0.051	3.334 ± 0.027 (↓1)	3.417 ± 0.031 (↓2)	3.492 ± 0.073 (↑4)	3.547 ± 0.050** (↑6)	3.609 ± 0.019** (↑7)
Absolute liver (g) (%)[b]	6.54 ± 0.17	6.68 ± 0.08 (↑2)	6.40 ± 0.12 (↓2)	6.74 ± 0.09 (↑3)	6.69 ± 0.08 (↑2)	6.74 ± 0.09 (↑3)
Relative liver ratio (%)[b]	32.250 ± 0.404	33.145 ± 0.274 (↑3)	33.359 ± 0.554 (↑3)	34.328** ± 0.328 (↑6)	35.213** ± 0.395 (↑9)	36.652** ± 0.376 (↑14)
Absolute lung (g) (%)[b]	1.35 ± 0.07	1.28 ± 0.04 (↓5)	1.26 ± 0.03 (↓7)	1.24 ± 0.04 (↓8)	1.28 ± 0.06 (↓5)	1.20 ± 0.05 (↓11)
Relative lung ratio (%)[b]	6.615 ± 0.269	6.369 ± 0.189 (↓1)	6.539 ± 0.140 (↓1)	6.341 ± 0.164 (↓4)	6.740 ± 0.310 (↑2)	6.542 ± 0.291 (↓1)

Table A-9. Absolute and Relative Organ Weights in Male and Female F1 Pups in the 13-Week Toxicity Study of SAN Trimer[a]

Endpoint	Dose Group ppm (mg/kg-day)					
	0	**100 (10)**	**200 (20)**	**400 (40)**	**800 (80)**	**1,600 (150)**
Absolute spleen (g) (%)[b]	0.535 ± 0.013	0.523 ± 0.008 (↓2)	0.528 ± 0.021 (↓1)	0.533 ± 0.012 (↓0)	0.550 ± 0.010 (↑3)	0.579* ± 0.008 (↑8)
Relative spleen ratio (%)[b]	2.639 ± 0.032	2.596 ± 0.038 (↓2)	2.748 ± 0.099 (↑4)	2.712 ± 0.042 (↑3)	2.897** ± 0.055 (↑10)	3.147** ± 0.051 (↑19)
Absolute thymus (g) (%)[b]	0.264 ± 0.007	0.249 ± 0.006 (↓6)	0.225** ± 0.005 (↓15)	0.243 ± 0.008 (↓8)	0.242 ± 0.012 (↓8)	0.244 ± 0.011 (↓8)
Relative thymus (%)[b]	1.301 ± 0.029	1.235 ± 0.028 (↓5)	1.174 ± 0.033 (↓10)	1.234 ± 0.033 (↓5)	1.275 ± 0.062 (↓2)	1.326 ± 0.052 (↑2)
Absolute uterus (g) (%)[b]	0.558 ± 0.070	0.564 ± 0.054 (↑1)	0.513 ± 0.066 (↓8)	0.655 ± 0.067 (↑17)	0.463 ± 0.065 (↓17)	0.643 ± 0.101 (↑15)
Relative uterus ratio (%)[b]	2.797 ± 0.395	2.795 ± 0.266 (0)	2.667 ± 0.340 (↓5)	3.327 ± 0.325 (↑19)	2.432 ± 0.335 (↓13)	3.497 ± 0.548 (↑25)

[a]NTP (2012).
[b]Mean ± standard error (% change from control).

*Williams' or Dunnett's test $p \leq 0.05$.
**Williams' or Dunnett's test $p \leq 0.01$.

Table A-10. Hematology, Clinical Chemistry, and Urinalysis Data for F1 Rats in the 13-Week Toxicity Study of SAN Trimer[a,b]

Endpoint	Exposure Group (Adjusted Daily Dose, mg/kg-day)[c]					
	0	10	20	40	80	150
Males						
Number of animals	10	10	10	10	10	10
Hematology						
Hemoglobin (g/dL)	15.5 ± 0.2	15.7 ± 0.1	15.4 ± 0.2	15.5 ± 0.1	15.5 ± 0.2	15.0 ± 0.1*
Erythrocytes (10^6/µL)	9.25 ± 0.08	9.44 ± 0.08	9.32 ± 0.09	9.27 ± 0.10	9.31 ± 0.10	8.88 ± 0.10*
Mean cell volume (fL)	54.3 ± 0.2	54.3 ± 0.1	54.0 ± 0.2	54.4 ± 0.2	54.3 ± 0/2	55.4 ± 0.3**
Clinical Chemistry						
Urea nitrogen (mg/dL)	14.9 ± 0.6	13.6 ± 0.7	13.6 ± 0.4	13.9 ± 0.9	15.5 ± 0.7	15.4 ± 0.6
Creatinine (mg/dL)	0.55 ± 0.02	0.53 ± 0.02	0.51 ± 0.01	0.58 ± 0.02	0.57 ± 0.02	0.60 ± 0.01
Total protein (g/dL)	6.5 ± 0.1	6.6 ± 0.1	6.5 ± 0.1	6.5 ± 0.1	6.8 ± 0.1*	6.5 ± 0.1
Albumin (g/dL)	4.5 ± 0.1	4.5 ± 0.1	4.5 ± 0.0	4.5 ± 0.0	4.7 ± 0.1**	4.6 ± 0.0**
Cholesterol (mg/dL)	79 ± 2	82 ± 2	80 ± 2	81 ± 2	78 ± 1	69 ± 1**
Triglycerides (mg/dL)	255 ± 16	206 ± 15	210 ± 17	163 ± 12**	199 ± 16**	155 ± 11**
Alanine aminotransferase (IU/L)	60 ± 3	52 ± 2	49 ± 2**	45 ± 1**	47 ± 2**	43 ± 1**
Aspartate aminotransferase (IU/L)	77 ± 5	66 ± 3*	63 ± 1**	62 ± 2**	64 ± 4**	55 ± 2**
Bile acids (µmol/L)	14.7 ± 2.2	11.7 ± 1.1	11.2 ± 1.2	7.3 ± 0.9**	11.0 ± 1.5	10.3 ± 1.4
Urinalysis						
Glucose (mg/dL)	35 ± 3	38 ± 2	37 ± 2	41 ± 4	46 ± 3*	37 ± 2
Glucose/creatinine ratio	0.18 ± 0.01	0.18 ± 0.00	0.18 ± 0.01	0.19 ± 0.01	0.20 ± 0.01*	0.20 ± 0.01*
Protein (mg/dL)	164 ± 14	166 ± 7	170 ± 13	170 ± 15	240 ± 19**	227 ± 25*
Protein/creatinine ratio	0.88 ± 0.71	0.79 ± 0.03	0.84 ± 0.04	0.81 ± 0.05	1.05 ± 0.06	1.20 ± 0.06**
Alkaline phosphatase/creatinine ratio	0.09 ± 0.01	0.10 ± 0.01	0.11 ± 0.01	0.09 ± 0.01	0.08 ± 0.00	0.10 ± 0.01
Aspartate aminotransferase (IU/L)	30 ± 14	18 ± 1	19 ± 2	23 ± 2	28 ± 3**	35 ± 6**
Aspartate aminotransferase/ creatinine ratio	0.02 ± 0.01	0.01 ± 0.00	0.01 ± 0.00	0.01 ± 0.00	0.01 ± 0.00*	0.02 ± 0.00**
N-Acetyl-*β*-D-glucosaminidase/creatinine ratio	0.02 ± 0.01	0.01 ± 0.00	0.01 ± 0.00	0.01 ± 0.00	0.01 ± 0.00	0.01 ± 0.00
Females						
Number of animals	10	10	10	10	10	10
Hematology						
Hemoglobin (g/dL)	14.5 ± 0.1	14.8 ± 0.1	14.7 ± 0.1	14.7 ± 0.1	14.3 ± 0.1	14.1 ± 0.2
Erythrocytes (10^6/µL)	8.23 ± 0.07	8.43 ± 0.08	8.39 ± 0.06	8.38 ± 0.07	8.16 ± 0.06	8.02 ± 0.08
Mean cell volume (fL)	56.3 ± 0.2	56.2 ± 0.2	56.0 ± 0.1	56.3 ± 0.1	56.3 ± 0.2	56.9 ± 0.2

Table A-10. Hematology, Clinical Chemistry, and Urinalysis Data for F1 Rats in the 13-Week Toxicity Study of SAN Trimer[a,b]

Endpoint	Exposure Group (Adjusted Daily Dose, mg/kg-day)[c]					
	0	10	20	40	80	150
Clinical Chemistry						
Urea nitrogen (mg/dL)	15.5 ± 0.6	15.8 ± 0.4	17.6 ± 0.6	15.7 ± 0.5	16.6 ± 0.5	18.2 ± 0.6**
Creatinine (mg/dL)	0.52 ± 0.01	0.53 ± 0.02	0.57 ± 0.02*	0.59 ± 0.01**	0.58 ± 0.01**	0.58 ± 0.01**
Total protein (g/dL)	6.4 ± 0.1	6.7 ± 0.1	6.6 ± 0.1	6.7 ± 0.1	6.6 ± 0.1	6.6 ± 0.1
Albumin (g/dL)	4.7 ± 0.1	4.9 ± 0.1	4.8 ± 0.1	4.8 ± 0.1	4.8 ± 0.1	4.8 ± 0.1
Cholesterol (mg/dL)	79 ± 2	85 ± 1	83 ± 2	83 ± 2	77 ± 2	74 ± 2
Triglycerides (mg/dL)	85 ± 9	87 ± 8	72 ± 7	72 ± 7	65 ± 9*	45 ± 6**
Alanine aminotransferase (IU/L)	43 ± 2	52 ± 3	46 ± 2	42 ± 2	41 ± 2	39 ± 1
Aspartate aminotransferase (IU/L)	66 ± 1	67 ± 3	66 ± 2	62 ± 1	60 ± 1*	57 ± 3**
Bile acids (µmol/L)	25.1 ± 1.8	15.4 ± 1.8*	17.0 ± 1.2	19.8 ± 2.7	15.3 ± 3.1**	21.5 ± 2.8
Urinalysis						
Glucose (mg/dL)	26 ± 3	29 ± 3	26 ± 3	27 ± 3	24 ± 4	29 ± 3
Glucose/creatinine ratio	0.20 ± 0.01	0.21 ± 0.01	0.21 ± 0.00	0.20 ± 0.00	0.21 ± 0.00	0.20 ± 0.00
Protein (mg/dL)	44 ± 5	46 ± 5	41 ± 5	46 ± 7	45 ± 10	73 ± 13
Protein/creatinine ratio	0.33 ± 0.01	0.33 ± 0.01	0.33 ± 0.01	0.32 ± 0.02	0.35 ± 0.02	0.47 ± 0.04
Alkaline phosphatase/creatinine ratio	0.09 ± 0.01	0.10 ± 0.00	0.09 ± 0.01	0.08 ± 0.00	0.08 ± 0.00	0.06 ± 0.00**
Aspartate aminotransferase (IU/L)	11 ± 2	10 ± 2	9 ± 1	8 ± 2	8 ± 1	15 ± 2
Aspartate aminotransferase/ creatinine ratio	0.01 ± 0.00	0.01 ± 0.00	0.01 ± 0.00	0.01 ± 0.00	0.02 ± 0.00	0.01 ± 0.00
N-Acetyl-*β*-D-glucosaminidase/creatinine ratio	0.01 ± 0.00	0.01 ± 0.00	0.01 ± 0.00	0.01 ± 0.00	0.01 ± 0.00**	0.01 ± 0.00**

[a]NTP (2012).
[b]Data are presented as mean ± standard error. Statistical tests were performed on unrounded data.
[c]Doses calculated by study authors.

*Significantly different ($p \leq 0.05$) from the control group by Dunn's or Shirley's test
**$p \leq 0.01$

Table A-11. Hematology, Clinical, and Urine Parameters in the F344/N Rats Treated with SAN Trimer for up to 78 Weeks[a,b]

Parameter	Exposure Group, mg/kg-day			
	0	20	40	75 (males)/ 85 (females)
Hematology				
Male Rat—27 Weeks				
Hemoglobin (g/dL)	16.2 ± 0.1	15.6 ± 0.2	15.8 ± 0.2	15.5 ± 0.1*
Reticulocytes (10^3/µL	194.5 ± 9.9	199.4 ± 7.5	197.4 ± 6.0	237.8 ± 10.2*
Mean cell hemoglobin (pg)	16.9 ± 0.1	16.6 ± 0.1*	16.7 ± 0.1	16.8 ± 0.1
Mean cell hemoglobin concentration (g/dL)	31.6 ± 0.2	31.2 ± 0.2	31.2 ± 0.1	31.0 ± 0.1**
Segmented neutrophils (10^3/µL)	1.51±0.08	1.51 ± 0.08	1.34 ± 0.06	1.28 ± 0.04*
Male Rat—52 Weeks				
Hemoglobin (g/dL)	16.1 ± 0.2	15.8 ± 0.2	15.7 ± 0.1	15.6 ± 0.2
Reticulocytes (10^3/µL	194.1 ± 4.9	209.0 ± 9.5	215.7 ± 8.8	221.5± 9.7
Mean cell hemoglobin (pg)	17.2 ± 0.1	17.0± 0.1	17.0 ± 0.1	17.0 ± 0.1
Mean cell hemoglobin concentration (g/dL)	33.0 ± 0.2	32.5 ± 0.1	32.5 ± 0.1	32.6± 0.2
Segmented neutrophils (10^3/µL)	1.76±0.17	1.56± 0.13	1.64± 0.08	1.74 ± 0.16
Male Rat—78 Weeks				
Hemoglobin (g/dL)	15.7 ± 0.2	15.9 ± 0.2	14.7 ± 1.2	15.6 ± 0.2
Reticulocytes (10^3/µL	224.7 ± 13.9	241.0 ± 15.2	314.6 ± 83.3	222.5± 7.9
Mean cell hemoglobin (pg)	17.2 ± 0.1	17.3± 0.2	16.7 ± 0.3	17.1 ± 0.1
Mean cell hemoglobin concentration (g/dL)	32.9 ± 0.1	33.1 ± 0.3	32.6 ± 0.3	32.7± 0.1
Segmented neutrophils (10^3/µL)	1.51±0.13	1.29± 0.11	1.57± 0.18	1.64 ± 0.27
Urinalysis/Clinical Chemistry				
Male Rat—26 Weeks (Urinalysis)/ 27 Weeks (Clinical Chemistry)				
Creatinine (mg/dL)	0.63 ± 0.02	0.64 ± 0.02	0.65 ± 0.02	0.69 ± 0.01**
Glucose (mg/dL)	128 ± 3	133 ± 2	125 ± 2	145 ± 9*
Sodium (mEq/L)	148 ± 0	149 ± 0	149 ± 0	151 ± 0**
Chloride (mEq/L)	97 ± 0	98 ± 0	97 ± 0	100 ± 0**
Albumin (g/dL)	4.6 ± 0.0	4.8 ± 0.0*	4.9 ± 0.1**	4.9 ± 0.1**
Triglycerides (mg/dL)	315 ± 27	249 ± 13	220 ± 14**	200 ± 12**
Alanine aminotransferase (IU/L)	144 ± 8	80 ± 3**	73 ± 2**	65 ± 2**
Alkaline phosphatase (IU/L)	169 ± 4	154 ± 3*	156 ± 4*	158 ± 4
Aspartate aminotransferase (IU/L)	122 ± 8	69 ± 3**	69 ± 3**	64 ± 3**
Lactate dehydrogenase (IU/L)	187 ± 19	133 ± 32	235 ± 30	111 ± 11*
Sorbitol dehydrogenase (IU/L)	29 ± 1	17 ± 1**	19 ± 1**	17 ± 1**

Table A-11. Hematology, Clinical, and Urine Parameters in the F344/N Rats Treated with SAN Trimer for up to 78 Weeks[a,b]

Parameter	Exposure Group, mg/kg-day			
	0	20	40	75 (males)/ 85 (females)
Male Rat—52 Weeks				
Creatinine (mg/dL)	195.2 ± 8.3	179.1 ± 8.1	168.7 ± 10.2	194.4 ± 9.9
Glucose (mg/dL)	39 ± 2	36 ± 2	35 ± 3	39 ± 2
Sodium (mEq/L)	149 ± 0	150 ± 0	149 ± 0	149 ± 0
Chloride (mEq/L)	98 ± 0	98 ± 0	98 ± 0	99 ± 0
Albumin (g/dL)	4.7 ± 0.0	4.8 ± 0.1	4.9 ± 0.0**	4.9 ± 0.1*
Triglycerides (mg/dL)	251 ± 15	255 ± 17	225 ± 13	224 ± 12
Alanine aminotransferase (IU/L)	123 ± 4	100 ± 8**	83 ± 4**	75 ± 3**
Alkaline phosphatase (IU/L)	152 ± 5	153 ± 4	147 ± 3	152 ± 4
Aspartate aminotransferase (IU/L)	122 ± 8	89 ± 4**	73 ± 4**	69 ± 2**
Lactate dehydrogenase (IU/L)	220 ± 35	203 ± 20	133 ± 25*	150 ± 52**
Sorbitol dehydrogenase (IU/L)	31 ± 1	27 ± 2*	22 ± 2**	19 ± 1**
Male Rat—78 Weeks				
Creatinine (mg/dL)	0.58 ± 0.01	0.63 ± 0.03	0.61 ± 0.01	0.64 ± 0.02*
Glucose (mg/dL)	141 ± 4	141 ± 3	133 ± 3	131 ± 3
Sodium (mEq/L)	151 ± 0	151 ± 0	151 ± 1	151 ± 0
Chloride (mEq/L)	101 ± 0	101 ± 1	101 ± 1	101 ± 0
Albumin (g/dL)	4.4 ± 0.0	4.3 ± 0.1	4.3 ± 0.1	4.4 ± 0.1
Triglycerides (mg/dL)	187 ± 9	194 ± 16	182 ± 8	155 ± 12
Alanine aminotransferase (IU/L)	151 ± 12	111 ± 9*	77 ± 7*	73 ± 5**
Alkaline phosphatase (IU/L)	151 ± 3	144 ± 10	144 ± 6	143 ± 6
Aspartate aminotransferase (IU/L)	143 ± 12	158 ± 45	89 ± 4**	88 ± 4**
Lactate dehydrogenase (IU/L)	210 ± 23	404 ± 186	154 ± 13	177 ± 18
Sorbitol dehydrogenase (IU/L)	37 ± 2	40 ± 11*	23 ± 2**	26 ± 1**
Parameter	**Exposure Group, mg/kg-day**			
	0	**20**	**40**	**85**
Hematology				
Female Rat—27 Weeks				
Hematocrit	47.1 ± 0.6	47.2 ± 0.5	46.7 ± 0.7	44.7 ± 0.7*
Hemoglobin (g/dL)	15.4 ± 0.2	15.3 ± 0.1	15.1 ± 0.2	14.5 ± 0.2**
Erythrocytes [b] ($\times 10^6$/MM^3)	8.52 ± 0.10	8.35 ± 0.08	8.31 ± 0.11	7.99 ± 0.11**
Mean cell hemoglobin (g/dl)	32.1 ± 0.1	32.3 ± 0.1	31.9 ± 0.2	31.9 ± 0.2
Platelets (10^3/µl)	615.9 ± 15.4	627.6 ± 31.1	579.9 ± 23.6	632.8 ± 19.3

Table A-11. Hematology, Clinical, and Urine Parameters in the F344/N Rats Treated with SAN Trimer for up to 78 Weeks[a,b]

Parameter	Exposure Group, mg/kg-day			
	0	20	40	75 (males)/ 85 (females)
Female Rat—52 Weeks				
Hematocrit	47.9 ± 0.5	46.9 ± 0.6	47.0 ± 0.5	45.3 ± 0.4**
Hemoglobin (g/dL)	16.0 ± 0.2	15.7 ± 0.2	15.6 ± 0.2	14.9 ± 0.2**
Erythrocytes [b] ($\times 10^6$/MM^3)	8.69 ± 0.09	8.53 ± 0.09	8.54 ± 0.08	8.16 ± 0.10**
Mean cell hemoglobin (g/dl)	33.4 ± 0.2	33.4 ± 0.2	33.1 ± 0.1	32.9 ± 0.1*
Platelets (10^3/µl)	625.9 ± 12.8	624.1 ± 18.2	648.8 ± 13.1	697.4 ± 3.9**
Female Rat—78 Weeks				
Hematocrit	49.0 ± 0.8	49.5 ± 0.7	47.0 ± 1.1	47.7 ± 0.7
Hemoglobin (g/dL)	15.8 ± 0.2	15.7 ± 0.2	15.0 ± 0.4	15.2 ± 0.2
Erythrocytes [b] ($\times 10^6$/MM^3)	8.45 ± 0.11	8.38 ± 0.11	8.07 ± 0.27	8.24 ± 0.15
Mean cell hemoglobin (g/dl)	33.3 ± 0.2	33.3 ± 0.1	33.2 ± 0.4	32.9 ± 0.2
Platelets (10^3/µl)	518.2 ± 15.7	507.3 ± 21.2	583.7 ± 47.7	514.1 ± 28.0
Urinalysis/Clinical Chemistry				
Female Rat—26 Weeks (Urinalysis)/ 27 Weeks (Clinical Chemistry)				
Urea nitrogen (mg/dL)	16.1 ± 0.6	16.0 ± 0.5	15.2 ± 0.3	16.6 ± 0.4
Creatinine (mg/dL)	0.67 ± 0.02	0.68 ± 0.01	0.70 ± 0.00	0.72 ± 0.02
Potassium (mEq/L)	5.2 ± 0.1	5.1 ± 0.2	5.1 ± 0.1	5.1 ± 0.1
Chloride (mEq/L)	100 ± 1	98 ± 1	99 ± 1	101 ± 1
Calcium (mg/dL)	11.6 ± 0.1	11.5 ± 0.1	11.7 ± 0.1	11.1 ± 0.1**
Phosphorus (mg/dL)	7.2 ± 0.2	6.8 ± 0.2	6.7 ± 0.3	5.9 ± 0.3**
Total protein (g/dL)	6.8 ± 0.1	7.1 ± 0.1	7.1 ± 0.1	6.8 ± 0.1
Albumin (g/dL)	4.8 ± 0.1	5.0 ± 0.1	5.0 ± 0.1	4.8 ± 0.0
Cholesterol (mg/dL)	115 ± 3	109 ± 3	111 ± 2	97 ± 2**
Triglycerides (mg/dL)	165 ± 17	121 ± 9	128 ± 11	72 ± 4**
Alanine aminotransferase (IU/L)	90 ± 8	55 ± 2**	56 ± 4**	54 ± 2**
Aspartate aminotransferase (IU/L)	91 ± 8	69 ± 3*	68 ± 4**	68 ± 2*
Sorbitol dehydrogenase (IU/L)	21 ± 1	17 ± 1*	18 ± 1*	15 ± 1**
Glucose/creatinine ratio	0.20 ± 0.01	0.19 ± 0.01	0.19 ± 0.00	0.19 ± 0.01
Protein (mg/dL)	36 ± 3	41 ± 3	51 ± 4**	53 ± 6*
Protein/creatinine ratio	0.28 ± 0.02	0.31 ± 0.01	0.35 ± 0.01**	0.40 ± 0.02**
Aspartate aminotransferase (IU/L)(Urine)	8 ± 1	8 ± 2	9 ± 1	11 ± 2
Female Rats—52 Weeks				
Urea nitrogen (mg/dL)	14.7 ± 0.4	15.1 ± 0.3	15.5 ± 0.5	16.5 ± 0.4**
Creatinine (mg/dL)	0.62 ± 0.01	0.69 ± 0.01**	0.68 ± 0.01**	0.73 ± 0.02**
Potassium (mEq/L)	5.5 ± 0.1	5.5 ± 0.1	5.6 ± 0.1	5.6 ± 0.1

Table A-11. Hematology, Clinical, and Urine Parameters in the F344/N Rats Treated with SAN Trimer for up to 78 Weeks[a,b]

Parameter	Exposure Group, mg/kg-day			
	0	20	40	75 (males)/ 85 (females)
Chloride (mEq/L)	99 ± 0	99 ± 0	98 ± 0	99 ± 0
Calcium (mg/dL)	12.3 ± 0.1	12.0 ± 0.1	12.2 ± 0.1	11.9 ± 0.1*
Phosphorus (mg/dL)	6.0 ± 0.2	5.1 ± 0.2*	5.3 ± 0.2*	5.1 ± 0.2**
Total protein (g/dL)	8.0 ± 0.1	8.0 ± 0.1	8.1 ± 0.1	7.8 ± 0.1
Albumin (g/dL)	5.6 ± 0.1	5.6 ± 0.1	5.7 ± 0.1	5.5 ± 0.1
Cholesterol (mg/dL)	139 ± 3	139 ± 2	137 ± 2	128 ± 3*
Triglycerides (mg/dL)	212 ± 11	208 ± 17	212 ± 30	178 ± 18
Alanine aminotransferase (IU/L)	69 ± 3	56 ± 2**	54 ± 2**	48 ± 1**
Aspartate aminotransferase (IU/L)	70 ± 6	56 ± 2	58 ± 3	55 ± 2*
Sorbitol dehydrogenase (IU/L)	18 ± 1	16 ± 1	15 ± 1*	15 ± 1**
Glucose/creatinine ratio	0.21 ± 0.01	0.23 ± 0.01	0.20 ± 0.01	0.20 ± 0.01
Protein (mg/dL)	66 ± 5	80 ± 20	71 ± 6	79 ± 9
Protein/creatinine ratio	0.42 ± 0.01	0.60 ± 0.12	0.49 ± 0.04	0.55 ± 0.04**
Female Rats—78 Weeks				
Urea nitrogen (mg/dL)	14.0 ± 0.6	14.6 ± 0.5	14.2 ± 0.3	14.9 ± 0.5
Creatinine (mg/dL)	0.58 ± 0.01	0.62 ± 0.01	0.62 ± 0.01	0.67 ± 0.02**
Potassium (mEq/L)	5.4 ± 0.1	4.9 ± 0.2*	5.2 ± 0.1	4.8 ± 0.1*
Chloride (mEq/L)	100 ± 0	100 ± 0	100 ± 0	99 ± 0*
Calcium (mg/dL)	12.0 ± 0.1	12.2 ± 0.1	12.0 ± 0.1	12.2 ± 0.1
Phosphorus (mg/dL)	6.4 ± 0.2	6.0 ± 0.1	5.7 ± 0.3	6.0 ± 0.3
Total protein (g/dL)	7.0 ± 0.1	7.3 ± 0.1*	7.5 ± 0.2**	7.4 ± 0.1**
Albumin (g/dL)	4.8 ± 0.0	5.0 ± 0.1*	5.2 ± 0.1**	5.1 ± 0.1**
Cholesterol (mg/dL)	123 ± 4	134 ± 4	127 ± 4	125 ± 5
Triglycerides (mg/dL)	162 ± 15	177 ± 13	178 ± 21	138 ± 15
Alanine aminotransferase (IU/L)	71 ± 8	62 ± 9	49 ± 3*	60 ± 7
Aspartate aminotransferase (IU/L)	92 ± 10	79 ± 11	67 ± 5*	73 ± 5
Sorbitol dehydrogenase (IU/L)	23 ± 2	20 ± 2	18 ± 1*	22 ± 2
Glucose/creatinine ratio	0.24 ± 0.00	0.24 ± 0.01	0.23 ± 0.01	0.22 ± 0.01**
Protein (mg/dL)	83 ± 12	75 ± 14	93 ± 12	86 ± 8
Protein/creatinine ratio	0.58 ± 0.11	0.62 ± 0.11	0.66 ± 0.06	0.65 ± 0.06

[a]NTP (2012).
[b]Data are presented as mean ± standard error. Statistical tests were performed on unrounded data.

*Significantly different ($p \leq 0.05$) from the control group by Dunn's or Shirley's test
**$p \leq 0.01$

Table A-12. Incidence of Histopathological Endpoints in F344/N Rats Exposed to SAN Trimer by Diet for 2 Years[a]

Parameter[b]	Exposure Group							
	Males				Females			
	0	20	40	75	0	20	40	85
Brain and Spinal Cord								
Spinal root degeneration (severity score)	34/47 (1.0)	37/48 (1.1)	37/50 (1.2)	43*/50 (1.3)	43/49 (1.2)	40/50 (1.2)	42/50 (1.3)	45/49 (1.5)
Sciatic nerve degeneration (severity score)	37/50 (1.1)	40/50 (1.2)	41/50 (1.3)	43/50 (1.3)	28/49 (1.0)	35/49 (1.1)	43**/49 (1.1)	40*/50 (1.1)
Liver								
Angiectasis (severity score)	1/50 (2.0)	5/50 (1.0)	1/50 (1.0)	9*/50 (1.1)	5/50 (NR)	5/50 (NR)	3/50 (NR)	5/50 (NR)
Eosinophilic foci (severity score)	17/50 (NR)	19/50 (NR)	22/50 (NR)	33*/50 (NR)	23/50 (NR)	31/50 (NR)	30/50 (NR)	29/50 (NR)
Active chronic inflammation (severity score)	34/50 (1.0)	40/50 (1.1)	38/50 (1.1)	43*/50 (1.0)	41/50 (NR)	45/50 (NR)	43/50 (NR)	47/50 (NR)
Mixed cell foci (severity score)	6/50 (NR)	19**/50 (NR)	12/50 (NR)	20**/50 (NR)	4/50 (NR)	8/50 (NR)	7/50 (NR)	13*/50 (NR)
Bone Marrow								
Bone marrow hyperplasia (severity score)	24/50 (1.9)	24/50 (1.8)	24/50 (1.8)	37**/50 (1.6)	16/50 (1.8)	25/50 (1.8)	25*/50 (2.0)	38**/50 (1.5)
Inflammation, granulomatous (severity score)	0/50 (NR)	0/50 (NR)	0/50 (NR)	3/50 (2.0)	0/50 (NR)	0/50 (NR)	6*/50 (1.2)	2/50 (1.5)
Bladder								
Hyperplasia of transitional epithelium (severity score)	0 (NR)	1 (NR)	0 (NR)	0 (NR)	1 (1.0)	0 (NR)	0 (NR)	12** (2.3)

[a]NTP (2012).
[b]Incidence, reported by the study authors.

Severity Score, average severity grade of lesions in affected animals: 1 = minimal, 2 = mild, 3 = moderate, 4 = marked.

*Significantly different from control ($p \leq 0.05$) using the Poly-3 test, as reported by the study authors.
**Significantly different from control ($p \leq 0.01$) using the Poly-3 test, as reported by the study authors.

NR = Not reported.

Table A-13. Tumor Incidence in F344/N Rats Administered SAN Trimer by Diet for 2 Years[a]

Parameter	Exposure Group, mg/kg-d (Human Equivalency Dose, mg/kg-day)[b]							
	Males				Females			
	0 (0)	20 (5.6)	40 (11)	75 (20)	0 (0)	20 (5)	40 (9.8)	85 (20)
Sample size	**50**	**50**	**50**	**50**	**50**	**50**	**50**	**50**
Brain and Spinal Cord								
Astrocytomas[c]	0	0	1 (2)	2 (4)	NR	NR	NR	NR
Granular cell tumors[c]	0	1 (2)	1 (2)	1 (2)	0	1 (2)	1 (2)	0
Glioma, Mixed Cell	NR	NR	NR	NR	1 (2)	1 (2)	1 (2)	0
Granular cell tumor or meningioma	NR	NR	NR	NR	0	1 (2)	1 (2)	0
Pituitary gland								
Adenoma[c,e,f]	16 (32)	10 (20)	13 (26)	4 (8)[d]	22 (44)	12 (24)[d]	19 (38)	9 (18)[d]
Adenoma or carcinoma[c,f]	NR	NR	NR	NR	22 (44)	12 (24)[d]	20 (40)	9 (18)[d]
Testes								
Bilateral, interstitial cell, adenoma[c,g]	33 (66)	45 (90)[h]	39 (78)	49 (98)[h]	NA	NA	NA	NA
Unilateral, interstitial cell, adenoma[c]	8 (16)	4 (8)	5 (10)	0 (0)	NA	NA	NA	NA
Overall, interstitial cell, adenoma (bilateral + unilateral, combined)[c,e]	41 (82)	49 (98)[d]	44 (88)	49 (98)[d]	NA	NA	NA	NA
Mammary gland								
Fibroadenoma[c,f]	1 (2)	2 (4)	1 (20)	3 (6)	36 (72)	31 (62)	26 (52)[d]	20 (40)[d]
Fibroadenoma or adenoma[c,f]	NR	NR	NR	NR	36 (72)	31 (62)	27 (54)[d]	20 (40)[d]
Fibroadenoma, adenoma, or carcinoma[c,f]	NR	NR	NR	NR	36 (72)	31 (62)	27 (54)[d]	21 (42)[d]
All organs								
Mononuclear cell leukemia[c,e,f]	15 (30)	7 (14)[d]	5 (10)[d]	3 (6)[d]	13 (26)	2 (4)[d]	3 (6)[d]	2 (4)[d]
Benign neoplasms[c]	46 (92)	49 (98)	47 (94)	49 (98)	42 (84)	37 (74)	39 (78)	34 (68)[d]
Malignant neoplasms[c,e]	22 (44)	17 (34)	11 (22)[d]	8 (16)[d]	19 (38)	5 (10)[d]	6 (12)[d]	12 (24)
Benign or malignant neoplasms[c,f]	49 (98)	50 (100)	49 (98)	49 (98)	44 (88)	41 (82)	41 (82)	35 (70)[d]

[a]NTP (2012).
[b]Doses were converted from adjusted daily doses to human equivalency doses using the following formula: $Dose_{HED} = Dose_{ADJ} \times (\text{body weight animal} \div \text{body weight human})^{(0.25)}$.
[c]Incidence, (corresponding percentage).
[d]Significantly different from control ($p \leq 0.05$) using the Poly-3 test, as reported by the study authors.
[e]Statistically significant trend using Poly-3 test for dose-response relationship in male rats, as reported by the study authors.
[f]Statistically significant trend using Poly-3 test for dose-response relationship in female rats, as reported by the study authors.
[g]Statistically significant trend using Cochran-Armitage test for dose-response relationship in male rats, as performed for this PPRTV assessment.
[h]Significantly different from control ($p \leq 0.05$) using the Fisher's Exact test, as performed for this PPRTV assessment.

NA = Not applicable, NR = Not reported.

APPENDIX B. BENCHMARK DOSE CALCULATIONS FOR THE SUBCHRONIC p-RfD AND CHRONIC p-RfD

MODELING PROCEDURE FOR DICHOTOMOUS DATA

The BMD modeling of dichotomous data was conducted with the U.S. EPA's BMDS (version 2.2.2). For these data, all of the dichotomous models (i.e., Gamma, Multistage, Logistic, Log-logistic, Probit, Log-probit, and Weibull models) available within the software were fit using a default BMR of 10% extra risk based on the U.S. EPA's *Benchmark Dose Technical Guidance Document* (U.S. EPA, 2012). Adequacy of model fit was judged based on the χ^2 goodness-of-fit *p*-value ($p > 0.1$), magnitude of scaled residuals in the vicinity of the BMR, and visual inspection of the model fit. Among all models providing adequate fit, the lowest BMDL was selected if the BMDLs estimated from different models varied greater than 3-fold; otherwise, the BMDL from the model with the lowest AIC was selected as a potential POD from which to derive a p-RfD.

In addition, data from exposures much higher than the study LOAEL do not provide reliable information regarding the shape of the response curve at low doses. However, such exposures can have a strong effect on the shape of the fitted model in the low-dose region of the dose-response curve in some cases. Thus, if lack of fit is due to characteristics associated with dose-response data for high doses, then the U.S. EPA's *Benchmark Dose Technical Guidance Document* allows for data to be adjusted by eliminating high-dose groups (U.S. EPA, 2012).

MODELING PROCEDURE FOR CONTINUOUS DATA

The BMD modeling of continuous data from the NTP (2012) study was conducted with the U.S. EPA's BMDS (version 2.2.2). For these data, all continuous models available within the software were fit using a default BMR of 1 SD relative risk. For changes in liver, body, and kidney weights, a BMR of 10% change relative to the control mean was also used. An adequate fit was judged based on the goodness-of-fit *p*-value ($p > 0.1$), magnitude of the scaled residuals in the vicinity of the BMR, and visual inspection of the model fit. In addition to these three criteria for judging adequacy of model fit, a determination was made as to whether the variance across dose groups was constant. If a constant variance model was deemed appropriate based on the statistical test provided in BMDS (i.e., Test 2), the final BMD results were estimated from a constant variance model. If the test for homogeneity of variance was rejected ($p < 0.1$), the model was run again while modeling the variance as a power function of the mean to account for this nonconstant variance. If this nonconstant variance model did not adequately fit the variance data (i.e., Test 3; $p < 0.1$), the data set was considered unsuitable for BMD modeling. Among all models providing adequate fit, the lowest BMDL was selected if the BMDLs estimated from different models varied greater than 3-fold; otherwise, the BMDL from the model with the lowest AIC was selected as a potential POD from which to derive a p-RfD.

INCREASED ABSOLUTE LIVER WEIGHT IN MALE F344/N RATS TREATED WITH SAN TRIMER FOR 13 WEEKS (NTP, 2012)

All available continuous models in BMDS (version 2.2.2) were fit to the increased absolute liver weight data from male F344/N rats exposed to SAN Trimer for 13 weeks (NTP, 2012) (see Table A-9). For increased absolute liver weight, a BMR of a 10% change relative to the control mean was used. In addition, a BMR of 1 SD relative risk was also estimated for comparison purposes based on the U.S. EPA's BMD guidance (U.S. EPA, 2012). For increased

absolute liver weight in male F1 rats, data were modeled without the highest dose of 150 mg/kg-day because there was a statistically significant decrease in body weight at that dose that confounds the interpretation of organ weight changes. Therefore, only the BMD modeling results based on data without the highest dose group are summarized in Table B-1 and Figure B-1. As assessed by the χ^2 goodness-of-fit statistic, AIC score, and visual inspection, after excluding the highest dose group, the Hill model provided the best model fit based on the lowest AIC and BMDL (see Table B-1 and Figure B-1). Test 2 indicated that using a constant variance model was appropriate for modeling these data. Estimated doses associated with a 10% BMR and the 95% lower confidence limit on these doses (BMD_{10} and $BMDL_{10}$ values, respectively) were 25 and 10 mg/kg-day.

Table B-1. Model Predictions for Absolute Liver Weight in Male Rats[a]									
Model[b]	**BMD_{10}**	**$BMDL_{10}$**	**BMD_{1SD}**	**$BMDL_{1SD}$**	***p*-Value Test 2[b]**	***p*-Value Test 3[b]**	**Goodness-of-Fit *p*-Value[b]**	**AIC**	**Conclusion**
Exponential (M2)	59	45	39	30	0.215	0.215	0.174	29.38	
Exponential (M3)	59	45	39	30	0.215	0.215	0.174	29.38	
Exponential (M4)	27	12	15	6.8	0.215	0.215	0.594	27.45	
Exponential (M5)	27	12	15	6.8	0.215	0.215	0.594	27.45	
Hill	**25**	**10**	**13**	**5.0**	**0.215**	**0.215**	**0.721**	**27.06**	**Lowest AIC**
Power	57	43	37	28	0.215	0.215	0.200	29.05	
Polynomial	57	43	37	28	0.215	0.215	0.200	29.05	
Linear	57	43	37	28	0.215	0.215	0.200	29.05	

[a]NTP (2012) 13-week study.
[b]Values <0.10 fail to meet conventional goodness-of-fit criteria.

AIC = Akaike's Information Criteria; BMD = Benchmark dose; BMDL = Lower confidence limit (95%) on the benchmark dose.

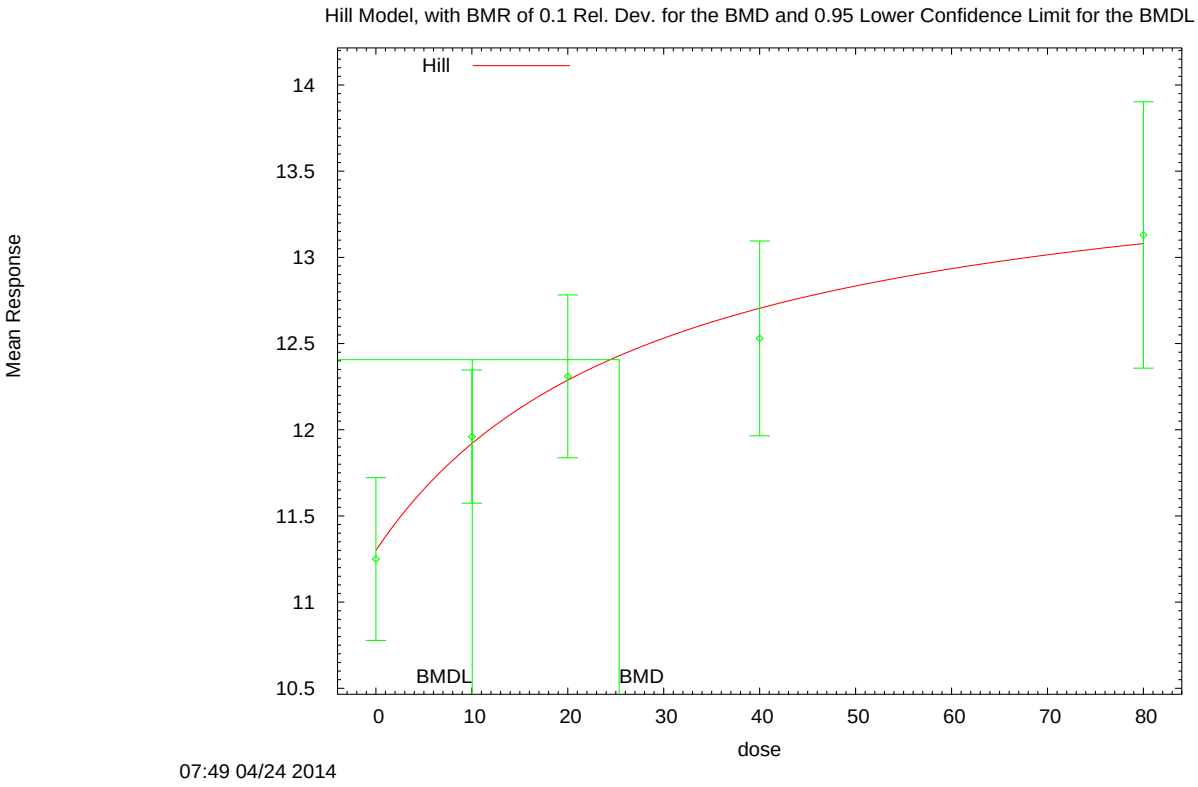

Figure B-1. Hill BMD Model for Increased Absolute Liver Weight Data in Male Rats at 13 Weeks
(NTP, 2012)

Text Output for Hill BMD Model for Increased Absolute Liver Weight in Male Rats Treated with SAN Trimer for 13 weeks (NTP, 2012)

```
================================================================
        Hill Model. (Version: 2.17;  Date: 01/28/2013)
        Input Data File: C:/Users/JKaiser/Desktop/modeling
results/hil_aliver_sant_mf1_nhd_Hil-ConstantVariance-BMR10-Restrict.(d)
        Gnuplot Plotting File:  C:/Users/JKaiser/Desktop/modeling
results/hil_aliver_sant_mf1_nhd_Hil-ConstantVariance-BMR10-Restrict.plt
                                          Wed Apr 30 14:10:33 2014
 ================================================================

 BMDS Model Run
~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~

   The form of the response function is:

   Y[dose] = intercept + v*dose^n/(k^n + dose^n)


   Dependent variable = mean
   Independent variable = dose
   rho is set to 0
   Power parameter restricted to be greater than 1
   A constant variance model is fit

   Total number of dose groups = 5
   Total number of records with missing values = 0
   Maximum number of iterations = 500
   Relative Function Convergence has been set to: 1e-008
   Parameter Convergence has been set to: 1e-008



                  Default Initial Parameter Values
                          alpha =      0.59066
                            rho =            0   Specified
                      intercept =        11.25
                              v =         1.88
                              n =     0.667711
                              k =      23.4286


           Asymptotic Correlation Matrix of Parameter Estimates

           ( *** The model parameter(s)  -rho    -n
                 have been estimated at a boundary point, or have been specified by
the user,
                 and do not appear in the correlation matrix )

                   alpha    intercept            v            k

     alpha             1    -2.6e-007    -1.9e-007    -3.8e-007

 intercept    -2.6e-007            1        0.033         0.57

         v    -1.9e-007        0.033            1         0.79

         k    -3.8e-007         0.57         0.79            1
```

Parameter Estimates

Variable	Estimate	Std. Err.	95.0% Wald Confidence Interval Lower Conf. Limit	Upper Conf. Limit
alpha	0.538601	0.10772	0.327473	0.749729
intercept	11.2787	0.233129	10.8217	11.7356
v	2.42814	0.671689	1.11165	3.74462
n	1	NA		
k	29.2305	23.7117	-17.2436	75.7046

NA - Indicates that this parameter has hit a bound
implied by some inequality constraint and thus
has no standard error.

Table of Data and Estimated Values of Interest

Dose	N	Obs Mean	Est Mean	Obs Std Dev	Est Std Dev	Scaled Res.
0	10	11.3	11.3	0.66	0.734	-0.124
10	10	12	11.9	0.54	0.734	0.269
20	10	12.3	12.3	0.66	0.734	0.193
40	10	12.5	12.7	0.79	0.734	-0.653
80	10	13.1	13.1	1.08	0.734	0.314

Model Descriptions for likelihoods calculated

Model A1: Yij = Mu(i) + e(ij)
Var{e(ij)} = Sigma^2

Model A2: Yij = Mu(i) + e(ij)
Var{e(ij)} = Sigma(i)^2

Model A3: Yij = Mu(i) + e(ij)
Var{e(ij)} = Sigma^2
Model A3 uses any fixed variance parameters that
were specified by the user

Model R: Yi = Mu + e(i)
Var{e(i)} = Sigma^2

Likelihoods of Interest

Model	Log(likelihood)	# Param's	AIC
A1	-9.203119	6	30.406238
A2	-6.306204	10	32.612408
A3	-9.203119	6	30.406238
fitted	-9.530512	4	27.061024
R	-22.901815	2	49.803629

Explanation of Tests

```
 Test 1:  Do responses and/or variances differ among Dose levels?
          (A2 vs. R)
 Test 2:  Are Variances Homogeneous? (A1 vs A2)
 Test 3:  Are variances adequately modeled? (A2 vs. A3)
 Test 4:  Does the Model for the Mean Fit? (A3 vs. fitted)
 (Note:  When rho=0 the results of Test 3 and Test 2 will be the same.)

                    Tests of Interest

   Test    -2*log(Likelihood Ratio)  Test df        p-value

   Test 1              33.1912          8          <.0001
   Test 2              5.79383          4          0.2151
   Test 3              5.79383          4          0.2151
   Test 4             0.654786          2          0.7208

The p-value for Test 1 is less than .05.  There appears to be a
difference between response and/or variances among the dose levels
It seems appropriate to model the data

The p-value for Test 2 is greater than .1.  A homogeneous variance
model appears to be appropriate here


The p-value for Test 3 is greater than .1.  The modeled variance appears
 to be appropriate here

The p-value for Test 4 is greater than .1.  The model chosen seems
to adequately describe the data


        Benchmark Dose Computation

Specified effect =           0.1

Risk Type        =     Relative deviation

Confidence level =           0.95

             BMD =        25.3548

            BMDL =       10.0451
```

INCREASED INCIDENCE OF CHRONIC ACTIVE HEPATIC INFLAMMATION IN MALE F334/N RATS TREATED WITH SAN TRIMER FOR 2 YEARS (NTP, 2012)

All available dichotomous models in BMDS (version 2.2.2) were fit to the chronic active hepatic inflammation data from male F344/N rats treated with SAN Trimer for 2 years (NTP, 2012) (see Table A-12). A BMR of 10% extra risk was used in all model runs. As assessed by the χ^2 goodness-of-fit statistic, AIC score, and visual inspection, the Log-Logistic model provided the best model fit (see Table B-2 and Figure B-2). Estimated doses associated with 10% extra risk and the 95% lower confidence limit on these doses (BMD_{10} and $BMDL_{10}$ values, respectively) were 8.0 and 3.2 mg/kg-day.

Table B-2. Model Predictions for Chronic Active Hepatic Inflammation in Males[a]

Model	BMD_{10}	$BMDL_{10}$	χ^2 *p*-Value	AIC	Conclusion
Gamma	11	5.7	0.575	213.45	
Logistic	12	6.8	0.571	213.47	
Log-Logistic[b]	**8.0**	**3.2**	**0.579**	**213.42**	**Lowest AIC**
LogProbit	21	11	0.480	213.84	
Multistage	11	5.7	0.575	213.45	
Probit	12	7.3	0.570	213.48	
Weibull	11	5.7	0.575	213.45	

[a]NTP (2012) 2-year study.
[b]The Log-Logistic model was selected as a best-fitting model based on lowest AIC.

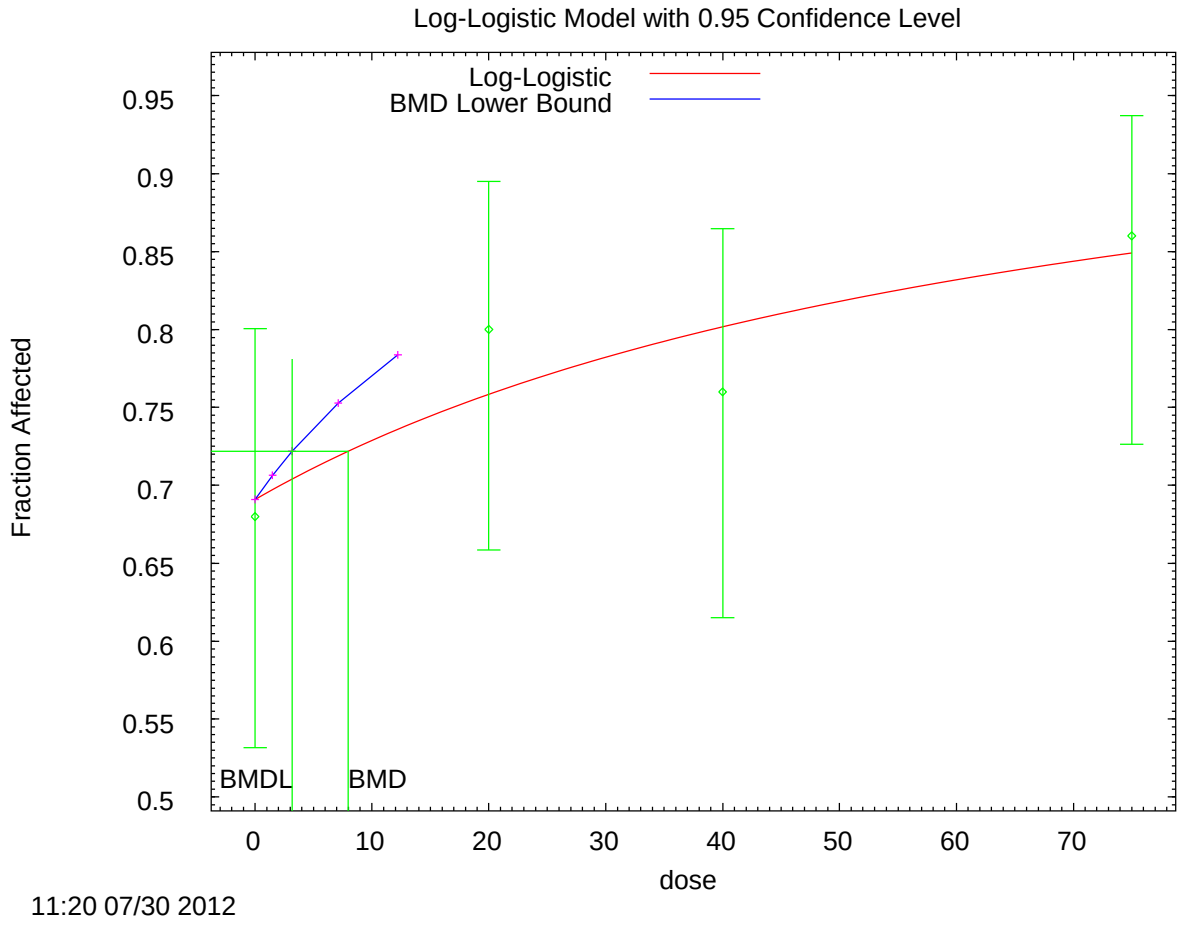

Figure B-2. Log-Logistic Model Fit for Chronic Active Hepatic Inflammation in Males (NTP, 2012)

Text Output for Log-Logistic BMD Model for Increased Incidence of Chronic Active Hepatic Inflammation in Male Rats Treated with SAN Trimer for 2 Years (NTP, 2012)

```
================================================================
         Logistic Model. (Version: 2.13; Date: 10/28/2009)
         Input Data File:
C:/USEPA/BMDS220/BMDS220/Data/SessionFiles/lnl_santa_cai_m_Lnl-BMR10-Restrict.(d)
         Gnuplot Plotting File:
C:/USEPA/BMDS220/BMDS220/Data/SessionFiles/lnl_santa_cai_m_Lnl-BMR10-Restrict.plt
                                                Mon Jul 30 11:20:23 2012
 ================================================================

 BMDS_Model_Run
~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~

   The form of the probability function is:

   P[response] = background+(1-background)/[1+EXP(-intercept-slope*Log(dose))]


   Dependent variable = Response
   Independent variable = Dose
   Slope parameter is restricted as slope >= 1

   Total number of observations = 4
   Total number of records with missing values = 0
   Maximum number of iterations = 250
   Relative Function Convergence has been set to: 1e-008
   Parameter Convergence has been set to: 1e-008



   User has chosen the log transformed model


                  Default Initial Parameter Values
                     background =         0.68
                      intercept =     -4.10081
                          slope =            1


           Asymptotic Correlation Matrix of Parameter Estimates

           ( *** The model parameter(s)  -slope
                 have been estimated at a boundary point, or have been specified by
the user,
                 and do not appear in the correlation matrix )

             background    intercept

background            1        -0.75

 intercept        -0.75            1


                                Parameter Estimates

                                                        95.0% Wald Confidence
Interval
       Variable         Estimate        Std. Err.     Lower Conf. Limit   Upper Conf.
Limit
```

background	0.690985	*	*	*
intercept	-4.27239	*	*	*
slope	1	*	*	*

* - Indicates that this value is not calculated.

Analysis of Deviance Table

Model	Log(likelihood)	# Param's	Deviance	Test d.f.	P-value
Full model	-104.166	4			
Fitted model	-104.71	2	1.08917	2	0.5801
Reduced model	-106.633	1	4.93401	3	0.1767

AIC: 213.421

Goodness of Fit

Dose	Est._Prob.	Expected	Observed	Size	Scaled Residual
0.0000	0.6910	34.549	34.000	50	-0.168
20.0000	0.7584	37.919	40.000	50	0.687
40.0000	0.8017	40.083	38.000	50	-0.739
75.0000	0.8490	42.449	43.000	50	0.218

Chi^2 = 1.09 d.f. = 2 P-value = 0.5788

Benchmark Dose Computation

Specified effect = 0.1

Risk Type = Extra risk

Confidence level = 0.95

BMD = 7.96586

BMDL = 3.16409

INCREASED INCIDENCE OF SCIATIC NERVE DEGENERATION IN FEMALE F334/N RATS TREATED WITH SAN TRIMER FOR 2 YEARS (NTP, 2012)

All available dichotomous models in BMDS (version 2.2.2) were fit to the sciatic nerve degeneration data from female F344/N rats treated with SAN Trimer for 2 years (NTP, 2012) (see Table A-12). A BMR of 10% extra risk was used in all model runs. The initial modeling of these data including all dose groups failed to provide an adequate fit to the data, as assessed by the χ^2 goodness-of-fit test. Therefore, only the BMD modeling results based on data without the high-dose group included are summarized in Table B-3 and Figure B-3. As assessed by the χ^2 goodness-of-fit statistic, AIC score, and visual inspection, after excluding the high-dose group (85 mg/kg-day) in order to improve fit, the Probit model provided the best model fit (see Table B-3 and Figure B-3). Estimated doses associated with 10% extra risk and the 95% lower confidence limit on these doses (BMD_{10} and $BMDL_{10}$ values, respectively) were 4.7 and 3.5 mg/kg-day.

Table B-3. Model Predictions for Sciatic Nerve Degeneration in Females[a]

Model	BMD_{10}	$BMDL_{10}$	χ^2 *p*-Value	AIC	Conclusion
Gamma	9.6	2.5	NDr	167.99	
Logistic	4.5	3.2	0.605	166.26	
Log-Logistic	10	1.6	NDr	167.99	
LogProbit	11	4.7	NDr	167.99	
Multistage	7.8	2.5	NDr	167.99	
Probit[b]	**4.7**	**3.5**	**0.659**	**166.18**	**Lowest AIC**
Weibull	8.7	2.5	NDr	167.99	

[a]NTP (2012) 2-year study.
[b]The Probit model was selected as a best-fitting model based on lowest AIC.

NDr = Not determined.

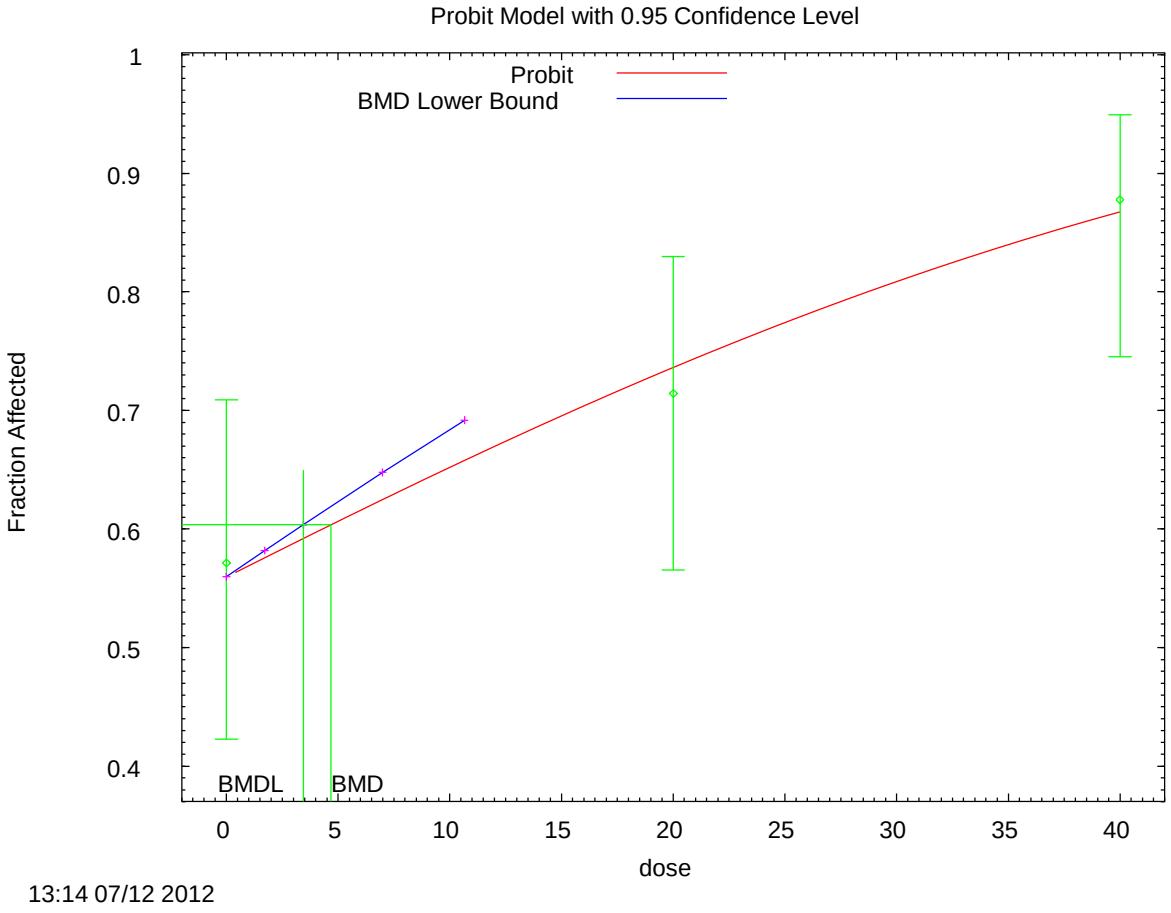

Figure B-3. Probit Model Fit for Sciatic Nerve Degeneration in Females (NTP, 2012)

Text Output for Probit BMD Model for Increased Incidence of Sciatic Nerve Degeneration in Female Rats Treated with SAN Trimer for 2 Years (NTP, 2012)

```
================================================================
        Probit Model. (Version: 3.2;  Date: 10/28/2009)
        Input Data File: C:/Documents and Settings/JKaiser/Desktop/modeling
results/pro_santa_srd_f_Pro-BMR10.(d)
        Gnuplot Plotting File:  C:/Documents and Settings/JKaiser/Desktop/modeling
results/pro_santa_srd_f_Pro-BMR10.plt
                                          Thu Jul 12 13:14:24 2012
 ================================================================

 BMDS_Model_Run
~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~

   The form of the probability function is:

   P[response] = CumNorm(Intercept+Slope*Dose),

   where CumNorm(.) is the cumulative normal distribution function


   Dependent variable = Response
   Independent variable = Dose
   Slope parameter is not restricted

   Total number of observations = 3
   Total number of records with missing values = 0
   Maximum number of iterations = 250
   Relative Function Convergence has been set to: 1e-008
   Parameter Convergence has been set to: 1e-008



                 Default Initial (and Specified) Parameter Values
                    background =            0   Specified
                     intercept =     0.144466
                         slope =    0.0241571


           Asymptotic Correlation Matrix of Parameter Estimates

           ( *** The model parameter(s)  -background
                 have been estimated at a boundary point, or have been specified by
the user,
                 and do not appear in the correlation matrix )

              intercept        slope

 intercept            1        -0.73

     slope        -0.73            1


                                Parameter Estimates

                                                        95.0% Wald Confidence
Interval
       Variable         Estimate        Std. Err.     Lower Conf. Limit   Upper Conf.
Limit
```

intercept	0.150347	0.166797	-0.176569	0.477264
slope	0.0241112	0.00717716	0.0100442	0.0381782

Analysis of Deviance Table

Model	Log(likelihood)	# Param's	Deviance	Test d.f.	P-value
Full model	-80.9947	3			
Fitted model	-81.0914	2	0.193293	1	0.6602
Reduced model	-87.0126	1	12.0357	2	0.002435

AIC: 166.183

Goodness of Fit

Dose	Est._Prob.	Expected	Observed	Size	Scaled Residual
0.0000	0.5598	27.428	28.000	49	0.165
20.0000	0.7365	36.088	35.000	49	-0.353
40.0000	0.8675	42.509	43.000	49	0.207

Chi^2 = 0.19 d.f. = 1 P-value = 0.6593

Benchmark Dose Computation

Specified effect = 0.1

Risk Type = Extra risk

Confidence level = 0.95

BMD = 4.67809

BMDL = 3.45186

APPENDIX C. DOSIMETRY CALCULATION EXAMPLES FOR F0 DAMS AND F1 PUPS IN THE 2-WEEK, 13-WEEK, AND 2-YEAR PERINATAL AND POSTNATAL FEED STUDIES OF SAN TRIMER (NTP, 2012)

NTP (2012) 2-WEEK STUDY

For this study, the study authors reported doses from GD 7 to 14, GD 15 to 18, PND 1 to 7, PND 8 to 14, and PND 15 to 20. A time-weighted average (TWA) dose was calculated for both the gestational and lactational components of the study.

Gestational Component:

$$\text{TWA Dose} = \frac{(D_1 \times T_1) + (D_2 \times T_2)}{T_1 + T_2}$$

Where

D1 = Dose received from GD 7 to 14 with T1 = 8 days
D2 = Dose received from GD 15 to 18 with T2 = 4 days.

Example for 250 ppm:

D1 = 17 mg/kg-day as calculated by the study authors, T1 = 8 days; D2 = 20 mg/kg-day as calculated by the study authors, T2 = 4 days.

$$\text{TWA Dose} = \frac{(17 \text{ mg/kg-day} \times 8 \text{ days}) + (20 \text{ mg/kg-day} \times 4 \text{ days})}{12 \text{ days}}$$

$$\text{TWA Dose} = 18 \text{ mg/kg-day}$$

Lactational Component:

$$\text{TWA Dose} = \frac{(D_1 \times T_1) + (D_2 \times T_2) + (D_3 \times T_3)}{T1 + T2 + T3}$$

Where

D_1 = Dose received from PND 1 to 7 with T_1 = 7 days
D_2 = Dose received from PND 8 to 14 with T_2 = 7 days
D_3 = Dose received from PND 15 to 20 with T_3 = 6 days.

Example for 250 ppm:

D_1 = 30 mg/kg-day as calculated by the study authors, T_1 = 7 days
D_2 = 43 mg/kg-day as calculated by the study authors, T_2 = 7 days
D_3 = 49 mg/kg-day as calculated by the study authors, T_2 = 6 days.

$$\text{TWA Dose} = \frac{(30 \text{ mg/kg-day} \times 7 \text{ days}) + (43 \text{ mg/kg-day} \times 7 \text{ days}) + (49 \text{ mg/kg-day} \times 6 \text{ days})}{20 \text{ days}}$$

TWA Dose = 40 mg/kg-day

NTP (2012) 13-WEEK STUDY

For the 13-week and 2-year studies (NTP, 2012), the study authors did not present food consumption data during gestation and lactation so it was not possible to calculate food factors for these studies. Therefore, the food factors were calculated from the body weight and food consumption data reported in dams from the 2-week NTP (2012) study and used to calculate gestational and lactational doses for the 13-week and 2-year NTP (2012) studies.

Dose calculation examples for the gestational component of the 13-week NTP (2012) study:

Table C-1. Average Food Factor Calculations for the Gestational Component of the 2-Week NTP (2012) Study

GD 7−14			
PPM	**Food Consumption/Day[a]**	**Body Weight[a]**	**Food Factor[b]**
0	13	191	0.068
250	14	196	0.071
500	12	185	0.065
1,000	12	197	0.061
2,000	11	190	0.058
Average food factor for GD 7−14 in the 2-week NTP study			**0.065**
GD 15−18			
PPM	**Food Consumption/Day[a]**	**Body Weight[a]**	**Food Factor[b]**
0	16	213	0.075
250	17	219	0.078
500	16	206	0.078
1,000	17	220	0.077
2,000	16	213	0.075
Average food factor for GD 15−18 in the 2-week NTP study			**0.077**

[a]Data were reported by the study authors.
[b]Food factor = Food consumption per day × (1 ÷ Body Weight).

Table C-2. Dose Calculations for the Gestational Component of the 13-Week NTP (2012) Study

PPM	Average Food Factor (GD 7−14)[a]	Dose (GD 7−14)[b]	Average Food Factor (GD 15−18)[a]	Dose (GD 15−18)[b]	TWA Dose[c]
0	0.065	0	0.077	0	0
100	0.065	6.5	0.077	7.7	6.9
200	0.065	13	0.077	15	14
400	0.065	26	0.077	31	28
800	0.065	52	0.077	62	55
1600	0.065	104	0.077	123	110

[a]As presented in Table C-1.
[b]Dose = Feed concentration × food factor × (days dosed ÷ total days).
[c]TWA doses were calculated as described above.

Dose calculation examples for the lactational component of the 13-week NTP (2012) study:

Table C-3. Average Food Factor Calculations for the Lactational Component of the 2-Week NTP (2012) Study

PND 1−7			
PPM	**Food Consumption/Day[a]**	**Body Weight[a]**	**Food Factor[b]**
0	24	204	0.12
250	26	214	0.12
500	25	199	0.13
1,000	26	210	0.12
2,000	23	202	0.11
Average food factor for PND 1−7 in the 2-week NTP study			**0.12**
PND 8−14			
PPM	**Food Consumption/Day[a]**	**Body Weight[a]**	**Food Factor[b]**
0	41	222	0.18
250	39	230	0.17
500	39	215	0.18
1,000	40	227	0.18
2,000	40	216	0.19
Average food factor for PND 8−14 in the 2-week NTP study			**0.18**
PND 15−20			
PPM	**Food Consumption/Day[a]**	**Body Weight[a]**	**Food Factor[b]**
0	47	234	0.20
250	46	238	0.19
500	47	226	0.21
1,000	48	238	0.20
2,000	43	222	0.19
Average food factor for PND 15−20 in the 2-week NTP study			**0.20**

[a]Data were reported by the study authors.
[b]Food factor = Food consumption per day × (1 ÷ Body Weight).

Table C-4. Dose Calculations for the Lactational Component of the 13-Week NTP (2012) Study

PPM	Average Food Factor (PND 1−7)[a]	Dose (PND 1−7)[b]	Average Food Factor (PND 8−14)[a]	Dose (PND 8−14)[b]	Average Food Factor (PND 15−20)[a]	Dose (PND 15−20)[b]	TWA Dose[c]
0	0.12	0	0.18	0	0.20	20	0
100	0.12	12	0.18	18	0.20	40	17
200	0.12	24	0.18	36	0.20	80	33
400	0.12	48	0.18	72	0.20	160	66
800	0.12	96	0.18	144	0.20	320	132
1600	0.12	192	0.18	288	0.20	20	264

[a]As presented in Table C-3.
[b]Dose = Feed concentration × food factor × (days dosed ÷ total days).
[c]TWA doses were calculated as described above.

NTP (2012) 2-YEAR STUDY

The same methodology used for the 13-week study was also used to perform dosimetry calculations for the gestational and lactational components of the 2-year NTP (2012) study and are not shown here.

APPENDIX D. REFERENCES

ATSDR (Agency for Toxic Substances and Disease Registry). (1997). Chemical specific health consultation: Toxicological issues related to chemicals by the New Jersey Department of Health and Senior Services. Atlanta, GA: Agency for Toxic Substances & Disease Registry.

ATSDR (Agency for Toxic Substances and Disease Registry). (1998). Public health concerns in Dover Township. ATSDR's public health response. Progress report, May 1998. Atlanta, GA: Agency for Toxic Substances & Disease Registry.

Behl, M; Elmore, SA; Malarkey, DE; Hejtmancik, MR; Gerken, DK; Chhabra, RS. (2013). Perinatal toxicity and carcinogenicity studies of styrene-acrylonitrile trimer, a ground water contaminant. Toxicology 314: 84-94. http://dx.doi.org/10.1016/j.tox.2013.09.006

Gargas, ML; Collins, B; Fennell, TR; Gaudette, NF; Sweeney, LM. (2008). Disposition of styrene-acrylonitrile (SAN) trimer in female rats: single dose intravenous and gavage studies. Toxicol Lett 178: 1-8. http://dx.doi.org/10.1016/j.toxlet.2008.01.016

Haseman, JK; Arnold, J; Eustis, SL. (1990). Tumor incidences in Fischer 344 rats. In GA Borman; SL Eustis; MR Elwell; CA Montgomery, Jr.; WF MacKenzie (Eds.), Pathology of the Fischer rat (pp. 555-564). San Diego, CA: Academic Press.

Hobbs, CA; Chhabra, RS; Recio, L; Streicker, M; Witt, KL. (2012). Genotoxicity of styrene-acrylonitrile trimer in brain, liver, and blood cells of weanling F344 rats. Environ Mol Mutagen 53: 227-238. http://dx.doi.org/10.1002/em.21680

Huntingdon Life Sciences. (1999a). SAN Trimer (AN2S): 14-day oral gavage toxicity study in the rat (final report November 19, 1999). Cambridgeshire, UK.

Huntingdon Life Sciences. (1999b). SAN Trimer: Acute oral LD50 study in rats (final report December 9, 1999).

IARC (International Agency for Research on Cancer). (1997). Dry cleaning, some chlorinated solvents and other industrial chemicals. In IARC monographs on the evaluation of caarcinogenic risks to humans. Geneva, Switzerland: World Health Organization. http://monographs.iarc.fr/ENG/Monographs/vol63/volume63.pdf

IRIS (Integrated Risk Information System). (2011). Summary sheet for trichloroethylene (CASRN 79-01-6). Washington, DC: U.S. Environmental Protection Agency, Integrated Risk Information System. http://www.epa.gov/iris/subst/0199.htm

IRIS (Integrated Risk Information System). (2012). Summary sheet for perchloroethylene (CASRN: 127-18-4). Washington, DC: U.S. Environmental Protection Agency, Integrated Risk Information System. http://www.epa.gov/iris/subst/0106.htm

MA Bioservices. (1998a). Bacterial Reverse Mutation Assay with an Independent Repeat Assay. Test article SAN trimer (AN2S) (draft report July 24, 1998). Rockville, MD.

MA Bioservices. (1998b). In vitro mammalian cell gene mutation test with an independent repeat assay. Test article SAN trimer (AN2S), draft report 9/4/1998. Rockville, MD.

MA Bioservices. (1998c). In vitro mammalian chromosome aberration test. Test article SAN trimer (AN2S), draft report 9/17/1998. Rockville, MD.

MA Bioservices. (1998d). Mammalian bone marrow chromosome aberration and micronucleus test. Test article SAN trimer (AN2S), draft report 7/30/1998. Rockville, MD.

NJ DHSS (New Jersey Department of Health and Senior Services). (1997). Childhood cancer incidence health consultation: A review and analysis of cancer registry data, 1979-1995 for Dover Township (Ocean County), New Jersey, December 1997. Trenton, NJ. http://www.state.nj.us/health/eoh/hhazweb/dovertwp.shtml

NJ DHSS (New Jersey Department of Health and Senior Services). (2001). Public health consultation: drinking water quality analyses, March 1996 to June 1999, united water Toms River. Dover Township, Ocean, New Jersey. Trenton, NJ: NJDHSS, Division of Epidemiology, Environmental and Occupational Health. http://www.state.nj.us/health/eohs/ocean/toms_river/toms_river_dover_twp/trdwhcfin.pdf

NJ DHSS (New Jersey Department of Health and Senior Services). (2003a). Case-control study of childhood cancers in Dover Township (Ocean County), New Jersey. Volume I: Summary of the final technical report. Trenton, NJ. http://www.state.nj.us/health/eohs/ocean/toms_river/toms_river_dover_twp/vol_i.pdf

NJ DHSS (New Jersey Department of Health and Senior Services). (2003b). Childhood cancer incidence update: A review and analysis of cancer registry data, 1979-2000 for Dover Township (Ocean County), New Jersey. Division of Epidemiology, Environmental and Occupational Health, Consumer and Environmental Services, Trenton, New Jersey. http://www.state.nj.us/health/eohs/ocean/toms_river/toms_river_dover_twp/tomsriverchildhoodcancerupdate.pdf

NJ DHSS (New Jersey Department of Health and Senior Services). (2008). Childhood cancer incidence update: A review and analysis of cancer registry data, 2001-2005 for Township of Toms River, Ocean County, New Jersey. http://www.state.nj.us/health/eohs/ocean/toms_river/toms_river_dover_twp/toms_river_childhood_cancer_update.pdf

NJDOH. (1996). The Dover Township Childhood Cancer Investigation, Public Health Response Plan, Trenton, New Jersey: New Jersey Department of Health. NJDOH. http://www.nj.gov/health/eohs/ocean/toms_river/toms_river_dover_twp/phrdover.pdf

NTP. (2012). Toxicology and carcinogenesis study of styrene-acrylonitrile trimer in F344/N rats (perinatal and postnatal feed studies). (NTP Technical Report 573). NTP. http://ntp.niehs.nih.gov/ntp/htdocs/LT_rpts/TR573_508.pdf

Richardson, SD; Collette, TW; Price, PC; Genicola, FA; Jenks, JW; Thruston, AD; Ellington, JJ. (1999). Identification of drinking water contaminants in the course of a childhood cancer investigation in Toms River, New Jersey. J Expo Anal Environ Epidemiol 9: 200-216.

Sills, RC; Hailey, JR; Neal, J; Boorman, GA; Haseman, JK; Melnick, RL. (1999). Examination of low-incidence brain tumor responses in F344 rats following chemical exposures in National Toxicology Program carcinogenicity studies. Toxicol Pathol 27: 589-599.

Takaki, Y; Kitamura, S; Uekusa, T; Honma, S; Aze, Y; Wakabayashi, K; Kuwabara, N; Fukuda, Y. (1989). Spontaneous tumors in F-344 rats Jcl rats. J Toxicol Sci 14: 181-195.

U.S. EPA (U.S. Environmental Protection Agency). (1988). Recommendations for and documentation of biological values for use in risk assessment. (EPA/600/6-87/008). Cincinnati, OH: U.S. Environmental Protection Agency, National Center for Environmental Assessment. http://cfpub.epa.gov/ncea/cfm/recordisplay.cfm?deid=34855

U.S. EPA (U.S. Environmental Protection Agency). (1991). Guidelines for developmental toxicity risk assessment. (EPA/600/FR-91/001). Washington, DC: U.S. Environmental Protection Agency, Risk Assessment Forum. http://www.epa.gov/raf/publications/guidelines-dev-toxicity-risk-assessment.htm

U.S. EPA (U.S. Environmental Protection Agency). (1996). Guidelines for reproductive toxicity risk assessment [EPA Report]. (EPA/630/R-96/009). Washington D.C.: Risk Assessment Forum, U.S. Environmental Protection Agency.

U.S. EPA (U.S. Environmental Protection Agency). (2002). A review of the reference dose and reference concentration processes. (EPA/630/P-02/002F). Washington, DC: U.S.

Environmental Protection Agency, Risk Assessment Forum. http://cfpub.epa.gov/ncea/cfm/recordisplay.cfm?deid=51717

U.S. EPA (U.S. Environmental Protection Agency). (2005). Guidelines for carcinogen risk assessment. (EPA/630/P-03/001F). Washington, DC: U.S. Environmental Protection Agency, Risk Assessment Forum. http://www.epa.gov/cancerguidelines/

U.S. EPA (U.S. Environmental Protection Agency). (2011). Recommended use of body weight 3/4 as the default method in derivation of the oral reference dose. (EPA/100/R11/0001). Washington, DC: U.S. Environmental Protection Agency, Risk Assessment Forum. http://www.epa.gov/raf/publications/interspecies-extrapolation.htm

U.S. EPA (U.S. Environmental Protection Agency). (2012). Benchmark dose technical guidance. (EPA/100/R-12/001). Washington, DC: Risk Assessment Forum. http://www.epa.gov/raf/publications/pdfs/benchmark_dose_guidance.pdf

Vacek, PM; Messier, T; Rivers, J; Sullivan, L; O'Neill, JP; Finette, BA. (2005). Somatic mutant frequency at the HPRT locus in children associated with a pediatric cancer cluster linked to exposure to two superfund sites. Environ Mol Mutagen 45: 339-345. http://dx.doi.org/10.1002/em.20101