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HEALTH AND ENVIRONMENTAL EFFECTS DOCUMENT FOR 1,3,5-TRINITROBENZENE

Prepared for

OFFICE OF SOLID WASTE AND
EMERGENCY RESPONSE

Prepared by

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PREFACE

Health and Environmental Effects Documents (HEEDs) are prepared for the Office of Solid Waste and Emergency Response (OSWER). This document series is intended to support listings under the Resource Conservation and Recovery Act (RCRA) as well as to provide health-related limits and goals for emergency and remedial actions under the Comprehensive Environmental Response, Compensation and Liability Act (CERCLA). Both published literature and information obtained for Agency Program Office files are evaluated as they pertain to potential human health, aquatic life and environmental effects of hazardous waste constituents. The literature searched for in this document and the dates searched are included in "Appendix: Literature Searched." Literature search material is current up to 8 months previous to the final draft date listed on the front cover. Final draft document dates (front cover) reflect the date the document is sent to the Program Officer (OSWER).

Several quantitative estimates are presented provided sufficient data are available. For systemic toxicants, these include Reference doses (RfDs) for chronic and subchronic exposures for both the inhalation and oral exposures. The subchronic or partial lifetime RfD is an estimate of an exposure level that would not be expected to cause adverse effects when exposure occurs during a limited time interval i.e., for an interval that does not constitute a significant portion of the lifespan. This type of exposure estimate has not been extensively used, or rigorously defined as previous risk assessment efforts have focused primarily on lifetime exposure scenarios. Animal data used for subchronic estimates generally reflect exposure durations of 30-90 days. The general methodology for estimating subchronic RfDs is the same as traditionally employed for chronic estimates, except that subchronic data are utilized when available.

In the case of suspected carcinogens, RfDs are not estimated. Instead, a carcinogenic potency factor, or q_1^* (U.S. EPA, 1980a), is provided. These potency estimates are derived for both oral and inhalation exposures where possible. In addition, unit risk estimates for air and drinking water are presented based on inhalation and oral data, respectively.

Reportable quantities (RQs) based on both chronic toxicity and carcinogenicity are derived. The RQ is used to determine the quantity of a hazardous substance for which notification is required in the event of a release as specified under the Comprehensive Environmental Response, Compensation and Liability Act (CERCLA). These two RQs (chronic toxicity and carcinogenicity) represent two of six scores developed (the remaining four reflect ignitability, reactivity, aquatic toxicity, and acute mammalian toxicity). Chemical-specific RQs reflect the lowest of these six primary criteria. The methodology for chronic toxicity and cancer based RQs are defined in U.S. EPA, 1984 and 1986a, respectively.

EXECUTIVE SUMMARY

1,3,5-Trinitrobenzene is a yellow crystalline solid at room temperature; it is soluble in both polar and nonpolar organic solvents and sparingly soluble in water (Sax and Lewis, 1987; Windholz et al., 1983). Information on current methods of production is lacking in the available literature. Only Eastman Kodak Co. in Rochester, NY, produced this material in 1977 (TSCAPP, 1989). 1,3,5-Trinitrobenzene is used mainly in explosive compositions (Sax and Lewis, 1987; Windholz et al., 1983). It is also used in the vulcanization of rubber (Barnhart, 1981) and as a pH indicator (Durst and Bates, 1981).

Sufficient data do not exist in the available literature to accurately predict the environmental fate of 1,3,5-trinitrobenzene but the following processes are expected to occur in the environment: In the atmosphere, 1,3,5-trinitrobenzene should exist partially in the vapor phase, and partially in the particulate form. Destruction by the gas-phase reaction with photochemically produced OH radical or by the reaction with ozone is not expected to be significant. Both wet and dry deposition may occur. In water, neither volatilization to the atmosphere nor hydrolysis are expected to be important processes. Information sufficient to predict the importance of microbial degradation of 1,3,5-trinitrobenzene were not located in the available literature; however, rapid anaerobic degradation may occur under the proper conditions. 1,3,5-Trinitrobenzene adsorbs light in the environmentally significant range >290 nm (Burlinson et al., 1973; Spanggord et al., 1980; Capellos and Suryanarayanan, 1973) but the light-induced transformations of this compound are not well understood. Therefore, the photolytic destruction of 1,3,5-trinitrobenzene in the environment cannot be accurately predicted.

1,3,5-Trinitrobenzene is a man-made organic compound that can enter the environment as a component of wastewater effluent of plants that synthesize, produce or demilitarize explosives or munitions. 1,3,5-Trinitrobenzene may also enter the environment through the disposal of solid wastes (Ryon et al., 1984, Spalding and Fulton, 1988; Spanggord et al., 1982a). Limited monitoring data are available on the concentration of this compound in the environment. Sufficient monitoring data are not available to estimate the exposure of 1,3,5-trinitirobenzene to the general population.

Existing data indicate that trinitrobenzene is highly toxic to aquatic fauna, tut the compound is not likely to concentrate in them. Acute toxicity data have been reported for four species of fish (fathead minnows, channel catfish, bluegills and rainbow trout) and one invertebrate, the water flea. LC_{50} values for fathead minnows range from 0.49-1.1 ppm, and the other three species displayed sensitivity to trinitrobenzene at LC_{50} levels <1.0 ppm and ranging from 0.38 to 0.85 (Bailey and Spanggord, 1983; Liu et al., 1983; Pearson et al., 1979; van der Schalie, 1983; van der Schalie et al., 1988). The water flea, D. magna, was less sensitive, showing EC_{50} values ranging from 2.7-4.1 ppm trinitrobenzene (Liu et al., 1983; Pearson et al., 1979; van der Schalie, 1983).

Chronic toxicity data are provided for fathead minnows, rainbow trout, bluegills, and the water flea. Early life stage tests indicate that fathead minnows are highly sensitive to trinitrobenzene, with survival significantly reduced at exposures $\geq$0.12 ppm for a period of 32 days. Rainbow trout were similarly sensitive to trinitrobenzene, showing a LOEL of 0.17 ppm for survival, fry length and fry weight (van der Schalie, 1983). Ventilatory effects were noted in bluegills exposed to treatment levels $\geq$0.128 ppm for 6 days (van der Schalie et al., 1988). Signs of respiratory distress

(opercular movement increases, excitability and violent swimming) were noted in K. sandvicensus exposed to ≥0.1 ppm trinitrobenzene for short durations (Hiatt et al., 1957).

A BCF of 6.36 has been calculated for trinitrobenzene by Liu et al. (1983) from the estimated log value of 1.36. Bioconcentration data reported by van der Schalie (1983) for fathead minnows, rainbow trout and the water flea support the conclusion that trinitrobenzene does not significantly bioaccumulate in aquatic animals.

Toxic effects of trinitrobenzene in the alga, S. capricornutum, was investigated by van der Schalie (1983). Significant reduction in growth was noted at all levels tested (0.01-17.32 ppm) after 5 and 14 days exposure, concentrations of 1.18 ppm were algicidal, and lower concentrations were algistatic.

Data pertinent to the pharmacokinetic behavior of 1,3,5-trinitrobenzene in mammalian systems are not available.

Information regarding the chronic or subchronic toxicity of 1,3,5-trinitrobenzene following inhalation exposure is unavailable. Information regarding oral exposure of chronic or subchronic duration is limited to an abstract of a Russian report (Korolev et al., 1977). "Prolonged" oral administration of 1,3,5-trinitrobenzene to mice, rats and guinea pigs altered the activities of peroxidase, alkaline phosphatase and ceroplasmin in the blood, but further details were unavailable.

Published oral LD_{50} values for 1,3,5-trinitrobenzene include the following: 600 mg/kg in white mice, 450 mg/kg in white rats, 730 mg/kg in guinea pigs (Korolev et al., 1977) and 600 mg/kg in mice (Timosievskaya and Rokionova, 1973).

Administration of single oral (0.4 μmol/kg) or intraperitoneal (0.1 μmol/kg) doses to rats increased blood levels of methemoglobin (Senczuk et al., 1976; Watanabe et al., 1976). Intraperitoneal administration of isomers of dinitrobenzene had a similar effect.

Data regarding the carcinogenicity of 1,3,5-trinitrobenzene were limited to a single study employing dermal and intraperitoneal administrations. Single topical applications of 1,3,5-trinitrobenzene to the skin of mice elicited responses (inflammation, epidermal hyperplasia and cell darkening) similar to those caused by TPA, a demonstrated promoter of mouse skin tumors. Direct evidence for the carcinogenic potential of 1,3,5-trinitrobenzene to cause mouse skin tumors was unavailable (Slaga et al., 1985). Multiple intraperitoneal injections of 1,3,5-trinitrobenzene (3 times/week for 8 weeks) did not cause lung tumors in mice, but neither did benzo(a)-pyrene, a known carcinogen in the positive controls in this study (Slaga et al., 1985).

1,3,5-Trinitrobenzene was mutagenic in assays for reverse mutations in S. typhimurium strains, and the mutagenic activity was reduced (but not abolished) by the presence of a metabolic activating system (McGregor et al., 1980; Spanggord et al., 1982b; Kawai et al., 1987).

Data regarding teratogenic and other reproductive effects of 1,3,5-trinitrobenzene were not available.

As data regarding the carcinogenicity of 1,3,5-trinitrobenzene are insufficient to assess the carcenogenic potential in humans, 1,3,5-trinitrobenzene was assigned to EPA Group D - not classifiable as to human carcinogenicity. A subchronic oral RfD of 5×10^{-4} mg/kg/day and a chronic oral RfD of 5×10^{-5} mg/kg/day were derived for 1,3,5-trinitrobenzene, based on analogy to the RfD for 1,3-dinitrobenzene. An RQ of 100 for chronic toxicity was derived, based on analogy to 1,3-dinitrobenzene.

TABLE OF CONTENTS

TABLE OF CONTENTS (cont.)

TABLE OF CONTENTS (cont.)

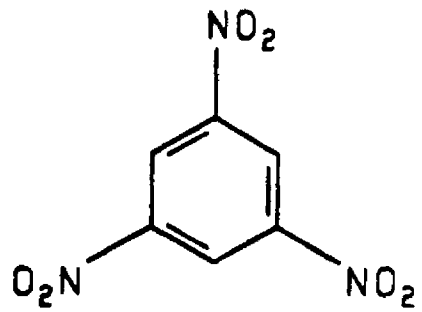
LIST OF ABBREVIATIONS

BCF	Bioconcentration factor
CAS	Chemical Abstract Service
CS	Composite score
DNA	Deoxyribonucleic acid
EC_{50}	Concentration effective to 50% of recipients (and all other subscripted concentration levels)
GMAV	Genus mean acute value
GMCV	Genus mean chronic value
K_{oc}	Soil sorption coefficient standardized with respect to organic carbon
K_{ow}	Octanol/water partition coefficient
LC_{50}	Concentration lethal to 50% of recipients (and all other subscripted dose levels)
LD_{50}	Dose lethal to 50% of recipients
MATC	Maximum acceptable toxicant concentration
MED	Minimum effective dose
ppm	Parts per million
RQ	Reportable quantity
RV_d	Dose-rating value
RV_e	Effect-rating value
SD	Standard deviation
TL_m	Median tolerance limit
TNT	Trinitrotoluene
TPA	7,12-dimethylbenz(a)anthracene(DMBA)-12-O-tetradecanoyl-phorbol-13-acetate
wt/wt	Weight per weight

1. INTRODUCTION

1.1. STRUCTURE AND CAS NUMBER

1,3,5-Trinitrobenzene is also known by the synonyms s-, sym-, symmetric and syn-trinitrobenzene and by the acronym TNB (CAS, 1989; Chemline, 1989; SANSS, 1989). The structure, CAS number, empirical formula and molecular weight are as follows:



CAS Registry number: 99-35-4

Empirical formula: $C_6H_3N_3O_6$

Molecular weight: 213.11

1.2. PHYSICAL AND CHEMICAL PROPERTIES

1,3,5-Trinitrobenzene is a yellow crystalline solid at room temperature (Sax and Lewis, 1987). It is a dimorphous solid; the most common form melts at 122.5°C and the rare form melts at 61°C (Weast et al., 1988; Windholz et al., 1983). It is soluble in polar organic solvents such as alcohol, acetone, ether and methanol and in nonpolar organic solvents such as benzene, carbon disulfide and petroleum ethers (Sax and Lewis, 1987; Weast et al., 1988; Windholz et al., 1983). It is also moderately soluble in water (Windholz et al., 1983). Selected physical properties for 1,3,5-trinitrobenzene are as follows:

Melting point:		
Common form	122.5°C	Windholz et al., 1983
Rare form	61°C	
Boiling point:	315°C	Weast et al., 1988

Density:	1.688 g/cm^3	Sax and Lewis, 1987
Water solubility:		
at 25°C	0.035 g/100 g	Windholz et al., 1983
at 20°C	0.034 g/100 g	Spanggord et al., 1980
Vapor pressure:		
at 20°C solid	3.2×10^{-6} mm Hg	Spanggord et al., 1980
supercooled liquid	2.2×10^{-4} mm Hg	Spanggord et al., 1980
Log K_{ow}:	1.18	Hansch and Leo, 1985
Conversion factors:	1 ppm = 8.66 mg/m^3 1 mg/m^3 = 0.115 ppm	

1.3. PRODUCTION DATA

Data from the U.S. EPA TSCA production file (TSCAPP, 1989) indicate that during 1977, only Eastman Kodak Co. in Rochester, NY, produced 1,3,5-trinitrobenzene. More current United States production data were not located in the available literature cited in Appendix A.

Data on the commercial methods used in the production of 1,3,5-trinitrobenzene were not located in the available literature. 1,3,5-Trinitrobenzene can be synthesized by treating benzene with a mixture of fuming nitric and fuming sulfuric acids (Purcell, 1981), by the action of alkali on 2,4,6-trinitrobenzaldehyde or the sequential oxidation/decarboxylation of 2,4,6-TNT (Windholz et al., 1983). 1,3,5-Trinitrobenzene is also produced as a by-product of the production of TNT (Spanggord et al., 1982a; Spalding and Fulton, 1988).

1.4. USE DATA

1,3,5-Trinitrobenzene is used in explosive compositions. It is less sensitive to impact than TNT but more powerful and brisant (Sax and Lewis, 1987; Windholz et al., 1983). It is also used as a vulcanizing agent for rubber (Barnhart, 1981) and as a pH indicator (Durst and Bates, 1981).

1.5. SUMMARY

1,3,5-Trinitrobenzene is a yellow crystalline solid at room temperature; it is soluble in both polar and nonpolar organic solvents and sparingly soluble in water (Sax and Lewis, 1987; Windholz et al., 1983). Information on current methods of production is lacking in the available literature. Only Eastman Kodak Co. in Rochester, NY, produced this material in 1977 (TSCAPP, 1989). 1,3,5-Trinitrobenzene is used mainly in explosive compositions (Sax and Lewis, 1987; Windholz et al., 1983). It is also used in the vulcanization of rubber (Barnhart, 1981) and as a pH indicator (Durst and Bates, 1931).

2. ENVIRONMENTAL FATE AND TRANSPORT

2.1. AIR

Given the available data on the vapor pressure of 1,3,5-trinitrobenzene at 20°C, 2.2×10^{-4} mm Hg (supercooled liquid) and 3.2×10^{-6} mm Hg (solid), it is expected that this compound will exist both in the vapor phase and in the particulate form in the ambient atmosphere (Eisenrich et al., 1981).

2.1.1. **Reaction with Hydroxyl Radicals.** Using the method of Atkinson (1985), a rate constant for the gas phase reaction of 1,3,5-trinitrobenzene with photochemically produced hydroxyl radicals in the atmosphere can be estimated to be 1.3×10^{-13} cm^3/molecule-sec. If an average atmospheric hydroxyl radical concentration is 5×10^5 molecules/cm^3, then the half-life for the vapor-phase destruction of 1,3,5-trinitrobenzene in the atmosphere would be 12,440 days. Consequently, this should not be an environmentally significant process.

2.1.2. **Reaction with Ozone.** 1,3,5-Trinitrobenzene is not expected to be susceptible to atmospheric degradation by ozone (Atkinson, 1985; U.S. EPA, 1987).

2.1.3. **Photolysis.** 1,3,5-Trinitrobenzene is known to absorb light in the environmentally significant range of >290 nm (Spanggord et al., 1980; Capellos and Suryanarayanan, 1973). Sufficient information does not exist in the available literature to accurately predict the photolytic fate of 1,3,5-trinitrobenzene in the atmosphere (Section 2.2.3.).

2.1.4. **Physical Removal Processes.** 1,3,5-Trinitrobenzene in the atmosphere is expected to exist partially in the particulate form and dry deposition may partially remove this compound from the atmosphere. The water solubility of 1,3,5-trinitrobenzene, 0.035 g/100 g at 25°C (Windholz et al., 1983), suggests that partial removal by wet deposition may also occur.

2.2. WATER

2.2.1. Hydrolysis. 1,3,5-Trinitrobenzene is not expected to hydrolyze under environmental conditions since it contains no hydrolyzable functional groups (Lyman et al., 1982).

2.2.2. Oxidation. Oxidation of 1,3,5-trinitrobenzene in water by electrophilic alkoxy or akylperoxy radicals is not expected to be an important fate process.

2.2.3. Photolysis. 1,3,5-Trinitrobenzene is known to adsorb light in the environmentally significant range >290 nm (Spanggord et al., 1980; Burlinson et al., 1973); however, the photolysis of aqueous 1,3,5-trinitrobenzene in the laboratory produced no reaction after 6 hours (Burlinson et al., 1973). Capellos and Suryanarayanan (1973) have shown that in the laboratory, photolytic decomposition of 1,3,5-trinitrobenzene only occurs in the presence of both oxygen and polar, nucleophilic reagents, such as methanol. Sunlight photolysis of 2,4,6-TNT in river water produced a 10% yield of 1,3,5-trinitrobenzene after 8 days (Spanggord et al., 1980). Thus, the rate of photolysis for 1,3,5-tri-nitrobenzene must be slower than for TNT, otherwise it would not have been detected in this experiment.

The above data suggest that when 1,3,5-trinitrobenzene is exposed to light, photochemical transformations unique to this compound may occur. Thus, the photolytic fate of 1,3,5-trinitrobenzene based on analogies to other nitro-aromatic compounds cannot be predicted. More information is necessary to accurately predict what process may occur when 1,3,5-trinitrobenzene in water is exposed to sunlight.

2.2.4. Microbial Degradation. Insufficient data in the available literature cited in Appendix A preclude the prediction of the microbial degradation of 1,3,5-trinitrobenzene in water. 1,3,5-Trinitrobenzene did not support growth of Nocardia V. cultures (Rodriguez-Villanueva, 1960).

Cell-free extracts of Veillonella alkalescens degraded 1,3,5-trinitrobenzene to the amino compound in the presence of hydrogen at a rate equal to 288 nmol H_2/min/mg protein, the most rapid rate obtained for the 40 nitrobenzene derivatives studied (McCormick et al., 1976). It is not possible to directly extrapolate from these laboratory studies to the behavior of 1,3,5-trinitrobenzene in the environment, but it is possible that anaerobic biodegradation may occur under the proper conditions.

2.2.5. **Bioconcentration.** The BCF for 1,3,5-tri-nitrobenzene can be calculated to range from 5-23 based on its water solubility, 0.035 mg/ℓ at 25°C (Windholz et al., 1983), and the log K_{ow}, 1.81 (Hansch and Leo, 1985). The respective regression equations, log BCF = 0.76 log K_{ow} - 0.23 and log BCF = 2.791 - 0.564 log s, were used in this estimation (Lyman et al., 1982). These values suggest that bioconcentration in fish and aquatic organisms is not an important fate process.

2.2.6. **Adsorption.** Using the regression equations log K_{oc} = 0.544 log K_{ow} + 1.377 and log K_{oc} = -0.55 log s + 3.64 (Lyman et al., 1982), the K_{oc} for 1,3,5-trinitrobenzene is calculated to range from 104-178 based on the water solubility, 340 mg/ℓ at 25°C (Windholz et al., 1983), and the log K_{ow}, 1.81 (Hansch and Leo, 1985), respectively. These values suggest that adsorption to sediment and suspended organic matter are not expected to be important fate processes.

2.2.7. **Volatilization.** Using the method of Hine and Mookerjee (1975), an estimated Henry's Law constant of 3.08×10^{-9} atm m³/mol at 25°C can be calculated for 1,3,5-trinitrobenzene. This value suggests that volatilization from water to the atmosphere is very slow. The volatilization half-life from a model river 1 m deep, flowing at 1 m/sec with a wind velocity of 3 m/sec is 47.5 years (Lyman et al., 1982).

2.3. SOIL

2.3.1. Microbial Degradation. Sufficient data are not available to predict the biodegradation of 1,3,5-trinitrobenzene in soil. Oxygen uptake that was not significantly higher than endogenous respiration was obtained when 1,3,5-trinitrobenzene was exposed to organisms obtained from soil, compost or a waste lagoon and enriched using phenol as the carbon source (Chambers and Kabler, 1964; Chambers et al., 1963; Tabak et al., 1964; Barth and Bunch, 1979). The results of these studies suggest that little or no aerobic biodegradation took place; however, anaerobic degradation may occur in soil under the proper conditions (see Section 2.2.4.).

2.3.2. Adsorption. K_{OC} values for 1,3,5-trinitrobenzene can be estimated to lie in the range from 104-178 (see Section 2.2.6.). These values suggest that 1,3,5-trinitrobenzene will display moderate to high mobility in soil (Swann et al., 1983).

2.3.3. Volatilization. The vapor pressure of 1,3,5-trinitrobenzene, 3.2×10^{-6} mm Hg at 20°C (Spanggord et al., 1980), suggests that volatilization from soil to the atmosphere is not a significant fate process.

2.4. SUMMARY

Sufficient data do not exist in the available literature to accurately predict the environmental fate of 1,3,5-trinitrobenzene but the following processes are expected to occur in the environment: In the atmosphere, 1,3,5-trinitrobenzene should exist partially in the vapor phase, and partially in the particulate form. Destruction by the gas-phase reaction with photochemically produced OH radical or by the reaction with ozone is not expected to be significant. Both wet and dry deposition may occur. In water, neither volatilization to the atmosphere nor hydrolysis are expected to be important processes. Information sufficient to predict the importance

of microbial degradation of 1,3,5-trinitrobenzene were not located in the available literature; however, rapid anaerobic degradation may occur under the proper conditions. 1,3,5-Trinitrobenzene adsorbs light in the environmentally significant range >290 nm (Burlinson et al., 1973; Spanggord et al., 1980; Capellos and Suryanarayanan, 1973) but the light-induced transformations of this compound are not well understood. Therefore, the photolytic destruction of 1,3,5-trinitrobenzene in the environment cannot be accurately predicted.

3. EXPOSURE

1,3,5-Trinitrobenzene is a man-made compound that is usually associated with the production of munitions and armaments. Limited data are available on the quantity of 1,3,5-trinitrobenzene in environmental media. Based on the available data, it appears that 1,3,5-trinitrobenzene can enter the environment in wastewater effluent from facilities that synthesize, produce or demilitarize munitions, or from the disposal of solid TNT wastes (Ryon et al., 1984; Spalding and Fulton, 1988; Spanggord et al., 1982a).

3.1. WATER

1,3,5-Trinitrobenzene was identified in the condensate wastewater effluent from the production of TNT. It was found in 3.8% of the samples taken weekly throughout 1 year in concentrations ranging from 0.06-0.20 ppm for the positive samples. 1,3,5-Trinitrobenzene was thought to be produced from the sequential oxidation and decarboxylation of TNT, which can occur at elevated temperatures during the distillation, condensation and subsequent release of the wastewater (Spanggord et al., 1982a). 1,3,5-Trinitrobenzene was qualitatively detected in groundwater samples contaminated with by-products from the manufacture of cyclonite and TNT near the Cornhusker Army Ammunition Plant, Nebraska (Spalding and Fulton, 1988). 1,3,5-Trinitrobenzene was identified as a by-product in wastewater effluent from the synthesis of p-nitrobenzoic acid, an intermediate used for the synthesis of pharmaceuticals, at a concentration of <1% (wt/wt) (Wennersten, 1980).

3.2. FOOD

Pertinent data regarding the detection of 1,3,5-trinitrobenzene in food were not located in the available literature cited in Appendix A.

3.3. INHALATION

Pertinent data regarding the exposure to 1,3,5-trinitrobenzene through inhalation were not located in the available literature cited in Appendix A.

3.4. DERMAL

Pertinent data regarding the dermal exposure to 1,3,5-trinitrobenzene were not located in the available literature cited in Appendix A.

3.5. SUMMARY

1,3,5-Trinitrobenzene is a man-made organic compound that can enter the environment as a component of wastewater effluent of plants that synthesize, produce or demilitarize explosives or munitions. 1,3,5-Trinitrobenzene may also enter the environment through the disposal of solid TNT wastes (Ryon et al., 1984, Spalding and Fulton, 1988; Spanggord et al., 1982a). Limited monitoring data are available on the concentration of this compound in the environment. Sufficient monitoring data are not available to estimate the exposure of 1,3,5-trinitirobenzene to the general population.

4. ENVIRONMENTAL TOXICOLOGY

4.1. AQUATIC TOXICOLOGY

4.1.1. Acute Toxic Effects on Fauna. The static acute toxicity of 1,3,5-trinitrobenzene to the fathead minnow, Pimephales promelas, was evaluated by Bailey and Spanggord (1983). Juvenile fish averaging 0.28 g were deprived of food for 24 hours before and during testing. Tests were conducted in 19 ℓ pickle jars containing 10 fish/jar and 15 ℓ of test solution. Dissolved oxygen, temperature, pH and mortality were checked daily. Hardness, alkalinity and conductivity of the diluent dechlorinated tap water were also monitored regularly (interval not reported). The 96-hour LC_{50} for fathead minnows was 1.1 mg/ℓ. Dissolved oxygen concentrations ranged from 2.8-9.4 mg/ℓ; pHs ranged from 6-9.2 mg/ℓ (mean 7.6); and mean temperatures ranged from 19.5-22°C (mean 20.3°C). Hardness ranged from 12-43 mg/ℓ as $CaCO_3$ and mean alkalinity ranged from 15-60 mg/ℓ as $CaCO_3$.

The acute static toxicity of trinitrobenzene to laboratory-reared fathead minnows, P. promelas, and water fleas, Daphnia magna, was determined by Pearson et al. (1979). Dechlorinated water was used in tests of both species. Tests were conducted at 20°C, pH of 7.2-8.6 and an average hardness and alkalinity of 26 mg/ℓ (as $CaCO_3$) and 45 mg/ℓ (as $CaCO_3$), respectively. The 96-hour LC_{50} for fathead minnows was 1.03 mg/ℓ. The 48-hour EC_{50} for D. magna was 2.7 mg/ℓ.

Liu et al. (1983) assessed the static acute toxicity of trinitrobenzene to fathead minnows, P. promelas, aged 90 (±2) days, and to water fleas, D. magna, first instars, obtained from laboratory-reared stocks and deprived of food during testing. Tests were conducted in dechlorinated tap water at 20°C having an average hardness of 33.8 mg/ℓ (SD=19.0) as calcium

carbonate ($CaCO_3$), pH of 7.7 (SD=0.35), alkalinity of 38.0 mg/ℓ (SD=20.0) as $CaCO_3$, and residual chlorine of 2.2 µg/ℓ (SD=0.95). Water temperatures were 20°C for minnows and 12°C for trout. The 96-hour LC_{50} estimate of static acute toxicity (and 95% confidence levels) for fathead minnows was 1.1 (1-1.2) mg/ℓ, and for water fleas, 2.7 (2.4-3.1) mg/ℓ.

Static acute toxicity of trinitrobenzene to the water flea, D. magna, bluegill, Lepomis macrochirus, rainbow trout, Salmo gairdneri, fathead minnow, P. promelas, and channel catfish, Ictalurus punctatus, was determined by van der Schalie (1983). Filtered, aerated and sterilized well water was used in the tests and monitored weekly for pH, hardness, alkalinity, total organic carbon, suspended solids and ammonia. Animals were acclimated to the well water for ≤30 weeks, transferred to holding tanks and held without food for 48 hours before starting the tests. The fish tests were conducted in 19 ℓ jars containing 14 ℓ of test solution. Three jars containing 10 fish, or two jars of 15 smaller fish, were used for each treatment level.

The jars were held in a tank at 22 (±2)°C for the warmwater fish species, and 12(±2)°C for the trout. The 96-hour LC_{50} values in mg/ℓ, (and 95% confidence limits) were reported as follows: bluegills, 0.85 (0.52-1.38); rainbow trout, 0.52 (0.32-0.8); fathead minnows, 0.49 (0.44-0.56); and channel catfish, 0.38 (0.34-0.43).

van der Schalie (1983) conducted a static acute test of trinitrobenzene in the water flea, D. magna, identically to that described above for fish, except that daphnids to be tested were obtained from females isolated from stock cultures <24 hours before starting the test. Trout chow and yeast food were provided up to the time that the young were pooled for testing.

Five neonates were transferred by eyedropper into six 200 mℓ beakers/ treatment level. The 48-hour EC_{50} level (and 95% confidence limit) was 2.98 (2.63-3.38) mg/ℓ. A higher 48-hour EC_{50} value of 4.1 (2.6-7.7) mg/ℓ was determined for this species when fed vitamin-enriched algae, Ankistrocesmus falcatus, twice daily (Section 4.1.2.1.).

van der Schalie et al. (1988) determined the acute flowthrough toxicity of trinitrobenzene to bluegills, L. macrochirus. Filtered and sterilized well water was used in the tests and was maintained at 22(±2)°C; dissolved oxygen concentration ranged from 88-98% saturation, pH was 8.2-8.3, alkalinity (as $CaCO_3$) averaged 241 (range 227-250) mg/ℓ, and hardness (as $CaCC_3$) averaged 173 (range 170-174) mg/ℓ. Ten fish were randomly assigned to two test aquaria containing 7.6 ℓ of test solution. Six treatment levels (0.10, 0.14, 0.34, 0.69, 1.29 and 3.07 mg/ℓ, mean measured concentration) were tested. Fish were not fed for 48 hours before starting the tests. Approximately five tank volumes were delivered by a proportional diluter to each tank per day. The 96-hour LC_{50} was 0.57 (95% fiducial limits of 0.50-0.65 mg/ℓ).

4.1.2. Chronic Effects on Fauna.

4.1.2.1. **TOXICITY** -- Early life stage tests of trinitrobenzene's toxicity were conducted on fathead minnows, P. promelas, and rainbow trout, S. gairdneri, by van der Schalie (1983). The water source and treatment were handled similarly to the method described in Section 4.1.1. Fathead minnow eggs were obtained from adults that had been held in aquaria containing spawning substrates. Larvae <24 hours old were used to start the test. Approximately 450 eggs were pooled, then randomized into beakers containing well water and egg cups. Each cup contained 35 eggs, with two replicate cups for each of the five treatment levels (0.72, 0.32, 0.18, 0.12 and 0.08

mg/ℓ) and controls (0.05 mg/ℓ). These cups were randomly placed in the water bath, and a rocker-arm apparatus kept the eggs in motion. Temperature was maintained at 25°C (range 23.8-26.3°C), dissolved oxygen levels were between 7.9 and 8.9 mg/ℓ and pH ranged from 7.9-8.2. When ≥90% hatch had occurred, fry were released into the test tank and fed twice/day. The test duration was 32 days. No effects were noted on hatching rate, hatching success or morphology of offspring at any treatment level. Significantly reduced survival was noted for all but the lowest dose. Fry at all treatment levels were less active than the controls. The LOAEL, based on mortality, is 0.12 ppm for 1,3,5-trinitrobenzene. Effects noted on fry length and weight do not appear to be treatment-related.

van der Schalie (1983) obtained eyed eggs of rainbow trout, S. gairdneri, from a national hatchery for early life stage tests. Eggs were placed in groups of 60 into egg baskets suspended in two 19ℓ aquaria containing 15 ℓ of test solution/treatment level. Treatment levels ranged from 0.09-0.71 mg/ℓ, but because significant effects were noted at the lowest concentration, a second test was conducted. Treatment levels for the second test were 0.17, 0.082, 0.045, 0.022, 0.015 and 0.01 mg/ℓ trinitrobenzene. Data discussed here are taken from the second test. Temperatures were maintained within 1° of 12°C, dissolved oxygen concentrations averaged 8.5-9.0 mg/ℓ and pH averaged between 8.0 and 8.2 (range 7.9-8.5). Feeding (twice daily) was initiated when fry swam out of the egg basket. Total duration of the rainbow trout test was 71 days (61 days following the 50% hatch day for controls). At concentrations of 0.17 and 0.082 mg/ℓ, fish showed erratic swimming patterns and were lighter in color than the controls. Significant differences in survival, fry length and fry weight were noted at 0.17 ppm, a LOAEL. No significant effects were reported on

hatching success, time to hatch, time to swim-up or fry morphology at treatment levels $\leq$0.71 mg/ℓ. A NOEL of 0.045 ppm and a NOAEL of 0.082 are thus identified for these study conditions.

Long-term effects on these same fish species were noted in flowthrough acute toxicity tests conducted by van der Schalie (1983). The same test conditions were used here as described in the static acute test (see Section 4.1.1.), except that fish were tested in 19 ℓ aquaria containing 15 ℓ of test solution. These aquaria measured 40 x 20 x 25 cm and had a drain hole at a height of 19 cm. For five treatment levels and a control, two replicate tanks housing 15 fish/tank were used. (In the rainbow trout tests, two replicates of 10 fish each were used.) For fathead minnows, a 10-day LC_{50} of 0.46 mg/ℓ was obtained (95% confidence limits = 0.42-0.53 ppm). The rainbow trout dynamic acute test results were an 18-day LC_{50} (and 95% confidence limits) of 0.4 mg/ℓ (0.24-0.73) and a 10-day LC_{50} (and 95% confidence limits) of 0.52 (0.37-0.73) mg/ℓ.

van der Schalie et al. (1988) determined the effects on ventilatory patterns and whole-body movement rates caused by chronic exposure of bluegills, L. macrochirus, to trinitrobenzene. Five fish/group, except for the highest treatment level in which three/group were tested, were exposed to six treatment levels (at mean measured trinitrobenzene concentrations of 0.613, 0 279, 0.128, 0.061, 0.034 and 0.02 mg/ℓ or below detection limit) for 6 days. Parameters monitored were ventilatory rate, ventilatory depth, cough rate and percent movement. The monitoring apparatus was that described by van der Schalie (1980) but was modified from an anterior/posterior electrode arrangement for monitoring fish ventilatory signals to a dorsal/ventral pattern. A proportional toxicant diluter delivered test solutions to the ventilatory chambers. Ventilatory parameters were

monitored continuously by microcomputer and compiled every 15 minutes. No significant differences in ventilatory signal minima were noted among control and treated fish at any exposure level. Effects were noted on ventilatory maxima for all parameters except ventilatory rate. Ventilatory depth was the most sensitive parameter tested, showing significant responses at concentrations $\geq$0.128 mg/ℓ. Significant effects on cough rate and percent movement were noted at 0.613 mg/ℓ. A concentration of 0.128 ppm thus represents a LOAEL and 0.061 ppm a NOEL.

Hiatt et al (1957) exposed marine fish, Kuhlia sandvicensis, to 1.3.5-trinitrobenzene concentrations of 10, 50, 100, 1000 and 10,000 µg/ℓ (0.01, 0.05, 0.1, 1 and 10 ppm, respectively) for short durations (time period not specified) and noted slight irritant effects (excitability, violent swimming and opercular movement increases), suggesting respiratory distress at the 0.1 ppm exposure level. These reactions were violent at the higher exposure levels and were not noted with exposures of $\leq$0.05 ppm. Lack of information on duration of this study precludes identification of effect levels for risk assessment purposes.

Chronic data were generated by van der Schalie (1983) for D. magna exposed to trinitrobenzene. Animals used in these tests were raised in an in-house culture unit in 2 ℓ tanks with 10 daphnids/tank. Aerated well water was maintained at 20°C (range 19-21 days), and light intensity was 150-350 lux. Daphnids were fed twice daily with ≈2 mg/ℓ (dry weight) vitamin-enriched alga, Ankistrodesmus falcatus. Daphnids were exposed to trinitrobenzene for 21 days. Ten daphnids were placed in four replicate tanks for each treatment level of 2.68, 1.32, 0.75, 0.47, 0.24 and 0.025 (control). Endpoints monitored were immobilization, young/replicate tank, young/female/reproductive day (total young divided by total days alive after

onset of reproduction in the test tank) and growth. Significant effects were noted at the three highest concentrations (mean total young/tank and total lergth). At concentrations ≥1.32 ppm, number of young/female/reproductive day differed significantly from that of controls. Daphnids subjected to 2.68 ppm exhibited a marked spinning motion when swimming. No significant differences were noted for any treatment level on survival. A concentration of 0.47 ppm represents a chronic NOEL and 0.75 a chronic LOEL.

4.1.2.2. **BIOACCUMULATION/BIOCONCENTRATION** -- Liu et al. (1983) calculated a steady-state BCF for trinitrobenzene of 6.36 from the estimated log K_{ow} value of 1.36. This value suggests that trinitrobenzene will not significantly bioaccumulate in aquatic organisms.

4.1.3. Effects on Flora.

4.1.3.1. **TOXICITY** -- van der Schalie et al. (1983) tested the toxicity of trinitrobenzene in the alga, Selenastrum capricornutum. Algae were exposed to solutions of trinitrobenzene in 100 mℓ of algal assay medium contained in 500 mℓ Erlenmeyer flasks. Triplicate flasks for each treatment level were inoculated with 20,000 cells of S. capricornutum/mℓ/flask and reared under 4300 lux of cool white fluorescent light for 14 days at 26°C (range 25-27°C). Cell counts were made using an electronic particle counter. Growth, measured in cells/mℓ, was monitored. Significant reduction in growth compared with that of controls was noted at all treatment levels on days 5 and 14 of the test. Concentrations >1.18 ppm were algicidal, and concentrations <1.18 ppm were algistatic.

4.1.3.2. **BIOCONCENTRATION** -- Pertinent data regarding the bioaccumulation/bioconcentration potential of trinitrobenzene in aquatic flora were not located in the available literature cited in Appendix A.

4.1.4. Effects on Bacteria. Pertinent data regarding the effects of exposure of aquatic bacteria to trinitrobenzene were not located in the available literature cited in Appendix A.

4.2. TERRESTRIAL TOXICOLOGY

4.2.1. Effects on Fauna. Pertinent data regarding the effects of exposure of terrestrial fauna to trinitrobenzene were not located in the available literature cited in Appendix A.

4.2.2. Effects on Flora. Pertinent data regarding the effects of exposure of terrestrial flora to trinitrobenzene were not located in the available literature cited in Appendix A.

4.3. FIELD STUDIES

Pertinent data regarding the effects of trinitrobenzene on flora and fauna in the field were not located in the available literature cited in Appendix A.

4.4. AQUATIC RISK ASSESSMENT

The lack of pertinent data regarding the effects of exposure of aquatic fauna and flora to trinitrobenzene precluded the development of a freshwater criterion by the method of U.S. EPA/OWRS (1986) (Figure 4-1). Additional data required for the development of a freshwater criterion include the results of acute assays with benthic crustaceans, an insect, a nonarthropod and nonchordate species and an insect or species from a phylum not previously represented. The development of a freshwater criterion also requires data from acceptable chronic toxicity tests with two species of fauna and at least one bioconcentration study.

The lack of pertinent data regarding the effects of exposure of aquatic fauna and flora to trinitrobenzene also precluded the development of a saltwater criterion by the method of U.S. EPA/OWRS (1986). Additional data

Family	TEST TYPE		
	GMAV* (ppm)	GMCV* (ppm)	BCF*
#1 Chordate (Salmonid-fish)	0.52*	NA	NA
#2 Chordate (warmwater fish)	0.0.884*	NA	NA
#3 Chordate (fish or amphibian)	0.57*	NA	NA
#4 Crustacean (planktonic)	3.072*	0.47[i]	NA
#5 Crustacean (benthic)	NA	NA	NA
#6 Insectan	NA	NA	NA
#7 non-Arthropod/-Chordate	NA	NA	NA
#8 New insectan or phylum representative	NA	NA	NA
#9 algae	XXXXXXXXXXX XXXXXXXXXXX	0.10[j]	NA
#10 Vascular plant	XXXXXXXXXXX XXXXXXXXXXX	NA	NA

*NA=Not available;*96-hr LC_{50} for rainbow trout, Salmo gairdneri; *Mean 96-hr LC_{50} for fathead minnows, (Pimephales promelas); *96-hr LC_{50} for bluegill sunfish, Lepomis macrochirus; *Mean 48-hr EC_{50} for the water flea, Daphnia magna; **1-day NOEC for the water flea, D. magna; *14-day LOEC for the alga, Selenastrum capricornutum.

FIGURE 4-1

Organization Chart for Listing GMAVs, GMCVs and BCFs Required to Derive Numerical Water Quality Criteria by the Method of U.S. EPA/OWRS (1986) for the Protection of Freshwater Aquatic Life from Exposure to Trinitrobenzene

required for the development of a saltwater criterion include the results of acute assays with two chordate species, a nonarthropod and nonchordate species, a mysid or panaeid crustacean, two additional nonchordate species and one other species of marine fauna. The development of a saltwater criterior also requires data from chronic toxicity tests with two species of fauna anc one species of algae or vascular plant and at least one bioconcentration study.

4.5. SUMMARY

Existing data indicate that trinitrobenzene is highly toxic to aquatic fauna but that the compound is not likely to concentrate in them. Acute toxicity data have been reported for four species of fish (fathead minnows, channel catfish, bluegills and rainbow trout) and one invertebrate, the water flea. LC_{50} values for fathead minnows range from 0.49-1.1 ppm, and the othe· three species displayed sensitivity to trinitrobenzene at LC_{50} levels <·.0 ppm and ranging from 0.38 to 0.8-5 (Bailey and Spanggord, 1983; Liu et al., 1983; Pearson et al., 1979; van der Schalie, 1983; van der Schalie et al., 1988). The water flea, D. magna, was less sensitive, showing LC_{50} values ranging from 2.7-4.1 ppm trinitrobenzene (Liu et al., 1983; Pearson et al., 1979; van der Schalie, 1983).

Chronic toxicity data are provided for fathead minnows, rainbow trout, bluegills and the water flea. Early life stage tests indicate that fathead minnows are highly sensitive to trinitrobenzene, with survival significantly reduced at exposures $\geq$0.12 ppm for 32 days. Rainbow trout were similarly sensitive to trinitrobenzene, showing a LOEL of 0.17 ppm for survival, fry length and fry weight (van der Schalie, 1983). Ventilatory effects were noted in bluegills exposed to treatment levels $\geq$0.128 ppm for 6 days (van der Schalie et al., 1988). Signs of respiratory distress (opercular

movement increases, excitability and violent swimming) were noted in K. sandvicensus exposed to ≥0.1 ppm trinitrobenzene for short durations (Hiatt et al., 1957).

A BCF of 6.36 has been calculated for trinitrobenzene by Liu et al. (1983) from the estimated log K_{ow} value of 1.36. Bioconcentration data reported by van der Schalie (1983) for fathead minnows, rainbow trout and the water flea support the conclusion that trinitrobenzene does not significantly bioaccumulate in aquatic animals.

Toxic effects of trinitrobenzene in the alga, S. capricornutum, was investigated by van der Schalie (1983). Significant reduction in growth was noted at all levels tested (0.01-17.32 ppm) after 5 and 14 days exposure; concentrations of 1.18 ppm were algicidal, and lower concentrations were algistatic.

5. PHARMACOKINETICS

5.1. ABSORPTION

Pertinent data regarding the absorption of 1,3,5-trinitrobenzene were not located in the available literature cited in Appendix A.

5.2. DISTRIBUTION

Pertinent data regarding the distribution of 1,3,5-trinitrobenzene were not located in the available literature cited in Appendix A.

5.3. METABOLISM

Pertinent data regarding the metabolism of 1,3,5-trinitrobenzene were not located in the available literature cited in Appendix A.

5.4. EXCRETION

Pertinent data regarding the excretion of 1,3,5-trinitrobenzene were not located in the available literature cited in Appendix A.

6. EFFECTS

6.1. SYSTEMIC TOXICITY

6.1.1. Inhalation Exposure. Data regarding the subchronic and chronic toxicity of 1,3,5-trinitrobenzene following inhalation exposure were not located in the available literature cited in Appendix A.

6.1.2. Oral Exposure. Data regarding the subchronic and chronic toxicity of 1,3,5-trinitrobenzene following oral exposure were not located in the available literature cited in Appendix A.

6.1.3. Other Relevant Information. Korolev et al. (1977) reported oral LD_{50} values of 600 mg/kg in white mice, 450 mg/kg in white rats and 730 mg/kg in guinea pigs. Toxicity was characterized by central nervous system and respiratory disorders and cyanosis. Ten rats were given daily oral doses of 90 mg/kg (in an unspecified vehicle) for 30 days, and two died. In a longer-term study (in the English abstract, the experimental period was referred to only as "prolonged"), orally administered 1,3,5-trinitrobenzene (0.02-2 mg/kg) altered peroxidase, alkaline phosphatase and ceroplasmin activities in the blood. A 5 mg/kg dose reportedly had weakly allergenic properties. Further details were unavailable.

Timofievskaya and Rodionova (1973) cited oral LD_{50} values of 572 mg/kg for 1,3,5-trinitrobenzene in mice. Exposure to air saturated with trinitrobenzene for 24 hours was not toxic to mice, but details regarding measured endpoints were unavailable. Timoievskaya and Rodionova (1973) also reported that application of trinitrobenzene to the shaved skin of mice caused hyperemia, edema and hemorrhages and that instillation of 50 mg of trinitrobenzene into the eyes of rabbits caused irritation. Further details, however, were unavailable.

Watarabe et al. (1976) compared the ability of various nitrobenzenes to cause methemoglobin formation in the blood of rats. Groups of five male Wistar rats were given single intraperitoneal doses of 100 μmols/kg of the compounds dissolved in polypropylene glycol. Five hours after injection, blood was examined for methemoglobin level. 1,3-Dinitrobenzene and 1,4-dinitrobenzene caused greater formation of methemoglobin than 1,3,5-trinitrobenzene, but the difference was statistically significant only for 1,4-nitrobenzene. Data for control animals were not reported. In an in vitro assay, 1,3,5-trinitrobenzene, in addition to the three isomers of dinitrobenzene, caused greater formation of methemoglobin than controls.

Senczuk et al. (1976) reported that single oral doses of trinitrobenzene (0.4 μmols/kg) also caused methemoglobin formation in Wistar rats. Further details were unavailable.

6.2. CARCINOGENICITY

6.2.1. Inhalation. Pertinent data regarding the carcinogenicity of inhaled 1,3,5-trinitrobenzene were not located in the available literature cited in Appendix A.

6.2.2. Oral. Pertinent data regarding the carcinogenicity of ingested 1,3,5-trinitrobenzene were not located in the available literature cited in Appendix A.

6.2.3. Other Relevant Information. Single applications of 10 or 50 mg 1,3,5-trinitrobenzene (dissolved in acetone) to the skin of mice increased the incidence of inflammation, epidermal hyperplasia and dark cells (Slaga et al., 1985). The response elicited by these dose levels was similar to the maximum response obtained with TPA, a potent promoter of two-stage carcinogenic tumors in the skin of SENCAR mice. 1,3,5-Trinitrobenzene

tested negatively in assays for initiation of TPA-promoted skin carcinogenicity. Postinitiation assays for the promotion of skin cancers were not conducted with 1,3,5-trinitrobenzene.

Intraperitoneal administration of 1,3,5-trinitrobenzene dissolved in corn oil (0, 600, 1500 and 3000 mg/kg 3 times weekly for 8 consecutive weeks) did not cause lung tumors in male A/Jax mice (Slaga et al. 1985). The lungs of five mice/dose were examined 16 weeks after the last injection. These results provided equivocal data regarding the carcinogenic potential of 1,3,5-trinitrobenzene because demonstrated carcinogens [benzo(a)pyrene and 4-nitroquinoline-N-oxide] did not produce lung tumors in the assay.

6.3. MUTAGENICITY

1,3,5-Trinitrobenzene produced reverse mutations in Salmonella typhimurium strains in three separate studies (McGregor et al., 1980; Spanggord et al., 1982b; Kawai et al., 1987) (Table 6-1). In each study, the presence of the S-9 activating system reduced, but did not abolish, the mutagenic activity of 1,3,5-trinitrobenzene. Evidence for 1,3,5-trinitrobenzene-induced genetic toxicity was not found in DNA repair assays with Escherichia coli strains or in mitotic recombination assays with Saccharomyces cerevisiae D5 (McGregor et al., 1980).

6.4. TERATOGENICITY

Pertinent data regarding the teratogenicity of 1,3,5-trinitrobenzene were not located in the available literature cited in Appendix A.

6.5. OTHER REPRODUCTIVE EFFECTS

Pertinent data regarding other reproductive effects were not located in the available literature cited in Appendix A.

6.6. SUMMARY

Information regarding the chronic or subchronic toxicity of 1,3,5-trinitrobenzene following inhalation exposure is unavailable. Information

TABLE 6-1

Mutagenicity Data for 1,3,5-Trinitrobenzene

Assay	Indicator/ Organism	Compound and/or Purity	Application	Concentration or Dose	Activating System	Response	Comment	Reference
Reverse mutation	Salmonella typhimurium TA1535, TA1537, TA1538, TA98, TA100	98.8%	plate incorporation	0-100 µg/g/plate	±S-9	+ +	Presence of S-9 reduced mutagenic activity	McGregor et al., 1980
Reverse mutation	S. typhimurium TA1535, TA1537, TA1538, TA98, TA100	NR	plate incorporation	10-70 µg/g plate	±S-9	+ +	4/5 of strains were positive without S-9; 3/5 of strains were positive with S-9	Spanggord et al., 1982b
Reverse mutation	S. typhimurium TA100, TA98	NR	plate incorporation	0, 100 or 250 µg/g plate	±S-9	+ +	Presence of S-9 reduced mutagenic activity	Kawai et al., 1987
DNA repair	Escherichia coli W3100/pol A+ p 3478/pol A-	98.8%	plate incorporation	0.1-10 mg/plate	±S-9	- -	Precipitates formed at ≥1 mg; compound toxic at all levels tested; no differential toxicity between pol A+ and pol A- strains	McGregor et al., 1980
Mitotic recombination	Saccharomyces cerevisiae D5	98.8%	incubation medium	2 mg/mt	±S-9	- -	Precipate occurred at this level; no toxicity in absence of S-9; toxicity in presence of S-9	McGregor et al., 1980

NR = Not reported

regarding oral exposure of chronic or subchronic duration is limited to an abstract of a Russian report (Korolev et al., 1977). "Prolonged" oral administration of 1,3,5-trinitrobenzene to mice, rats and guinea pigs altered the activities of peroxidase, alkaline phosphatase and ceroplasmin in the blood, but further details were unavailable.

Published oral LD_{50} values for 1,3,5-trinitrobenzene include: 600 mg/kg in white mice, 450 mg/kg in white rats, 730 mg/kg in guinea pigs (Korolev et al., 1977) and 600 mg/kg in mice (Timosievskaya and Rokionova, 1973).

Administration of single oral (0.4 μmol/kg) or intraperitoneal (0.1 μmol/kg) doses to rats increased blood levels of methemoglobin (Senczuk et al., 1975; Watanabe et al., 1976). Intraperitoneal administration of isomers of dinitrobenzene had a similar effect.

Data regarding the carcinogenicity of 1,3,5-trinitrobenzene were limited to a single study employing dermal and intraperitoneal administrations. Single topical applications of 1,3,5-trinitrobenzene to the skin of mice elicited a response (inflammation, epidermal hyperplasia and cell darkening) similar to that caused by TPA, a demonstrated promoter of mouse skin tumors. Direct evidence for the carcinogenic potential of 1,3,5-trinitrobenzene to cause mouse skin tumors was unavailable. Multiple intraperitoneal injections of 1,3,5-trinitrobenzene (3 times/week for 8 weeks) did not cause lung tumors in mice, but neither did benzo(a)pyrene, a known carcinogen (Slaga et al., 1985).

1,3,5-Trinitrobenzene was mutagenic in assays for reverse mutations in S. typhimurium strains, and the mutagenic activity was reduced, but not abolished, by the presence of a metabolic activating system (McGregor et al., 1980; Spanggord et al., 1982b; Kawai et al., 1987).

Data regarding teratogenic and other reproductive effects of 1,3,5-trinitrobenzene were not available.

7. EXISTING GUIDELINES AND STANDARDS

7.1. HUMAN

A chronic oral RfD for 1,3,5-trinitrobenzene of 0.05 µg/kg/day was adopted by the U.S. EPA (1988). This value was based on the RfD for 1,3-dinitrobenzene because of the structural similarity between the two molecules and the insufficient data on the toxicity of 1,3,5-trinitrobenzene.

7.2. AQUATIC

Pertinent data regarding additional guidelines and standards for 1,3,5-trinitrobenzene were not located in the available literature cited in Appendix A.

8. RISK ASSESSMENT

8.1. CARCINOGENICITY

8.1.1. Inhalation. Pertinent data regarding the carcinogenicity of 1,3,5-trinitrobenzene to animals or humans by inhalation exposure were not located in the available literature cited in Appendix A.

8.1.2. Oral. Pertinent data regarding the carcinogenicity of 1,3,5-trinitrobenzene to animals or humans by oral exposure were not located in the available literature cited in Appendix A.

8.1.3. Other Routes. Single topical applications of 1,3,5-trinitrobenzene (10 and 50 mg) to the skin of mice caused inflammation, epidermal hyperplasia and cell darkening. This response was similar to the maximum response caused by TPA, a demonstrated promoter of mouse skin tumors. 1,3,5-Trinitrobenzene did not initiate TPA-promoted mouse skin tumors, but tests of the ability of 1,3,5-trinitrobenzene to promote skin tumors in the presence of a known initiator were not conducted. Multiple intraperitoneal injections of 1,3,5-trinitrobenzene (600, 1500 or 3000 mg/kg, 3 times/week for 8 weeks) did not cause lung tumors in mice (Slaga et al., 1985).

8.1.4. Weight of Evidence. The available data regarding the carcinogenicity of 1,3,5-trinitrobenzene are insufficient to adequately assess the carcinogenic potential of 1,3,5-trinitrobenzene in humans. The negative results for 1,3,5-trinitrobenzene in the only available study (Slaga et al., 1985) are inconclusive because 1) direct tests of the ability of 1,3,5-trinitrobenzene to promote mouse skin tumors in the presence of a known initiator were not conducted, and 2) intraperitoneal administration of benzo(a)pyrene, a known carcinogen included as a positive control, did not produce mouse lung tumors. Applying guidelines for carcinogenic risk assessment adopted by the U.S. EPA (1986b), 1,3,5-trinitrobenzene is assigned to EPA Group D - not classifiable as to human carcinogenicity.

8.1.5. Quantitative Risk Assessment.

8.1.5.1. INHALATION -- Data regarding the carcinogenicity of 1,3,5-trinitrobenzene by inhalation exposure are not available; therefore, estimates of carcinogenic potency cannot be derived.

8.1.5.2. ORAL -- Data regarding the carcinogenicity of 1,3,5-trinitrobenzene by oral exposure are not available; therefore, estimates of carcinogenic potency cannot be derived.

8.2. SYSTEMIC TOXICITY

8.2.1. Inhalation Exposure.

8.2.1.1. LESS THAN LIFETIME EXPOSURE (SUBCHRONIC) -- Data regarding subchronic inhalation toxicity of 1,3,5-trinitrobenzene are not available; therefore, an RfD for subchronic inhalation exposure cannot be derived.

8.2.1.2. CHRONIC EXPOSURE -- Data regarding chronic inhalation toxicity of 1,3,5-trinitrobenzene are not available; therefore, an RfD for chronic inhalation exposure cannot be derived.

8.2.2. Oral Exposure.

8.2.2.1. LESS THAN LIFETIME EXPOSURE (SUBCHRONIC) -- Data regarding subchronic or chronic oral toxicity of 1,3,5-trinitrobenzene are insufficient for derivation of an oral RfD. The U.S. EPA (1988), however, has adopted a chronic oral RfD for 1,3,5-trinitrobenzene derived from the RfD for the structurally similar 1,3-dinitrobenzene. Justification for this derivation included the fact that the LD_{50} value for 1,3-dinitrobenzene (83 mg/kg) in rats (Cody et al., 1981) is much lower than the LD_{50} value for 1,3,5-trinitrobenzene (450 mg/kg) in rats (Korolev et al., 1977). Additional justification is provided by the observation that administration of either 1,3-dinitrobenzene or 1,3,5-trinitrobenzene to rats caused similar increases in blood levels of methemoglobin (Watanabe et al., 1976).

The derivation of the oral RfD for 1,3-dinitrobenzene is based on the results of rat studies by Cody et al. (1981). Carworth Farm rats (20/sex/dose) were exposed to 0, 3, 8 or 20 ppm 1,3-dinitrobenzene in drinking water for 16 weeks. The highest concentration caused decreased body weight in females, decreased hemoglobin concentrations, testicular atrophy in males, splenic enlargement and splenic fibrosis with hemosiderin deposits in both sexes. In rats of both sexes treated with 8 ppm, increased spleen weights were also observed. No treatment-related effects were noted at the lowest dose in this study, but in a separate behavioral study of male rats given 3 and 8 ppm 1,3-dinitrobenzene in drinking water for 90 days, treated rats showed significantly increased activity on a running wheel (Cody et al., 1981). The concentration of 3 ppm is thus a NOAEL, and 8 ppm a LOAEL. Based on water consumption and body weight data, Cody et al. (1981) calculated that the 3-ppm level corresponded to an average dose of 0.40 mg/kg/day in male rats.

Adjusting for molecular weight differences, the NOAEL of 0.4 mg/kg/day for the subchronic toxicity of 1,3-dinitrobenzene corresponds to a 1,3,5-trinitrobenzene dose of 0.51 mg/kg/day. A subchronic oral RfD of 0.5 µg/kg/day for 1,3,5-trinitrobenzene is derived by dividing by an uncertainty factor of 1000 (10 for derivation of an RfD by structural analogy to reflect the lack of confidence in the data base, 10 for interspecies extrapolation and 10 for sensitive members of the human population). Confidence in the RfD is low as discussed in Section 8.2.2.2.

8.2.2.2. CHRONIC EXPOSURE -- A chronic oral RfD of 0.05 µg/kg/day is derived by dividing the subchronic oral RfD of 5×10^{-4} mg/kg/day by an additional factor of 10 to extrapolate from subchronic to chronic exposure.

Confidence in the key study for the RfD derivation is medium because both a NOAEL and LOAEL were identified; adequate numbers of animals were tested; and multiple endpoints were examined. However, the lack of toxicological and pharmacokinetic data on 1,3,5-trinitrobenzene makes confidence in the data base and RfD very low.

9. REPORTABLE QUANTITIES

9.1. BASED ON SYSTEMIC TOXICITY

As discussed in Chapter 6, data regarding the subchronic and chronic toxicity of 1,3,5-trinitrobenzene are not available. Oral RfDs were derived by analogy from the RfD for the structurally similar 1,3-dinitrobenzene. Following the same logic, a provisional RQ for 1,3,5-trinitrobenzene can be derived from subchronic toxicity data for 1,3-dinitrobenzene from the rat study by Cody et al. (1981). Increased spleen weight and increased running wheel activities were noted in rats provided with drinking water containing 1,3-dinitrobenzene at concentrations $\geq$8 ppm. Based on water consumption and body weight data, this concentration corresponded to an average intake of 1.13 mg/kg/day. An equivalent daily intake of 1,3,5-trinitrobenzene is 1.44 mg/kg/day, from which a human equivalent dose of 0.25 mg/kg/day is derived by multiplying by the cube root of the ratio of the reference body weight (0.35 kg) for rats (U.S. EPA, 1980a) to the reference human body weight (70 kg).

A chronic human MED of 1.7 mg/day is derived by multiplying the human equivalent dose of 0.25 mg/kg/day by 70 kg to express the dose in mg/day for a 70 kg human and dividing by 10 to approximate chronic from subchronic exposure. The MED of 1.7 mg/day corresponds to an RV_d of 5.15. An RV_e of 4 is assigned to the noted effects (increased running wheel activity and spleen weight); when it is multiplied by the RV_d of 5.15, a CS of 20.58 is derived. The CS of 20.58 corresponds to an RQ for 1,3,5-trinitrobenzene of 100 (Table 9-1). This RQ, however, is based on analogy to 1,3-dinitrobenzene and should therefore be considered provisional.

TABLE 9-1

1,3,5-Trinitrobenzene

Minimum Effective Dose (MED) and Reportable Quantity

Route:	oral
Species:	rats
Dose*:	1.7 mg/day
Duration:	16 weeks
Effect:	increased spleen weight
RV_d:	5.15
RV_e:	4
CS:	20.58
RQ:	100
Reference:	Cody et al., 1981

*Equivalent human dose, based on analogy to 1,3-dinitrobenzene

10. REFERENCES

Atkinson, R. 1985. Kinetics and mechanisms of the gas-phase reactions of the hydroxyl radical with organic compounds under atmospheric conditions. Chem. Rev. 85: 69-75, 185-193.

Bailey, H.C. and R.J. Spanggord. 1983. The relationship between the toxicity and structure of nitroaromatic chemicals. In: Proc. Aquatic Toxicology and Hazard Assessment, 6th Symposium, W.E. Bishop, R.D. Cardwell and B.B. Heidolph, Ed. American Society for Testing and Materials (ASTM) STP, Philadelphia, PA. 802: 98-107.

Barnhart, R.R. 1981. Rubber compounding. In: Kirk-Othmer Encyclopedia of Chemical Technology, 3rd ed., A. Stannden, Ed. John Wiley and Sons, New York, NY. 20: 393, 468.

Barth, E.F. and R.L. Bunch. 1979. Biodegradation and treatability of specific pollutants. EPA-600/9-79-034. U.S. EPA Municipal Environmental Research Lab, Cincinnati, OH. p. 1-60.

Burlinson, N.E., L.A. Kaplan and C.E. Adams. 1973. Photochemistry of TNT: Investigation of the pink water problem. Naval Ordinance Laboratory, NOCTR73-172. p. 1-14.

Capellos, C. and K. Suryanarayanan. 1973. Flash photolysis of s-trinitro-benzene solutions. Int. J. Chem. Kinetics. 5: 305-320.

CAS (Chemical Abstracts Service). 1989. Chemical Abstracts Registry File (database). Online: 2/28/89.

Chambers, C.W. and P.W. Kabler. 1964. Biodegradability of phenols as related to chemical structure. Dev. Indust. Microbiol. 5: 85-93.

Chambers, C.W., H.H. Tabak and P.W. Kabler. 1963. Degradation of aromatic compounds by phenol-adapted bacteria. J. Water Pollut. Control Fed. 35: 1517-1528.

Chemline. 1989. National Library of Medicine Chemline Database. Online: 2/23/89.

Cody, T.E., S. Witherup, L. Hastings, K. Stemmer and R.T. Chustion. 1981. 1-3-Dinitrobenzene: Toxic effect in vivo. J. Toxicol. Environ. Health. 7: 829-847.

Crockett, P.W., B. Kilian, K.S. Crump and R.B. Howe. 1985. Descriptive Methods for Using Data from Dissimilar Experiments to Locate a No-Adverse-Toxic-Effects Region in the Dose-Duration Plane. Prepared by K.S. Crump and Company, Inc., under Contract No. 6807-007 for Environmental Criteria and Assessment Office, U.S. EPA, Cincinnati, OH.

Durkin, P. and W. Meylan. 1988. User's Guide for D2PLOT: A Program for Dose/Duration Graphs. Prepared by Chemical Hazard Assessment Division, Syracuse Research Corporation under Contract No. 68-C8-0004 for Environmental Criteria and Assessment Office, U.S. EPA, Cincinnati, OH.

Durst, R.A. and R.G. Bates. 1981. Hydrogen-ion activity. In: Kirk-Othmer Encyclopedia of Chemical Technology, 3rd ed., A. Stannden, Ed. John Wiley and Sons, New York, NY. 13: 10-11.

Eisenreich, S.J., B.B. Looney and J.D. Thornton. 1981. Airborne organic contaminants in the Great Lakes ecosystem. Environ. Sci. Technol. 15: 30-38.

Hansch, C. and A.J. Leo. 1985. MedChem Project. Issue No. 26. Pomona College, Claremont, Ca.

Hiatt, R.W., et al. 1957. Relationship of chemical structure to irritant response in marine fish. Nature. 179: 904. (Cited in U.S. EPA, 1980b)

Hine, J. and P.K. Mookerjee. 1975. The intrinsic hydrophilic character of organic compounds. Correlations in terms of structural contributions. J. Org. Chem. 40: 292-298.

Kawai, A., S. Goto, Y. Matsumoto and H. Matsushita. 1987. Mutagenicity of aliphatic and aromatic nitro compounds: Industrial materials and related compounds. Jap. J. Ind. Health. 29(1): 34-54.

Korolev, A.A., T. Vo'itesekhovskaia, M.V. Bogdanov, M.V. Arsen'eva and T.A. Azkharova. 1977. Experimental data on the hygenic standardization of dinitroluene and trintrobenzol in the water reservoirs. Gig. Sanit. 10: 17-20. (Russ.)

Liu, D.H.W., H.C. Bailey and J.G. Pearson. 1983. Toxicity of a complex munitions wastewater to aquatic organisms. In: Proc. Aquatic Toxicology and Hazard Assessment, 6th Symposium, W.E. Bishop, R.D. Cardwell and B.B. Heidolph, Ed. American Society for Testing and Materials (ASTM) STP, Philadelphia, PA. 802: 135-150.

Lyman, W.J., W.F. Reehl and D.H. Rosenblatt. 1982. Handbook of Chemical Property Estimation Methods. McGraw Hill Book Co., New York, NY. p. 4-1 to 4-3, 5-4, Table 5-1, 7-4, 15-1 to 15-21, 15-26 to 15-30.

Mantel, N. and M.A. Schneiderman. 1975. Estimating "safe" levels, a hazardous undertaking. Cancer Res. 35: 1379-1386.

McCormick, N.G., F.E. Feeherry and H.S. Levinson. 1976. Microbial transformation of 2,4,6-trinitrotoluene and other nitroaromatic compounds. Appl. Environ. Microbiol. 31: 949-958.

McGregor, D.B., C.G. Riach, R.M. Hastwell and J.C. Dacre. 1980. Genotoxic activity in microorganisms of tetryl, 1,3-dinitrobenzene and 1,3,5-trinitrobenzene. Environ. Mutagen. 2(4): 531-541.

Pearson, J.G., J.P. Glennon, J.J. Barkley and J.W. Highfill. 1979. An approach to the toxicological evaluation of a complex industrial wastewater. In: Aquatic Toxocology, L.L. Marking and R.A. Kimerle, Ed. American Society for Testing and Materials (ASTM) STP. 667: 284-301.

Purcell, W.P. 1981. Benzene. In: Kirk-Othmer Encyclopedia of Chemical Technology, 3rd ed., A. Stannden, Ed. John Wiley and Sons, New York, NY. p. 749-771.

Rodriguez-Villanueva, J. 1960. The growth of Nocardia V. on various aromatic compounds. Microbiol. Espan. 13: 387-391.

Ryon, M.G., B.C. Pal, S.S. Talmage and R.H. Ross. 1984. Database assessment of the health and environmental effects of munition production waste products. Final Report. ORNL-6018. (NTIS DE84-016512). Oak Ridge National Lab., Oak Ridge, TN. p. 32, 35, 48, 50, 112-113.

SANSS (Structure and Nomenclature Search System). 1989. [Database]. Online: 3/28/89. p. 1-2.

Sax, N.I. and R.J. Lewis. 1987. Hawley's Condensed Chemical Dictionary, 11th ed. van Nostrand Reinhold, Co., New York, NY. p. 1190.

Senczuk, W., J. Jodynis and H. Rogal. 1976. Effect of the chemical structure of some aromatic compounds on their methemoglobin-inducing properties -- Aromatic nitro compounds. Bromat. Chem. Toksykol. 9(3): 289-294. (Russ.) Taken from NIOSH Abstr. 00107007).

Slaga, T.J., L.L. Triplett, L.H. Smith and H.P. Witshi. 1985. Carcinogenesis of Nitrated Toluenes and Benzenes, Skin and Lung Tumor Assays in Mice. Final Report. Govt. Reports Announcements and Index. 19: DE85012081. 33 p.

Spalding, R.F. and J.W. Fulton. 1988. Groundwater munition residues and nitrate near Grand Island, Nebraska. J. Contam. Hydrol. 2: 139-153.

Spanggord, R.J., T. Mill, T.W. Chou, W.R. Mabey, J.H. Smith and S. Lee. 1980. Environmental fate studies on certain munitions wastewater constituents. Final Report, Phase I - Literature Review. SRI Project No. LSU-7934. Contract No. DAMD 17-78-C-8081. U.S. Army Medical Res. and Develop. Command, Fort Detrick, MD.

Spanggord R.J., B.W. Gibson, R.G. Keck, D.W. Thomas and J.J. Barkley, Jr. 1982a. Effluent analysis of waste water generated in the manufacture of 2,4,6-trinitrotoluene. I. Characterization study. Environ. Sci. Technol. 16: 229-232.

Spanggord, R.J., K.E. Mortelmang, A.F. Griffin and V.F. Simmon. 1982b. Mutagenicity in Salmonella typhimurium and structure-activity relationships of waste-water components emanating from the manufacture of trinitrotoluene. Environ. Mutagen. 4(2): 163-180.

Swann, R.L., D.A. Laskowski, P.J. McCall, K. vander-Kuy and H.J. Dishburger. 1983. A rapid method for the estimation of the environmental parameters octanol/water partition coefficient, soil sorption constant, water to air ratio and water solubility. Res. Rev. 85: 17-28.

Tabak, H.H., C.W. Chambers and P.W. Kabler. 1964. Microbial metabolism of aromatic compounds. I. Decomposition of phenolic compounds and aromatic hydrocarbons by phenol-adapted bacteria. J. Bacteriol. 87: 910-919.

Timofievskaya, L.A. and R.P. Rodionova. 1973. Comparative evaluation of the toxicity of some aromatic polynitro compounds. Toksikol. Novykh Promysh. Khim. (Russ.). 13: 138-144.

TSCAPP. 1989. Computer print-out of non-confidential production data from TSCA inventory OPTS, CID, U.S. EPA, Washington, DC. Online: 3/28/89.

U.S. EPA. 1980a. Guidelines and Methodology Used in the Preparation of Health Effect Assessment Chapters of the Consent Decree Water Criteria Documents. Federal Register. 45(231): 79347-79357.

U.S. EPA. 1980b. Identification and listing of hazardous waste under Resource Conservation and Recovery Act (RCRA), Subtitle C, Section 3001; Health and Env. Effect Profiles (40 CFE 261). U.S. Environmental Protection Agency, Washington, DC. October, 1980. PB81-190019.

U.S. EPA. 1984. Methodology amd Guidelines for Ranking Chemicals Based on Chronic Toxicity Data. Prepared by the Office of Health and Environmental Assessment, Environmental Criteria and Assessment Office, Cincinnati, OH for the Office of Emergency and Remedial Response, Washington, DC.

U.S. EPA. 1986a. Methodology for Evaluating Reportable Quantity Adjustments Pursuant to CERCLA Section 102. Prepared by the Carcinogen Assessment Group, Office of Health and Environmental Assessment, Washington, DC for the Office of Emergency and Remedial Response, Washington, DC.

U.S. EPA. 1986b. Guidelines for Carcinogen Risk Assessment. Federal Register. 51(185): 33992-34003.

U.S. EPA. 1987. Graphical Exposure Modeling System (GEMS) Fate of Atmospheric Pollutants (FAP). Office of Toxic Substances, U.S. EPA, Washington, DC.

U.S. EPA 1988. Integrated Risk Information System. Online. Chemical File for 1,3,5-Trinitrobenzene. Office of Health and Environmental Assessment, Environmental Criteria and Assessment Office, Cincinnati, OH.

U.S. EPA/OWRS. 1986. Guidelines for Deriving Numerical National Water Quality Criteria for the Protection of Aquatic Organisms and Their Uses. U.S. EPA, Washington, DC. p. 22-58. NTIS PB85-227049/XAB.

van der Schalie, W.H. 1980. No title provided. In: Aquatic Toxicology. 3rd Symposium. ASTM STP 707. American Society for Testing and Materials, Philadelphia, PA. p. 233-242. (Cited in van der Schalie et al., 1988)

van der Schalie, W.H. 1983. Acute and chronic toxicity of 3,5-dinitroaniline, 1,3-dinitrobenzene and 1,3,5-trinitrobenzene to freshwater aquatic organisms. Report: USAMBRDL-TR-8305. No. AD-A138408. 57 p.

van der Schalie, W.H., T.R. Shedd and M.G. Zeeman. 1988. Ventilatory and movement responses of bluegills exposed to 1,3,5-trinitrobenzene. In: Aquatic Toxicology and Hazard Assessment, W.J. Adams, G.A. Chapman and W.G. Landis, Ed. American Society for Testing and Materials (ASTM) STP, Philadelphia, PA. 971: 307-315.

Watanabe, T., N. Ishihara and M. Ikeda. 1976. Toxicity of and biological monitoring for 1,3-diamino-2,4,6-trinitrobenzene and other nitro-amino derivatives of benzene and chlorobenzene. Intl. Arch. Occup. Environ. Health. 37: 157-168.

Weast, R.C., M.J. Astle and W.H. Beyer. 1988. CRC Handbook of Chemistry and Physics, 69th ed. CRC Press, Inc., Boca Raton, FL. p. C-119.

Wennersten, R. 1980. Extraction of organic pollutants from an effluent stream in the manufacture of p-nitrobenzoic acid. In: Proc. Int. Solvent Extr. Conf. Assoc. Ing. Univ. Liege, Liege, Belgium. 2: 1-6.

Windholz, M., S. Budavari, R.F. Blumetti and E.S. Otterbein. 1983. The Merck Index. Merck and Co., Inc., Rahway, NJ. p. 1389.

APPENDIX A

LITERATURE SEARCHED

This HEED is based on data identified by computerized literature searches of the following:

CHEMLINE
TSCATS
CASR online (U.S. EPA Chemical Activities Status Report)
TOXLINE
TOXLIT
TOXLIT 65
RTECS
OHM TADS
STORET
SRC Environmental Fate Data Bases
SANSS
AQUIRE
TSCAPP
NTIS
Federal Register
CAS ONLINE (Chemistry and Aquatic)
HSDB

These searches were conducted in May, 1989, and the following secondary sources were reviewed:

ACGIH (American Conference of Governmental Industrial Hygienists). 1986. Documentation of the Threshold Limit Values and Biological Exposure Indices, 5th ed. Cincinnati, OH.

ACGIH (American Conference of Governmental Industrial Hygienists). 1987. TLVs: Threshold Limit Values for Chemical Substances in the Work Environment adopted by ACGIH with Intended Changes for 1987-1988. Cincinnati, OH. 114 p.

Clayton, G.D. and F.E. Clayton, Ed. 1981. Patty's Industrial Hygiene and Toxicology, 3rd rev. ed., Vol. 2A. John Wiley and Sons, NY. 2878 p.

Clayton, G.D. and F.E. Clayton, Ed. 1981. Patty's Industrial Hygiene and Toxicology, 3rd rev. ed., Vol. 2B. John Wiley and Sons, NY. p. 2879-3816.

Clayton, G.D. and F.E. Clayton, Ed. 1982. Patty's Industrial Hygiene and Toxicology, 3rd rev. ed., Vol. 2C. John Wiley and Sons, NY. p. 3817-5112.

Grayson, M. and D. Eckroth, Ed. 1978-1984. Kirk-Othmer Encyclopedia of Chemical Technology, 3rd ed. John Wiley and Sons, NY. 23 Volumes.

Hamilton, A. and H.L. Hardy. 1974. Industrial Toxicology, 3rd ed. Publishing Sciences Group, Inc., Littleton, MA. 575 p.

IARC (International Agency for Research on Cancer). IARC Monographs on the Evaluation of Carcinogenic Risk of Chemicals to Humans. IARC, WHO, Lyons, France.

Jaber, H.M., W.R. Mabey, A.T. Lieu, T.W. Chou and H.L. Johnson. 1984. Data acquisition for environmental transport and fate screening for compounds of interest to the Office of Solid Waste. EPA 500/6-84-010. NTIS PB84-243906. SRI International, Menlo Park, CA.

NTP (National Toxicology Program). 1987. Toxicology Research and Testing Program. Chemicals on Standard Protocol. Management Status.

Ouellette, R.P. and J.A. King. 1977. Chemical Week Pesticide Register. McGraw-Hill Book Co., NY.

Sax, I.N. 1984. Dangerous Properties of Industrial Materials, 6th ed. Van Nostrand Reinhold Co., NY.

SRI (Stanford Research Institute). 1987. Directory of Chemical Producers. Menlo Park, CA.

U.S. EPA. 1986. Report on Status Report in the Special Review Program, Registration Standards Program and the Data Call in Programs. Registration Standards and the Data Call in Programs. Office of Pesticide Programs, Washington, DC.

USITC (U.S. International Trade Commission). 1986. Synthetic Organic Chemicals. U.S. Production and Sales, 1985, USITC Publ. 1892, Washington, DC.

Verschueren, K. 1983. Handbook of Environmental Data on Organic Chemicals, 2nd ed. Van Nostrand Reinhold Co., NY.

Windholz, M., Ed. 1983. The Merck Index, 10th ed. Merck and Co., Inc., Rahway, NJ.

Worthing, C.R. and S.B. Walker, Ed. 1983. The Pesticide Manual. British Crop Protection Council. 695 p.

In addition, approximately 30 compendia of aquatic toxicity data were reviewed, including the following:

Battelle's Columbus Laboratories. 1971. Water Quality Criteria Data Book. Volume 3. Effects of Chemicals on Aquatic Life. Selected Data from the Literature through 1968. Prepared for the U.S. EPA under Contract No. 68-01-0007. Washington, DC.

Johnson, W.W. and M.T. Finley. 1980. Handbook of Acute Toxicity of Chemicals to Fish and Aquatic Invertebrates. Summaries of Toxicity Tests Conducted at Columbia National Fisheries Research Laboratory. 1965-1978. U.S. Dept. Interior, Fish and Wildlife Serv. Res. Publ. 137, Washington, DC.

McKee, J.E. and H.W. Wolf. 1963. Water Quality Criteria, 2nd ed. Prepared for the Resources Agency of California, State Water Quality Control Board. Publ. No. 3-A.

Pimental, D. 1971. Ecological Effects of Pesticides on Non-Target Species. Prepared for the U.S. EPA, Washington, DC. PB-269605.

Schneider, B.A. 1979. Toxicology Handbook. Mammalian and Aquatic Data. Book 1: Toxicology Data. Office of Pesticide Programs, U.S. EPA, Washington, DC. EPA 540/9-79-003. NTIS PB 80-196876.

APPENDIX B

Summary Table for 1,3,5-Trinitrobenzene

	Species	Exposure	Effect	RfD or q_1*	Reference
Inhalation Exposure					
Subchronic	ID	ID	ID	ID	ID
Chronic	ID	ID	ID	ID	ID
Carcinogenicity	ID	ID	ID	ID	ID
Oral Exposure					
Subchronic	rat	3 ppm 1,3-dinitrobenzene in drinking water for 16 weeks (0.4 mg/kg/day equivalent to 0.51 mg/kg/day 1,3,5-trinitrobenzene)	increased spleen weight at higher levels	$5x10^{-4}$ mg/kg/day	Cody et al., 1981
Chronic	rat	3 ppm 1,3-dinitrobenzene in drinking water for 16 weeks (0.4 mg/kg/day equivalent to 0.51 mg/kg/day 1,3,5-trinitrobenzene)	increased spleen weight at higher levels	$5x10^{-5}$ mg/kg/day	Cody et al., 1981
Carcinogenicity	ID	ID	ID	ID	ID

REPORTABLE QUANTITIES

Based on Chronic Toxicity: 100

Based on Carcinogenicity: ID

ID = Insufficient Data

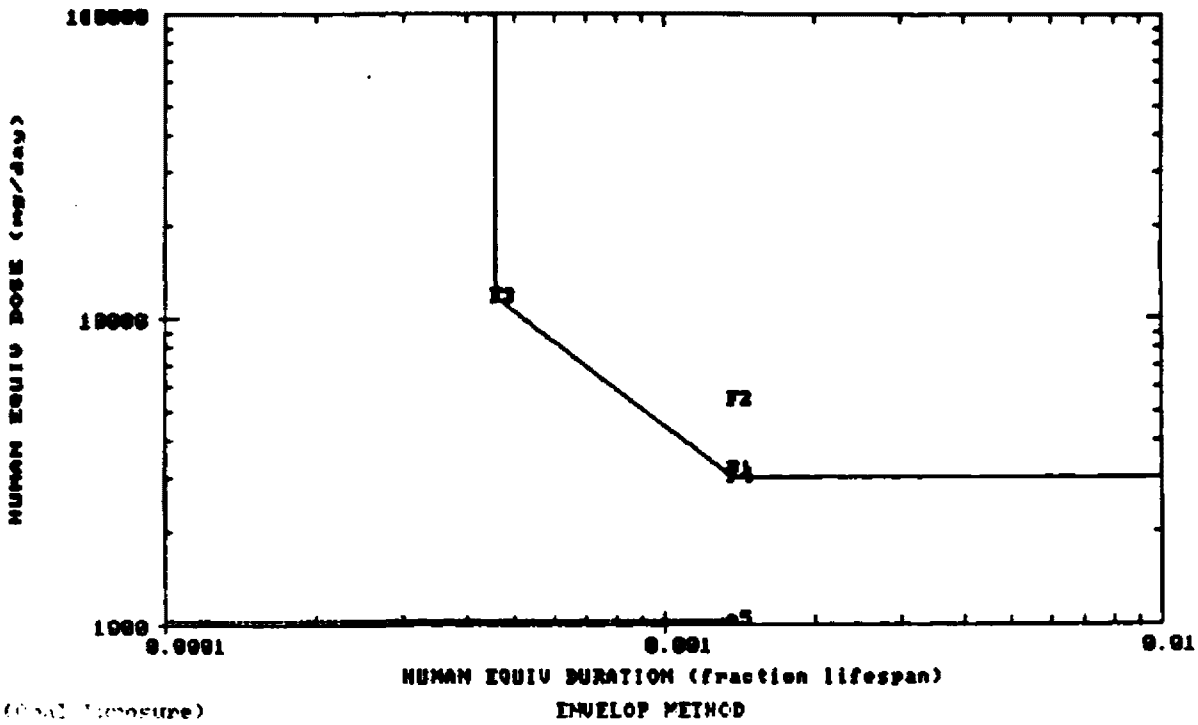
APPENDIX C

DOSE/DURATION RESPONSE GRAPHS FOR EXPOSURE TO 1,3,5-TRINITROBENZENE

C.1. DISCUSSION

Dose/duration-response graph(s) for inhalation and oral exposure to 1,3,5-trinitrobenzene generated by the method of Crockett et al. (1985), using the computer software by Durkin and Meylan (1988), and developed under contract to ECAO-Cincinnati is presented in Figure C-1. Data used to generate these graphs are presented in Section C.2. In the generation of this figure, all responses are classified as adverse (FEL, AEL or LOAEL) or nonadverse (NOEL or NOAEL) for plotting. For oral exposure, the ordinate expresses dosage as human equivalent dose. The animal dosage in mg/kg/day is multiplied by the cube root of the ratio of the animal:human body weight to adjust for species differences in basal metabolic rate (Mantel and Schneiderman, 1975). The result is then multiplied by 70 kg, the reference human body weight, to express the human equivalent dose as mg/day for a 70 kg human.

The boundary for adverse effects (solid line) is drawn by identifying the lowest adverse effect dose or concentration at the shortest duration of exposure at which an adverse effect occurred. From this point, an infinite line is extended upward, parallel to the dose axis. The starting point is then connected to the lowest adverse effect dose or concentration at the next longer duration of exposure that has an adverse effect dose or concentration equal to or lower than the previous one. This process is continued to the lowest adverse effect dose or concentration. From this point, a line is extended to the right, parallel to the duration axis. The region of adverse effects lies above the adverse effects boundary.



Key: F = FEL
N = NOAEL

FIGURE C-1

Dose/Duration - Response Graph for Inhalation Exposure to 1,3,5-Trinitrobenzene Envelope Method (expanded experimental concentration)

Using the envelope method, the boundary for no adverse effects (dashed line) is drawn by identifying the highest no adverse effects dose or concentration. From this point, a line parallel to the duration axis is extended to the dose or concentration axis. The starting point is then connected to the next lower or equal no adverse effect dose or concentration at a longer duration of exposure. When this process can no longer be continued, a line is dropped parallel to the dose or concentration axis to the duration axis. The no adverse effects region lies below the no adverse effects boundary. At both ends of the graph between the adverse effects and no adverse effects boundaries are regions of ambiguity. The area (if any) resulting from the intersection of the adverse effects and no adverse effects boundaries is defined as the region of contradiction.

In the censored data method, all no adverse effect points located in the region of contradiction are dropped from consideration and the no adverse effect boundary is redrawn so that it does not intersect the adverse effects boundary and no region of contradiction is generated. This method results in the most conservative definition of the no adverse effects region.

The lack of data for oral exposure to 1,3,5-trinitrobenzene is reflected in the large area of ambiguity defined in Figure C-1. The boundary for adverse effects for oral exposure to 1,3,5-trinitrobenzene is defined in Figure C-1 by two data points. Starting from the upper left, these points represent the LD_{50} value (730 mg/kg/day, Rec. #3) in guinea pigs (Korolev et al., 1977) and the lowest LD_{50} value (572 mg/kg/day, Rec. #4) from the study by Timofievskaya and Rodionova (1973). The boundary for no adverse effects for oral exposure to 1,3,5-trinitrobenzene is defined in Figure C-1 by the NOAEL for methemoglobin effects (86 mg/kg/day, Rec. #5) from the rat study by Senczuk et al. (1973).

C.2. DATA USED TO GENERATE DOSE/DURATION-RESPONSE GRAPHS

C.2.1. Inhalation Exposure

Chemical Name: 1,3,5-Trinitrobenzene
CAS Number: 99-35-4
Document Title: Health and Environmental Effects Document for 1,3,5-Trinitrobenzene
Document Number:
Document Date:
Document Type: HEED

RECORD #1:

Species:	Mice	Dose:	600.000
Sex:	NR	Duration Exposure:	1.0 days
Effect:	FEL	Duration Observation:	1.0 days
Route:	Oral (NOS)		

Number Exposed:	NR
Number Responses:	NR
Type of Effect:	DEATH
Site of Effect:	BODY
Severity Effect:	10

Comment: LD_{50} value for white mice.

Citation: Korolev et al., 1977

RECORD #2

Species:	Rats	Dose:	450.000
Sex:	NR	Duration Exposure:	1.0 days
Effect:	FEL	Duration Observation:	1.0 days
Route:	Oral (NOS)		

Number Exposed:	NR
Number Responses:	NR
Type of Effect:	DEATH
Site of Effect:	BODY
Severity Effect:	10

Comment: LD_{50} value for white rats.

Citation: Korolev et al., 1977

RECORD #3:

Species:	Guinea pigs	Dose:	730.000
Sex:	NR	Duration Exposure:	1.0 days
Effect:	FEL	Duration Observation:	1.0 days
Route:	Oral (NOS)		

Number Exposed:	NR
Number Responses:	NR
Type of Effect:	DEATH
Site of Effect:	BODY
Severity Effect:	10

Comment: LD_{50} value for guinea pigs.

Citation: Korolev et al., 1977

RECORD #4:

Species:	Mice	Dose:	572.000
Sex:	NR	Duration Exposure:	1.0 days
Effect:	FEL	Duration Observation:	1.0 days
Route:	Oral (NOS)		

Number Exposed:	NR
Number Responses:	NR
Type of Effect:	DEATH
Site of Effect:	BODY
Severity Effect:	10

Comment: LD_{50} value for mice.

Citation: Timofievskaya and Rodionova, 1973

RECORD #5:

Species:	Rats	Dose:	86.000
Sex:	NR	Duration Exposure:	1.0 days
Effect:	NOAEL	Duration Observation:	1.0 days
Route:	Oral (NOS)		

Number Exposed:	NR
Number Responses:	NR
Type of Effect:	ENZYM
Site of Effect:	BLOOD
Severity Effect:	2

Comment: Single oral dose (0.4 µmol/kg). Increased methemoglobin formation in blood. Degree of increase not specified in English abstract.

Citation: Senczuk et al. 1976

NR = Not reported