DRAFT
INTERACTION PROFILE FOR:
ARSENIC, HYDRAZINES, JET FUELS, STRONTIUM-90, AND TRICHLOROETHYLENE

U.S. Department of Health and Human Services
Public Health Service
Agency for Toxic Substances and Disease Registry

Public Comment Period Ends November 30, 2002

PREFACE

The Comprehensive Environmental Response, Compensation, and Liability Act (CERCLA) mandates that the Agency for Toxic Substances and Disease Registry (ATSDR) shall assess whether adequate information on health effects is available for the priority hazardous substances. Where such information is not available or under development, ATSDR shall, in cooperation with the National Toxicology Program, initiate a program of research to determine these health effects. The Act further directs that where feasible, ATSDR shall develop methods to determine the health effects of substances in combination with other substances with which they are commonly found. The Food Quality Protection Act (FQPA) of 1996 requires that factors to be considered in establishing, modifying, or revoking tolerances for pesticide chemical residues shall include the available information concerning the cumulative effects of substances that have a common mechanism of toxicity, and combined exposure levels to the substance and other related substances. The FQPA requires that the Administrator of the Environmental Protection Agency consult with the Secretary of the Department of Health and Human Services (which includes ATSDR) in implementing some of the provisions of the act.

To carry out these legislative mandates, ATSDR's Division of Toxicology (DT) has developed and coordinated a mixtures program that includes trend analysis to identify the mixtures most often found in environmental media, *in vivo* and *in vitro* toxicological testing of mixtures, quantitative modeling of joint action, and methodological development for assessment of joint toxicity. These efforts are interrelated. For example, the trend analysis suggests mixtures of concern for which assessments need to be conducted. If data are not available, further research is recommended. The data thus generated often contribute to the design, calibration or validation of the methodology. This pragmatic approach allows identification of pertinent issues and their resolution as well as enhancement of our understanding of the mechanisms of joint toxic action. All the information obtained is thus used to enhance existing or developing methods to assess the joint toxic action of environmental chemicals. Over a number of years, ATSDR scientists in collaboration with mixtures risk assessors and laboratory scientists have developed approaches for the assessment of the joint toxic action of chemical mixtures. As part of the mixtures program a series of documents, Interaction Profiles, are being developed for certain priority mixtures that are of special concern to ATSDR.

The purpose of an Interaction Profile is to evaluate data on the toxicology of the "whole" priority mixture (if available) and on the joint toxic action of the chemicals in the mixture in order to recommend approaches for the exposure-based assessment of the potential hazard to public health. Joint toxic action includes additivity and interactions. A weight-of-evidence approach is commonly used in these documents to evaluate the influence of interactions in the overall toxicity of the mixture. The weight-of-evidence evaluations are qualitative in nature, although ATSDR recognizes that observations of toxicological interactions depend greatly on exposure doses and that some interactions appear to have thresholds. Thus, the interactions are evaluated in a qualitative manner to provide a sense of what influence the interactions may have when they do occur.

The public comment period ends on November 30, 2002. Comments should be sent to:

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All reviewers were selected in conformity with the conditions for peer review specified in Section 104(I)(13) of the Comprehensive Environmental Response, Compensation, and Liability Act, as amended.

Scientists from the Agency for Toxic Substances and Disease Registry (ATSDR) have reviewed the peer reviewers' comments and determined which comments will be addressed in the profile. A listing of the peer reviewers' comments not incorporated in the profile, with a brief explanation of the rationale for their exclusion, exists as part of the administrative record for this mixture. A list of databases reviewed and a list of unpublished documents cited are also included in the administrative record.

The citation of the peer review panel should not be understood to imply its approval of the profile's final content. The responsibility for the content of this profile lies with the ATSDR.

SUMMARY

The mixture of jet fuels, hydrazines, trichloroethylene, arsenic, and strontium-90 was chosen to represent potential exposures in the vicinity of sites where past and/or present activities include use and/or release of these materials. Such sites might include rocket testing facilities, air force bases, and similar installations. Activities at such sites might include use of jet fuels and hydrazines as aircraft and rocket fuels and trichloroethylene as a solvent to clean engine components. Such sites sometimes include or are co-located with nuclear research facilities or radioactive waste storage sites, where strontium-90 may be found in spent nuclear fuel rods. Arsenic, although not necessarily used or produced at such sites, is frequently detected at hazardous waste sites and would not be unexpected at any specific site. The purposes of this profile are: (1) to evaluate data (if available) on the health hazards and corresponding dose-response relationships associated with exposure to this five-component mixture as a whole; (2) to evaluate data on the joint toxic actions of components of this mixture; and (3) to make recommendations for exposure-based assessments of the potential impact of joint toxic action of the mixture on public health.

The primary route of exposure for offsite receptors (i.e., receptors located beyond the borders of the site where the materials have been used or released) is expected to be oral for all five of these substances, resulting from contamination of soil and/or ground or surface water. Inhalation is also a potential route of exposure for jet fuels, hydrazines, and trichloroethylene, all of which are volatile. However, due to rapid degradation of hydrazine in air and dispersion of all chemicals during transport offsite, inhalation is expected to be a relatively minor route of exposure for offsite receptors under most conditions. Potential exceptions may occur when contaminated groundwater is used as household water, resulting in volatilization of the chemicals into indoor air, or when contamination of groundwater and subsurface soil results in migration of these chemicals into basements as soil gas. While inhalation is an important route of exposure to arsenic at industrial facilities that generate arsenic particulates (e.g., smelters), it is not relevant to arsenic at the sites being considered here. Catastrophic accidental release of strontium-90 to the air from nuclear facilities is possible, but is beyond the scope of this document.

No studies were located that examined health effects in humans or research animals exposed to mixtures containing jet fuels, hydrazines, trichloroethylene, arsenic, and strontium-90, and no physiologically-based pharmacokinetic/pharmacodynamic (PBPK/PD) models for this mixture have been developed. Binary weight-of-evidence (BINWOE) analysis of the joint toxic action of the component pairs was indeterminate for most pairs due to scarcity of data regarding joint toxic action of the component pairs and insufficient understanding of toxic and pharmacokinetic mechanisms of the individual substances, but did predict additivity for depression of the central nervous system from exposure to jet fuels and trichloroethylene and a greater-than-additive effect of strontium on arsenic toxicity due to inhibition by strontium of arsenic metabolism.

Although the BINWOEs were indeterminate for all of the remaining pairs due to insufficient data, the extensive overlap of toxic endpoints for the five mixture components suggests that there is a potential for joint toxic action among these substances. Therefore, it is reasonable to be cautious when evaluating public health concerns for this mixture by assuming additivity.

The hazard index approach is recommended as an additive component-based method for assessing possible health hazards from noncancer effects for mixtures of jet fuels, hydrazines, trichloroethylene, arsenic, and strontium-90. The hazard index approach allows for summing across routes of exposure to account for multiple pathways of exposure, which may be important for this mixture. For oral exposure, the lack of health guidance values is problematic and leaves only arsenic and trichloroethylene contributing to the hazard index for oral exposure to the mixture. Because these chemicals affect many of the same sensitive endpoints (neurological, renal, and immunological targets), it is recommended to calculate hazard indexes for oral exposure using both chemicals. For inhalation exposure, intermediate Minimal Risk Levels (MRLs) are available for all three chemicals for which this route is expected to potentially contribute to exposure to offsite receptors at rocket launch sites: jet fuels and hydrazines based on liver effects and trichloroethylene based on neurological effects. Because the central nervous system and the liver are sensitive targets for all three chemicals, it is recommended that inhalation hazard indexes be calculated using all three chemicals together. Application of the target-organ toxicity dose (TTD) modification of the hazard index method is not justified by the existing data set.

For cancer effects, the cancer risk for each substance (calculated from the lifetime average daily intake and the potency factor) is summed to provide an estimate of risk due to the whole mixture. Risk can be summed across routes to account for multiple pathways of exposure.

Additive approaches to assessment for this mixture are only needed when there is reason to believe that two or more chemicals in the mixture contribute significantly to the public health assessment. Therefore, hazard indexes are only calculated if two or more of the individual components have hazard quotients equaling or exceeding 0.1, and cancer risks are summed only if estimated risks exceed 1×10^{-6} for at least two components.

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LIST OF ACRONYMS, ABBREVIATIONS, AND SYMBOLS

ACGIH	American Conference of Governmental Industrial Hygienists
As	arsenic
As_2O_3	arsenic trioxide
ATSDR	Agency for Toxic Substances and Disease Registry
BINWOE	binary weight-of-evidence
BrdU	bromo-deoxyuridine
CERCLA	Comprehensive Environmental Response, Compensation, and Recovery Act
CI	confidence interval
DNA	deoxyribonucleic acid
DMA	dimethylarsinic acid
DT	Division of Toxicology
EDTA	ethylenediaminetetraacetic acid
EPA	Environmental Protection Agency
FQPA	Food Quality Protection Act
gastro	gastrointestinal
GST-P	glutathione S-transferase-placental
HEAST	Health Effects Assessment Summary Tables
IARC	International Agency for Research on Cancer
IRIS	Integrated Risk Information System
kg	kilogram
L	liter
LOAEL	lowest-observed-adverse-effect level
m^3	cubic meter
MeV	millions of electron volts
mg	milligram
MMA	monomethylarsonic acid
MRL	Minimal Risk Level
neuro	neurological
NOAEL	no-observed-adverse-effect level
NTP	National Toxicology Program
PBPK	physiologically based pharmacokinetic
PBPK/PD	physiologically-based pharmacokinetic/pharmacodynamic
pCi	picocurie
ppm	parts per million
RfC	Reference Concentration
RfD	Reference Dose
RR	relative risk
Sr	strontium
$SrCl_2$	strontium chloride
$t_{1/2}$	half-life
TPH	total petroleum hydrocarbons
TTD	target-organ toxicity dose
TUNEL	TdT-mediated dUTP digoxigenin nick end labeling
µg	microgram
µM	micromolar
U.S.	United States
WOE	weight-of-evidence
Y	Yttrium
Zn	Zinc
Zr	Zirconium
>	greater than
≥	greater than or equal to
=	equal to
<	less than
≤	less than or equal to

1. Introduction

The primary purpose of this Interaction Profile for jet fuels, hydrazines, trichloroethylene, arsenic, and strontium-90 is to evaluate data on the toxicology of the "whole" mixture and the joint toxic action of the chemicals in the mixture in order to recommend approaches for assessing the potential hazard of this mixture to public health. To this end, the profile evaluates the whole mixture data (if available), focusing on the identification of health effects of concern, adequacy of the data as the basis for a mixture Minimal Risk Level (MRL), and adequacy and relevance of physiologically-based pharmacokinetic/ pharmacodynamic models (PBPK/PD) for the mixture. The profile also evaluates the evidence for joint toxic action—additivity and interactions—among the mixture components. A weight-of-evidence (WOE) approach is commonly used in these profiles to evaluate the influence of interactions in the overall toxicity of the mixture. The weight-of-evidence evaluations are qualitative in nature, although the Agency for Toxic Substances and Disease Registry (ATSDR) recognizes that observations of toxicological interactions depend greatly on exposure doses and that some interactions appear to have thresholds. Thus, the interactions are evaluated in a qualitative manner to provide a sense of what influence the interactions may have when they do occur. The profile provides environmental health scientists with ATSDR Division of Toxicology's (DT) recommended approaches for the incorporation of the whole mixture data or the concerns for additivity and interactions into an assessment of the potential hazard of this mixture to public health. These approaches can then be used with specific exposure data from hazardous waste sites or other exposure scenarios.

The mixture of jet fuels, hydrazines, trichloroethylene, arsenic, and strontium-90 was chosen to represent potential exposures in the vicinity of sites where past and/or present activities include use and/or release of these materials. Such sites might include rocket testing facilities, air force bases, and similar installations. Activities at such sites might include use of jet fuels and hydrazines as aircraft and rocket fuels and trichloroethylene as a solvent to clean engine components. Such sites sometimes include or are co-located with nuclear research facilities or radioactive waste storage sites, where strontium-90 may be found in spent nuclear fuel rods. Arsenic, although not necessarily used or produced at such sites, is frequently detected at hazardous waste sites and would not be unexpected at any specific site.

The primary route of exposure for offsite receptors (i.e., receptors located beyond the borders of the site where the materials have been used or released) is expected to be oral for all five of these substances, resulting from contamination of soil and/or ground or surface water. Inhalation is also a potential route of exposure for jet fuels, hydrazines, and trichloroethylene, all of which are volatile. However, due to rapid degradation of hydrazine in air and dispersion of all chemicals during transport offsite, inhalation is expected to be a relatively minor route of exposure for offsite receptors under most conditions. Inhalation exposure may occur when contaminated groundwater is used as household water, resulting in volatilization of the chemicals into indoor air, or when contamination of groundwater and subsurface soil results in migration of these chemicals into basements as soil gas. While inhalation is an important route of

exposure to arsenic at industrial facilities that generate arsenic particulates (e.g., smelters), it is not relevant to arsenic at the sites being considered here. Catastrophic accidental release of strontium-90 to the air from nuclear facilities is possible, but is beyond the scope of this document.

Before evaluating the relevance of interactions data for these substances, an understanding of the end-points of concern for this mixture is needed. The endpoints of concern include the critical effects that are the bases for MRLs, as well as other sensitive endpoints of the individual substances. Endpoints in common to multiple substances that may become significant due to additivity or interactions are also considered.

Jet fuels are complex mixtures of hydrocarbons produced by distillation of petroleum crude oil. Most jet fuels (e.g., JP-5, JP-7, JP-8) are middle distillates similar in composition to kerosene, although some (e.g., JP-4) also include lower boiling naphtha streams, like those used to produce gasoline. For jet fuels and related substances, intermediate and chronic inhalation MRLs are available based on liver effects (hepatocellular fatty change, hepatic inflammation) in animal studies (ATSDR 1995a, 1995b, 1998). Liver effects were also reported after oral exposure to jet fuels, although the oral data were insufficient to support derivation of MRLs. Other endpoints of concern for jet fuels are central nervous system depression, which is a well-known effect of jet fuels in humans exposed by any route of exposure, and immunosuppression. While jet fuels have been shown to produce hyaline droplet nephropathy in male rats, this effect is not predictive of renal effects in humans and is, therefore, not considered in this analysis. Jet fuels are not genotoxic and have not been demonstrated to be carcinogenic. See Appendix A for more information.

The hydrazines considered in this document are hydrazine and 1,1-dimethylhydrazine, which have both been used as rocket fuel. Both of these compounds have intermediate inhalation MRLs based on liver effects (ATSDR 1997a). Oral data confirm that the liver is a target by this route as well, but the data are too limited to support MRL derivation. The central nervous system is a prominent target of hydrazines in humans and animals by any route of exposure. Other targets of concern for hydrazines include the respiratory tissues (following inhalation exposure), the blood (anemia), and the reproductive organs of both males and females (ovarian and testicular atrophy). Both hydrazine and 1,1-dimethylhydrazine are genotoxic carcinogens, producing multiple tumor types in rodents by inhalation, oral, and parenteral exposure. The U.S. Environmental Protection Agency (EPA) has derived an oral slope factor and inhalation unit risk for hydrazine (IRIS 2001). Further details regarding hydrazines can be found in Appendix B.

The most sensitive targets for trichloroethylene are the central nervous system (central nervous system depression, neurobehavioral deficits, hearing loss) and the liver (changes in serum cholesterol and bile acids, liver enlargement, and cellular hypertrophy). Trichloroethylene has acute and intermediate inhalation MRLs and a draft chronic reference concentration (RfC) based on central nervous system

effects, and an acute oral MRL based on neurological effects and draft chronic oral reference dose (RfD) based on liver effects (ATSDR 1997b; EPA 2001). Other sensitive targets for trichloroethylene are the kidneys (increased kidney weights and cytomegaly and karyomegaly in renal tubular epithelial cells), endocrine system (altered hormone levels), immune system (depressed immune function, autoimmune disease), male reproductive system (decreases in sperm count and motility), and developing fetus (cardiac and eye malformations, neurobehavioral alterations). Recent analyses have concluded that trichloroethylene is probably carcinogenic to humans (EPA 2001; IARC 1995; NTP 2001), and EPA (2001) has derived draft oral slope factors and inhalation unit risks for the chemical. Appendix C contains additional information regarding trichloroethylene.

For this mixture, exposure to arsenic is assumed to be entirely by the oral route, as discussed above. Chronic oral exposure to arsenic produces characteristic dermal lesions in humans that are the basis for the chronic oral MRL (ATSDR 2000) and EPA's chronic oral RfD (IRIS 2001). A provisional acute oral MRL was based on facial (periorbital) edema and gastrointestinal irritation in humans (ATSDR 2000). Other endpoints of concern for ingested arsenic are vascular disease, peripheral and central neuropathy, anemia, leukopenia, and renal effects, all of which have been observed in humans. Arsenic is a known human carcinogen, and EPA has derived an oral slope factor for this chemical (IRIS 2001). A point of interest is that there appears to be no good animal model for arsenic toxicity in humans. No other species has been found to develop the arsenic effect of greatest concern, cancer in the skin and other organs. Nor have the studied species of animals been found to develop the noncancer skin lesions seen in humans exposed to arsenic. The species most often used in interactions studies, the rat, is significantly different from humans in terms of arsenic metabolism, distribution, and health effects. For more information on arsenic, see Appendix D.

As discussed previously, exposure to strontium-90 for this mixture is assumed to be entirely by the oral route. ATSDR (2001c) did not derive oral MRLs for strontium-90, and EPA has not derived an RfD (IRIS 2001). Since radiostrontium is preferentially retained in bone, and therefore has a long biological half-life, internal exposures of any duration will lead to chronic internal exposure to ionizing radiation. Consequently, the most significant effects of exposure to absorbed radioactive strontium are necrosis and cancers of bone, bone marrow, and tissues adjacent to bone. Noncancer effects include dystrophic and osteolytic lesions in bone, anemia, and immunosuppression. Radioactive strontium is a known human carcinogen. EPA (1997) has calculated oral slope factors (lifetime risk per picocurie [pCi]) for ingested strontium-90 (4.09×10^{-11} for ^{90}Sr and 5.59×10^{-11} for ^{90}Sr plus disintegration products). For more details, see Appendix E.

Information on the toxicity of the individual substances in the jet fuels, hydrazines, trichloroethylene, arsenic, and strontium-90 mixture is summarized in Tables 1–3. Table 1 shows the availability of MRLs and RfDs/RfCs for the individual substances. The availability of cancer assessments is shown in Table 2. Table 3 displays the endpoints of concern for each substance. Additional information about the individual substances can be found in Appendices A–E.

Table 1. Critical Endpoints for Noncancer Health Guidance Values for the Mixture of Jet Fuels, Hydrazines, Trichloroethylene, Arsenic, and Strontium-90

	Inhalation				Oral			
	Acute MRL	Intermediate MRL	Chronic MRL	RfC	Acute MRL	Intermediate MRL	Chronic MRL	RfD
Jet fuels								
JP-4	—	Liver	—	—	—	—	—	—
JP-5	—	Liver	—	—	—	—	—	—
JP-7	—	—	Liver	—	—	—	—	—
JP-8	—	Liver	—	—	—	—	—	—
Kerosene	—	Liver	—	—	—	—	—	—
Hydrazines								
Hydrazine	—	Liver	—	—	—	—	—	—
1,1-Dimethyl-hydrazine	—	Liver	—	—	—	—	—	—
Trichloroethylene	Neuro	Neuro	—	Neuro	Neuro	—	—	Liver
Arsenic[a]					Dermal/ gastro[b]	—	Dermal	Dermal
Strontium-90[a]					—	—	—	—

[a]Inhalation exposure is not relevant for these chemicals under the assumed conditions.
[b]Provisional value

MRL = Minimal Risk Level; RfC = reference concentration; RfD = reference dose

Table 2. Cancer Assessments for the Mixture of Jet Fuels, Hydrazines, Trichloroethylene, Arsenic, and Strontium-90

	Weight-of-evidence category			Basis for quantitative assessment		
	IARC[a]	NTP[b]	EPA[c]	Inhalation unit risk	Oral slope factor	Genotoxicity
Jet fuels	3	—	—	—	—	Negative
JP-4						
JP-5						
JP-7						
JP-8						
Kerosene						
Hydrazines						
Hydrazine	2B	R	B2	Nasal tumors	Liver tumors	Positive
1,1-Dimethyl-hydrazine	2B	R	—	—	—	Positive
Trichloroethylene	2A	R	B1	Kidney tumors	Liver and kidney tumors, lymphoma	Weak positive
Arsenic[d]	1	K	A		Dermal tumors	Mixed: weak mutagen, but clastogenic
Strontium-90[d]	1	—	A		Internal tumors	Positive

[a]1 = carcinogenic to humans; 2A = probably carcinogenic to humans; 2B = possibly carcinogenic to humans; 3 = not classifiable as to carcinogenicity in humans; 4 = probably not carcinogenic to humans
[b]K = known to be a human carcinogen; R = reasonably anticipated to be a human carcinogen
[c]A = human carcinogen; B = probable human carcinogen (B1 based on human data, B2 based on animal data); C = possible human carcinogen; D = not classifiable as to human carcinogenicity; E = evidence of non-carcinogenicity for humans
[d]Inhalation exposure not relevant for these chemicals under the assumed conditions.

EPA = Environmental Protection Agency; IARC = International Agency for Research on Cancer; NTP = National Toxicology Program

Table 3. Potential Health Effects of Concern for Mixtures of Jet Fuels, Hydrazines, Trichloroethylene, Arsenic, and Strontium-90

Jet fuels	Hydrazines	Trichloroethylene	Arsenic	Strontium-90
Hepatic[a] Neurological Immunological	**Hepatic**[a] Respiratory Hematological Neurological Reproductive Cancer	**Neurological**[a] **Hepatic**[a] Renal Endocrine Immunological Reproductive Developmental Cancer	**Dermal**[a] **Gastrointestinal**[a] Cardiovascular Hematological Renal Neurological Immunological Cancer	Hematological Musculoskeletal Immunological Cancer

[a]basis for MRL/RfC/RfD

MRL = Minimal Risk Level; RfC = reference concentration; RfD = reference dose

2. Joint Toxic Action Data for the Mixture of Concern and Component Mixtures

This chapter provides a review and evaluation of the literature pertinent to joint toxic action of the mixture and its components. Few relevant data were located for the mixture of jet fuels, hydrazines, trichloroethylene, arsenic, and strontium-90.

2.1 Mixture of Concern

No studies were located that examined health effects or pharmacokinetic endpoints in humans or research animals exposed to mixtures containing jet fuels, hydrazines, trichloroethylene, arsenic, and strontium-90. No physiologically-based pharmacokinetic (PBPK) models were found for mixtures of these five components.

2.2 Component Mixtures

No studies were located that examined health effects or pharmacokinetic endpoints in humans or research animals exposed to three- or four-membered mixtures of the five components of concern. No PBPK models were found for three- or four-membered mixtures of these chemicals.

The following subsections present evaluations of health effects data and discussions of mechanistic information pertinent to the joint toxic action of each pair of components.

2.2.1 Jet Fuels and Hydrazines

No studies were located regarding possible joint toxic actions between jet fuels and hydrazines in affecting health-related endpoints in humans or research animals. No PBPK models for co-exposure to jet fuels and hydrazines were found. Jet fuels and hydrazines both produce effects in the liver and central nervous system. In the liver, both substances produce inflammation, fatty degeneration, and necrosis. For both substances, a proposed mechanism of hepatic effects involves metabolism and generation of reactive oxygen species; however, mechanistic understanding of the hepatic effects of jet fuels and hydrazines is not sufficient to make reliable predictions as to the hepatic effects of joint exposure. Both jet fuels and hydrazines have been shown to cause neurological effects. However, the mechanisms believed to be responsible for these effects differ for the two classes of compounds, with jet fuels believed to disrupt function of nerve cell membrane proteins by physical presence of the solvent in the membrane, whereas hydrazines are believed to form hydrazones with vitamin B6 derivatives, thereby inhibiting reactions that require vitamin B6 as a cofactor and inducing a functional deficiency of vitamin B6 (see Appendices A and B). Understanding of these mechanisms is inadequate to make reliable predictions as to the neurological effects of joint exposure. Hydrazines have been demonstrated to cause multiple tumor

types in animal studies. No mechanistic information was located as to potential effects of jet fuels, which have not been demonstrated to be carcinogenic, on the carcinogenic effects of hydrazines.

2.2.2 Jet Fuels and Trichloroethylene

In a cohort mortality study of 3,814 white male employees at a uranium processing plant, Ritz (1999) reported that the main exposures (classified into "light," "moderate," and "heavy;" actual exposure concentrations not reported) were to kerosene, trichloroethylene, and cutting fluids (complex mixtures of variable composition classified as straight oils, soluble, or synthetic fluids; no information was available regarding the specific cutting oils used at the plant being studied over the 30-year exposure period). Considerable overlap in exposures occurred between these three substances, though primarily only at the "light" exposure level. Moderate exposure to trichloroethylene for 5 or more years was associated with increased incidence of liver (relative risk [RR] 12.1, 95% confidence interval [CI] 1.03–144) and brain (RR 14.4, 95% CI 1.24–167) cancer, though these increases were each the result of a single case. Both light (RR 3.46, 95% CI 1.22–9.80) and moderate (RR 7.71, 95% CI 2.04–29.1) exposures to kerosene (>2 years duration) were associated with increases in cancers of the esophagus and stomach. However, no inferences as to potential joint toxic actions can be made for trichloroethylene and kerosene from this study due to co-exposure to other chemicals (i.e., cutting fluids).

Spirtas et al. (1991) reported on the mortality of a cohort of 14,457 workers at an aircraft maintenance facility. The primary exposures were to trichloroethylene, though co-exposure to a number of chemicals, including JP-4, also occurred, as reported in a subsequent exposure assessment (Stewart et al. 1991). A significant trend toward increased incidence of emphysema with increasing trichloroethylene exposure was noted in male workers. While increases in cohort cancer mortality were observed, neither trichloroethylene nor JP-4 exposure was associated with significant increases in mortality from any type of cancer examined.

No other studies were located regarding possible joint toxic actions between jet fuels and trichloroethylene in affecting health-related endpoints in humans or research animals. No PBPK models for co-exposure to jet fuels and trichloroethylene were found. Jet fuels and trichloroethylene both produce neurological, hepatic, and immunological effects. Both jet fuels and trichloroethylene are believed to inhibit neuronal function by their physical presence in neuronal membranes, and as such, are expected to produce additive effects on the central nervous system. However, data directly corroborating this are not available. Both jet fuels and trichloroethylene are believed to elicit hepatic and immunological effects as a result of metabolism to reactive products, possibly involving reactive oxidative species. However, understanding of these mechanisms is insufficient to reliably predict the result, which might involve competitive inhibition and induction of various cytochrome P-450 isozymes, of joint exposure. Trichloroethylene is a probable human carcinogen (see Appendix C). No mechanistic information as to

potential effects of jet fuels, which have not been demonstrated to be carcinogenic, on the carcinogenic effects of trichloroethylene was located.

2.2.3 Hydrazines and Trichloroethylene

No studies were located regarding possible joint toxic actions between hydrazines and trichloroethylene in affecting health-related endpoints in humans or research animals. No PBPK models for co-exposure to hydrazines and trichloroethylene were found. Important targets of toxicity common to hydrazines and trichloroethylene are the liver and central nervous system. Both hydrazines and trichloroethylene have been shown to cause hepatic effects, including inflammation, fatty degeneration, and necrosis (see Appendices B and C). Both are believed to do so as a result of metabolism resulting in a reactive intermediate, possibly resulting in oxygen radical formation, although hydrazines can also act by direct binding of the parent compound to cellular macromolecules. Mechanistic understanding of the hepatic effects of hydrazines and trichloroethylene is not sufficient to make reliable predictions as to the hepatic effects of joint exposure. Both hydrazines and trichloroethylene have been shown to cause neurological effects. However, the mechanisms for these effects appear to differ for the two classes of compounds, with hydrazines believed to interact with alpha-keto acids, such as vitamin B6, whereas trichloroethylene is thought to interact directly with neuronal membranes (see Appendices B and C). Understanding of these mechanisms is inadequate to make reliable predictions as to the neurological effects of joint exposure. The carcinogenic effects of hydrazines and trichloroethylene are well documented (see Appendices B and C). Limited understanding of the mechanisms of hydrazine carcinogenesis, as well as limited knowledge of the mechanisms of action of trichloroethylene, precludes a reliable prediction of the carcinogenic effects of joint exposure.

2.2.4 Jet Fuels and Arsenic

No studies were located regarding possible joint toxic actions between jet fuels and arsenic in affecting health-related endpoints in humans or research animals. No PBPK models for co-exposure to jet fuels and arsenic were found. Studies examining both jet fuels and arsenic have reported neurological effects. However, the mechanisms behind arsenic-induced neurological effects are not well understood. Thus, no reliable predictions of the neurological effects of joint exposure can be made. Similarly, understanding of the mechanisms of arsenic and jet fuel-induced effects on the immune system is inadequate to assess the potential effects of joint exposure on immunotoxicity. Other sensitive endpoints of arsenic toxicity (e.g., dermal, cardiovascular, hematological, and renal effects) are not believed to be sensitive endpoints of jet fuel exposure, and mechanistic understanding is insufficient to allow for reliable predictions of the effect of co-exposure on these endpoints. Arsenic is a confirmed human carcinogen (see Appendix D). However, the mechanisms of arsenic carcinogenesis are not sufficiently understood to allow for reliable predictions of the effect of exposure to jet fuels on arsenic-induced carcinogenesis.

2.2.5 Hydrazines and Arsenic

Yamamoto et al. (1995) treated groups of male F344 rats to multiple initiators, followed by 26 weeks of exposure to dimethylarsinic acid. Animals received a single intraperitoneal injection of 100 mg/kg of diethylnitrosamine on day 0 of the experiment, then intraperitoneal injections of 20 mg/kg of N-methyl-N-nitrosourea on days 5, 8, 11, and 14, followed by subcutaneous injections of 40 mg/kg of 1,2-dimethyl-hydrazine on days 18, 22, 26, and 30. Two groups received no initiating treatments. Beginning at week 6, initiated animals then received 0, 50, 100, 200, or 400 ppm of dimethylarsinic acid in the drinking water (0, 27.1, 54.3, 108.6, or 217.1 mg As/kg/day); animals with no initiation treatments received 100 or 400 ppm (108.6 or 217 mg As/kg/day). Animals were sacrificed at 30 weeks and examined for histologic changes, including examination of glutathione S-transferase-placental (GST-P)-positive foci in the liver. Dimethylarsinic acid treatment resulted in significantly decreased body weights at concentrations of 100 ppm or greater. In initiated groups, dimethylarsinic acid treatment resulted in dose-dependent increases in the incidence of tumors of the liver, bladder, kidneys, and thyroid gland; preneoplastic lesions in the liver (GST-P-positive foci) and kidney (atypical tubules) were also increased. No tumors or preneoplastic lesions were observed in uninitiated animals. This study suggests that dimethylarsinic acid, which is a major metabolite of arsenic in mammals, can promote tumors initiated by a combination of several chemicals, one of which was 1,2-dimethylhydrazine (a liver carcinogen and, although not used as a rocket fuel, is structurally similar to the hydrazines that have been used for that purpose). However, data from this study were inadequate to assess whether any joint toxic action specifically between dimethylarsinic acid and 1,2-dimethylhydrazine was additive or greater than additive.

No other studies were located regarding possible joint toxic actions between hydrazines and arsenic in affecting health-related endpoints in humans or research animals. No PBPK models for co-exposure to hydrazines and arsenic were found. Shared targets of toxicity of hydrazines and arsenic include the hematopoietic and neurological systems (see Appendices B and D). However, understanding of the mechanisms of arsenic-induced toxic effects is insufficient to allow for reliable predictions of the effects of joint exposure. The carcinogenic effects of both hydrazines and arsenic are well documented (see Appendices B and D). Arsenic is a known human carcinogen. As with toxicity endpoints, the mechanisms of action are not sufficiently understood to allow for reliable predictions of the carcinogenic effect of joint exposure.

2.2.6 Trichloroethylene and Arsenic

Constan et al. (1995, 1996) exposed groups (5/time interval) of rats to a mixture of 31 ppm arsenic (as arsenic trioxide), 50 ppm benzene, 15 ppm chloroform, 7 ppm chromium (as chromium chloride hexa-hydrate), 37 ppm lead (as lead acetate trihydrate), 34 ppm phenol, and 38 ppm trichloroethylene in drinking water for up to 6 months; control animals received untreated drinking water. No changes in

weight gain, body weight, liver weight, or liver-associated plasma enzymes were reported. The authors noted an increase in hepatocellular proliferation, as measured by increased bromo-deoxyuridine (BrdU) staining, that was seen around the large hepatic veins on days 3 and 10, and at 1 month of exposure. Similarly, at day 10 and 1 month of exposure, apoptosis of hepatocytes, assessed by TdT-mediated dUTP digoxigenin nick end labeling (TUNEL) stain, was elevated in large hepatic veins. Neither proliferation nor apoptosis were significantly different from controls at 3 and 6 months of treatment.

In a follow-up study, Benjamin et al. (1999) pretreated groups of rats with an intraperitoneal injection of 20 mg/kg of diethylnitrosamine on day 0, then exposed them to the same mixture (referred to as 10x), or the mixture at 1/10th the concentration (3.1 ppm arsenic [as arsenic trioxide], 5.0 ppm benzene, 1.5 ppm chloroform, 0.7 ppm chromium [as chromium chloride hexahydrate], 3.7 ppm lead [as lead acetate trihydrate], 3.4 ppm phenol, and 3.8 ppm trichloroethylene; referred to as 1x) in drinking water for 21 or 56 days. Treatment at the 1x concentration resulted in a significant increase in the area, but not total number, of GST-P-positive (i.e., preneoplastic) foci in the liver relative to the deionized water controls. Treatment with the 10x concentration did not significantly affect either the number or area of the foci. The researchers concluded that there was no evidence of tumor promotion in this study.

Pott et al. (1998a) reported that oral administration of a mixture of arsenic, trichloroethylene, vinyl chloride, and 1,2-dichloroethane, after 2 weeks of initiation with diethylnitrosamine, in male F344 rats resulted in a dose-related decrease in the area of hepatocellular foci, as well as a decrease in the number of large foci per animal. No pulmonary adenomas were seen in any of the treated groups, while animals initiated with diethylnitrosamine averaged 0.25 adenomas per animal; the difference was statistically significant. The incidence of pulmonary hyperplasia was also significantly lower in treated groups compared to the initiation-only control group.

In a series of studies, Vodela et al. (1997a, 1997b) exposed male and female broiler chickens to drinking water containing mixtures of either 0.8 ppm arsenic, 1.3 ppm benzene, 5.0 ppm cadmium, 6.7 ppm lead, and 0.65 ppm trichloroethylene (low) or the same components at 10-fold higher concentrations (high). In the first experiment (Vodela et al. 1997a), male broiler chickens were exposed to the low or high concentrations of the mixture in the drinking water for 49 days. Exposed animals showed decreased water intake, food intake, and body weight gain, as well as statistically significant, dose-related decreases in cell-mediated and humoral immune response in both dose groups, relative to pair-watered controls. In the second experiment (Vodela et al. 1997b), female chickens were exposed to the low- or high-dose levels of the mixture from week 29 to 39 of age (10 weeks). Water consumption was significantly decreased in the high-dose animals, but not the low-dose animals; pair-watered controls were therefore used. Body weights were linearly ($p \leq 0.01$) decreased in exposed hens. Increasing concentration of the exposure mixture resulted in decreasing egg production and decreased egg weights, neither of which were due to reduced water consumption.

All of these studies were performed with mixtures that included other chemicals in addition to arsenic and trichloroethylene. It is uncertain which, if any, effects were influenced by these two chemicals, and what any joint toxic actions may have been. No other studies were located regarding possible joint toxic actions between arsenic and trichloroethylene in affecting health-related endpoints in humans or research animals. No PBPK models for co-exposure to arsenic and trichloroethylene were found. The most sensitive effects of trichloroethylene exposure are neurological effects, believed to result from an interaction between trichloroethylene and the neuronal membrane (see Appendix C). Although arsenic also produces neurological effects, the available data are not sufficient to reliably predict the neurological effect of joint exposure. Similarly, while both arsenic and trichloroethylene have been shown to affect the immune system and kidneys, data are inadequate to reliably predict the effect of joint exposure. Other sensitive endpoints of arsenic toxicity (e.g., dermal, cardiovascular, and hematological effects) are not believed to be sensitive endpoints of trichloroethylene exposure, and mechanistic understanding is insufficient to allow for reliable predictions of the effect of co-exposure to trichloroethylene on these endpoints. Trichloroethylene is a probable human carcinogen (see Appendix C) and arsenic is an established human carcinogen (see Appendix D), but due to limited understanding of the mechanism of action of either chemical, it is unknown what the carcinogenic effect of joint exposure might be.

2.2.7 Jet Fuels and Strontium-90

No studies were located regarding possible joint toxic actions between jet fuels and strontium-90 in affecting health-related endpoints in humans or research animals. No PBPK models for co-exposure to jet fuels and strontium-90 were found. Exposures to either jet fuels or strontium-90 have been shown to result in a decreased immune response in animal studies, but the effects for jet fuels are not well studied (see A and E). Understanding of the mechanism(s) of jet fuel-induced immunotoxic effects is not sufficient to allow for reliable mechanistic inferences as to possible joint action of jet fuels and strontium-90. Other effects of strontium-90 (musculoskeletal and hematological effects and cancer) have not been demonstrated as endpoints of jet fuel toxicity, and plausible modes of joint action on these strontium-90 targets are not obvious (see Appendices A and E). Other effects of jet fuels (neurological and hepatic effects, see Appendix A) are not believed to be sensitive targets of strontium-90 radiation (see Appendix E). No data were located to indicate how exposure to radiation from strontium-90 might influence neurological effects from jet fuels itself or hepatic effects involving metabolites of jet fuels.

2.2.8 Hydrazines and Strontium-90

No studies were located regarding possible joint toxic actions of hydrazines and strontium-90 in humans or research animals. No PBPK models for co-exposure to hydrazines and strontium-90 were found. Strontium-90 is believed to cause hematological and immunological effects by localizing in bone and/or lymphatic tissues and subsequently irradiating the progenitor cells (see Appendix E). The mechanisms of hydrazine-induced hematological effects are not known with certainty, but are believed to involve either

direct binding to cellular molecules, particularly alpha-keto acids, or the generation of reactive metabolites (see Appendix B). Available data are insufficient to allow for reliable predictions of hematological or immunological changes following joint exposure. The mechanism of carcinogenesis for strontium-90 (ionization events leading to damage to cellular constituents, including deoxyribonucleic acid [DNA]) is well characterized. Hydrazines are also genotoxic carcinogens; however, understanding of the mechanisms of action of hydrazines is not sufficient to allow for reliable prediction of the carcinogenic effect of joint exposure.

2.2.9 Trichloroethylene and Strontium-90

Kilburn (1999) reported on a cohort of 154 jet engine repair workers who were exposed to a variety of metals (strontium chromate, manganese, nickel, beryllium, and others) and solvents (trichloroethylene, 1,1,1-trichloroethane, trichlorofluoroethane, and methanol) and 112 controls. Reported exposure levels, measured in six workers on a single day, were 0.006–0.29 mg/m^3 for strontium chromate and 4,800 mg/m^3 for trichloroethylene. Exposed workers were found to have significant differences in a number of respiratory parameters, including shortness of breath, wheezing, phlegm, and abnormal radiographs, relative to controls. The researchers noted that such effects are consistent with industrial bronchitis due to inhalation of welding fumes and of particulates from grinding stainless steel. Exposed workers also showed significant impairment of a number of neurological indices, including simple and choice reaction times, sway speeds (eyes open and closed), and color discrimination. The researchers tentatively attributed these effects to chlorinated solvent exposure, although it was noted that some of the metals present (e.g., manganese) may also have contributed. Due to co-exposures to other chemicals and the fact that strontium was in the form of strontium chromate, with chemical toxicity generally believed to be due to the chromate group, potential joint toxic actions between strontium-90 and trichloroethylene cannot be assessed from this study.

No other studies were located regarding possible joint toxic actions between strontium-90 and trichloroethylene in affecting health-related endpoints in humans or research animals. No PBPK models for co-exposure to trichloroethylene and strontium-90 were found. Exposures to either strontium-90 or trichloroethylene have been shown to result in a decreased immune response in animal studies, but the effects for trichloroethylene are not well studied (see Appendices C and E). Understanding of the mechanism(s) of trichloroethylene-induced immunotoxic effects is not sufficient to allow for reliable mechanistic inferences as to possible joint action of trichloroethylene and strontium-90. Other effects of strontium-90 (musculoskeletal and hematological effects) have not been demonstrated as sensitive targets of trichloroethylene, and plausible modes of joint action on these strontium-90 targets are not obvious (see Appendices C and E). Other effects of trichloroethylene (neurological, hepatic, and renal effects, see Appendix C) are not believed to be sensitive targets of strontium-90 radiation (see Appendix E). No data were located to indicate how exposure to radiation from strontium-90 might influence neurological effects from trichloroethylene itself or hepatic and/or renal effects involving metabolites of trichloroethylene.

The mechanism of carcinogenesis for strontium-90 (ionization events leading to cellular damage, including DNA) is well characterized. Trichloroethylene is also carcinogenic in some species; however, understanding of the mechanisms of action of trichloroethylene is not sufficient to allow for reliable prediction of the effect of trichloroethylene on strontium-90-induced carcinogenic effects.

2.2.10 Arsenic and Strontium-90

De Kimpe et al. (1999) examined the effect of a number of compounds, including stable strontium (as strontium nitrate), on the methylation of arsenic in freshly-isolated liver cytosol from adult male Flemish Giant rabbits. This species was chosen because previous *in vivo* studies by these researchers demonstrated inorganic arsenic metabolism very similar to humans in these rabbits. Over the tested range of 0.34–8.5 µM, strontium exposure resulted in a dose-dependent decrease in both the mono- and dimethylation of arsenic. Similar results were found for many other species of trace elements and anions, as well as some, but not all, chelating agents, organic methyltransferase inhibitors, and uremic toxins. In contrast, some trace elements acted as stimulating agents for methylation, most notably Zn^{2+}. The researchers suggested that inhibition of methylation by strontium and other divalent cations may result from competitive inhibition with the stimulatory divalent cation, zinc. The researchers suggested that the inhibitory effects of the chelating agents ethylenediaminetetraacetic acid (EDTA) and oxime indicate that zinc may be an essential co-factor for As(III) methylation. This study shows that strontium inhibits methylation of arsenic *in vitro*. Because methylation of inorganic arsenic is generally considered to be a detoxification reaction, it is plausible that strontium-90 will increase the toxic effects of arsenic. However, it is not clear that strontium would be present in the liver cell in sufficient quantities to have any effect in a complete organism (approximately 99% of the total body burden is contained in the skeleton, see Appendix E) and it must be noted that the methylation products of arsenic are not without toxic effects themselves (studies have shown effects on the respiratory tissues, gastrointestinal tract, liver, kidney, reproduction, development, and genetic material [ATSDR 2000] and there is some evidence that dimethylarsinic acid is a cancer promoter [Yamamoto et al. 1995]).

Liu et al. (1999) reported that addition of 75 mg/L of arsenic trioxide (As_2O_3) to the water of Wistar rats for 6 months resulted in significantly decreased levels of naturally-occurring strontium in the kidney, but not in the liver, compared with control rats. The effect in the kidney disappeared when the animals were co-treated with sodium fluoride along with the arsenic trioxide.

No other studies were located regarding possible joint toxic actions between arsenic and strontium-90 in affecting health-related endpoints in humans or research animals. No PBPK models for co-exposure to arsenic and strontium-90 were found. Strontium-90 is believed to cause hematological and immunological effects by localizing in bone and/or lymphatic tissues and subsequently irradiating the progenitor cells (see Appendix E). While arsenic is also capable of eliciting hematological and immunological effects (see Appendix D), the mechanisms by which it does so are not well understood. Therefore, no

reliable predictions as to the immunological and hematological effects of joint exposure to arsenic and strontium-90 can be made. The mechanism of carcinogenesis for strontium-90 (ionization events leading to damage to cellular constituents, including DNA) is well characterized. However, understanding of the mechanisms of action of arsenic is not sufficient to allow for reliable predictions of carcinogenic effects following joint exposure. Arsenic induces the metal-binding protein metallothionein in the liver, but binds to it with low affinity (see Appendix D). The extent to which strontium, which is sequestered in bone, might interact with metallothionein in the liver is unclear, as are any potential consequences for strontium-90 toxicity, which is focused on the bone and surrounding tissues (see Appendix E).

2.3 Relevance of the Joint Toxic Action Data and Approaches to Public Health

Due to the lack of data regarding toxicity of the mixture of jet fuels, hydrazines, trichloroethylene, arsenic, and strontium-90, a component-based approach is recommended to assess potential public health effects associated with exposure to this mixture. PBPK/PD models to predict dispositional and toxicological outcomes of joint action of these five components are not available, but the WOE approach can be used to evaluate the joint toxic action of the component pairs (ATSDR 2001a, 2001b).

The weight-of-evidence approach produces a qualitative binary weight-of-evidence (BINWOE) classification and associated score for the effect of each substance in the mixture on each other substance in the mixture. BINWOEs are based primarily on pairwise data regarding joint toxic action, but can also include inferences based on mechanistic understanding of the disposition and toxicity of the individual substances. Figure 1 shows the factors that contribute to a BINWOE classification and the associated scoring.

BINWOEs for the mixture of jet fuels, hydrazines, trichloroethylene, arsenic, and strontium-90 are shown in Tables 4–7. The selection of target organs or endpoints for BINWOE development takes into account the critical effects of the individual components. In addition, and particularly if the components do not have the same critical effect, the selection also takes into account other relatively sensitive effects in common across two or more components of the mixture. See Section 1 and Appendices A–E for information on the critical and other sensitive endpoints of the individual mixture components. The BINWOEs focus on repeated simultaneous exposure, since this is the exposure scenario most relevant to evaluation of public health risk associated with exposure to these substances at a waste site.

Due to the scarcity of data available regarding joint toxic action of the component pairs for the jet fuels, hydrazines, trichloroethylene, arsenic, and strontium-90 mixture, and insufficient understanding of toxic and pharmacokinetic mechanisms of the individual substances, the type of joint toxic action could not be predicted for 17 of the 20 BINWOEs for this mixture. The only joint action that could be projected was

for additive depression of the central nervous system from exposure to jet fuels and trichloroethylene (see Tables 4 and 5) and a greater-than-additive effect of strontium-90 on the general toxicity of arsenic by inhibition of methylation of the arsenic (see Table 6).

Although the BINWOEs were indeterminate for all of the remaining pairs due to insufficient data (see Table 7), the extensive overlap of toxic endpoints for the five mixture components suggests that there is a potential for joint toxic action among these substances. For example, similar effects on the liver are produced by jet fuels, hydrazines, and trichloroethylene. The possibility of joint toxic action on the central nervous system by jet fuels and trichloroethylene was recognized in the BINWOEs; in addition, the central nervous system is also affected by hydrazines and arsenic (although the peripheral nerves are a more sensitive target for this chemical). Immunosuppression is characteristic of four of the five mixture components (all but hydrazines, for which there is also some evidence of immune sensitivity), and similar hematological effects are produced by arsenic, strontium-90, and hydrazines. Renal effects, which are well-known for trichloroethylene, are also produced by arsenic. Hydrazines and strontium-90 are genotoxic carcinogens, and trichloroethylene and arsenic are also carcinogens, although the mechanisms of carcinogenicity for the latter two chemicals have not been fully elucidated.

Given this amount of overlap in toxic endpoints, it is reasonable to be cautious when evaluating public health concerns for this mixture by assuming additivity (dose additivity for noncancer effects and response additivity for cancer, as per ATSDR 2001a, 2001b).

Figure 1. Binary Weight-of-Evidence Scheme for the Assessment of Chemical Interactions*

	Classification	Factor
Direction of Interaction		**Direction**
=	Additive	0
>	Greater than additive	+1
<	Less than additive	–1
?	Indeterminate	0
Quality of the Data		**Weighting**
Mechanistic Understanding		
I.	Direct and Unambiguous Mechanistic Data: The mechanism(s) by which the interactions could occur has been well characterized and leads to an unambiguous interpretation of the direction of the interaction.	1.0
II.	Mechanistic Data on Related Compounds: The mechanism(s) by which the interactions could occur has not been well characterized for the chemicals of concern but structure-activity relationships, either quantitative or informal, can be used to infer the likely mechanisms(s) and the direction of the interaction.	0.71
III.	Inadequate or Ambiguous Mechanistic Data: The mechanism(s) by which the interactions could occur has not been well characterized or information on the mechanism(s) does not clearly indicate the direction that the interaction will have.	0.32
Toxicological Significance		
A.	The toxicological significance of the interaction has been directly demonstrated.	1.0
B.	The toxicological significance of the interaction can be inferred or has been demonstrated for related chemicals.	0.71
C.	The toxicological significance of the interaction is unclear.	0.32
Modifiers		
1.	Anticipated exposure duration and sequence.	1.0
2.	Different exposure duration or sequence.	0.79
a.	*In vivo* data	1.0
b.	*In vitro* data	0.79
i.	Anticipated route of exposure	1.0
ii.	Different route of exposure	0.79

Weighting Factor = Product of Weighting Scores: Maximum = 1.0, Minimum = 0.05

BINWOE = Direction Factor x Weighting Factor: Ranges from –1 through 0 to +1

*Source: ATSDR 2001a, 2001b

Table 4. Effect of **Jet Fuels** on **Trichloroethylene**

BINWOE: =IIIC (0)
neurological effects
BINWOE: ? (0)
hepatic effects
BINWOE: ? (0)
immunological effects

Direction of Interaction - Jet fuels and trichloroethylene are expected to produce additive effects on neurological endpoints. The direction of the interaction for hepatic and immunological effects cannot be predicted in the absence of (1) pertinent interaction data; (2) information clearly indicating that pharmacokinetic interactions with jet fuels will influence the toxicity of trichloroethylene; or (3) mechanistic understanding leading to an unambiguous projection of interactions between jet fuels and trichloroethylene.

Mechanistic Understanding - Jet fuels and trichloroethylene both produce neurological, hepatic, and immunological effects. Both jet fuels and trichloroethylene are believed to inhibit neuronal function by their physical presence in neuronal membranes, and as such, are expected to produce additive effects on the central nervous system. However, data directly corroborating this are not available; a rating of "III" was therefore assigned. Both jet fuels and trichloroethylene are believed to elicit hepatic and immunological effects as a result of metabolism to reactive products, possibly involving reactive oxidative species. However, understanding of these mechanisms is insufficient to reliably predict the influence, which might involve competitive inhibition and induction of various cytochrome P-450 isozymes, of exposure to jet fuels on the hepatic or immunological effects of trichloroethylene. Trichloroethylene is a probable human carcinogen (see Appendix C). No mechanistic information as to potential effects of jet fuels, which have not been demonstrated to be carcinogenic, on the carcinogenic effects of trichloroethylene was located.

Toxicologic Significance - The BINWOE for neurological effects contains a rating of "C" for toxicological significance because neither the joint toxic action nor any potentially related mechanistic changes have been demonstrated. Two cohort mortality studies (Ritz 1999; Spirtas et al. 1991) involving co-exposure to jet fuels and trichloroethylene have been reported. However, in both cases, high levels of co-exposure to other chemicals prevents a reliable determination of the potential joint toxic action of jet fuels and trichloroethylene. No other relevant interaction data on health effects following simultaneous exposure were located. No studies were located in which pretreatment with jet fuels prior to trichloroethylene exposure was examined.

Additional Uncertainties - Uncertainties have been addressed in the above discussion.

Table 5. Effect of **Trichloroethylene** on **Jet Fuels**

BINWOE: =IIIC (0)
neurological effects
BINWOE: ? (0)
hepatic effects
BINWOE: ? (0)
immunological effects

Direction of Interaction - Jet fuels and trichloroethylene are expected to produce additive effects on neurological endpoints. The direction of the interaction for hepatic and immunological effects cannot be predicted in the absence of (1) pertinent interaction data; (2) information clearly indicating that pharmacokinetic interactions with trichloroethylene will influence the toxicity of jet fuels; or (3) mechanistic understanding leading to an unambiguous projection of interactions between trichloroethylene and jet fuels.

Mechanistic Understanding - Jet fuels and trichloroethylene both produce neurological, hepatic, and immunological effects. Both jet fuels and trichloroethylene are believed to inhibit neuronal function by their physical presence in neuronal membranes, and as such, are expected to produce additive effects on the central nervous system. However, data directly corroborating this are not available; a rating of "III" was therefore assigned. Both trichloroethylene and jet fuels are believed to elicit hepatic and immunological effects as a result of metabolism to reactive products, possibly involving reactive oxidative species. However, understanding of these mechanisms is insufficient to reliably predict the influence, which might involve competitive inhibition and induction of various cytochrome P-450 isozymes, of exposure to trichloroethylene on the hepatic or immunological effects of jet fuels.

Toxicologic Significance - The BINWOE for neurological effects contains a rating of "C" for toxicological significance because neither the joint toxic action nor any potentially related mechanistic changes have been demonstrated. Two cohort mortality studies (Ritz 1999; Spirtas et al. 1991) involving co-exposure to trichloroethylene and jet fuels have been reported. However, in both cases, high levels of co-exposure to other chemicals prevents a reliable determination of the potential joint toxic action of trichloroethylene and jet fuels. No other relevant interaction data on health effects following simultaneous exposure were located. No studies were located in which pretreatment with trichloroethylene prior to jet fuel exposure was examined.

Additional Uncertainties - Uncertainties have been addressed in the above discussion.

Table 6. Effect of **Strontium-90** on **Arsenic**

BINWOE: >IIICb (+1 x 0.32 x 0.32 x 0.79 = +0.08)

Direction of Interaction - It is plausible that strontium-90 will increase the toxic effects of arsenic (>). There is evidence that strontium inhibits methylation of arsenic *in vitro* (De Kimpe et al. 1999). Because methylation of inorganic arsenic is generally considered to be a detoxification reaction (see caveat below), inhibition of methylation may reasonably be expected to produce a general increase in arsenic toxicity at targets throughout the body.

Mechanistic Understanding - Mechanistic understanding of the effect of strontium on arsenic is limited (III). Relevant data were from a single *in vitro* study (De Kimpe et al. 1999). The study was conducted in freshly-isolated liver cytosol from adult male Flemish Giant rabbits. This species has been shown to be an appropriate model for metabolism of arsenic in humans, and the liver is the primary site of arsenic methylation. While strontium was found to inhibit methylation of arsenic in this test system, so were many other inorganic ions and organic compounds. It was found that certain inorganic cations (most notably Zn^{2+}) stimulated methylation. The researchers presented some evidence to suggest that zinc may be an essential co-factor for As(III) methylation, and hypothesized that competitive inhibition between strontium (or other divalent cations) and zinc could be responsible for the observed inhibition of methylation in this test system. However, it is not clear that strontium would be present in the liver cell in sufficient quantities to have any effect in a complete organism. Although low concentrations of strontium can be found in soft tissues, approximately 99% of the total body burden is contained in the skeleton (see Appendix E). No studies have been done to investigate whether strontium would inhibit arsenic methylation in a whole animal model.

Toxicologic Significance - The toxicological significance of the interaction is not clear (C). Methylation of arsenic is generally considered a detoxification reaction because the methylation products, monomethylarsonic acid (MMA) and dimethylarsinic acid (DMA), are less acutely toxic than inorganic arsenic, have a lower affinity for tissue constituents and proteins, and are excreted more rapidly (De Kimpe et al. 1999). However, MMA and DMA are not without toxic effects themselves. Studies of MMA and DMA have shown effects on the respiratory tissues, gastrointestinal tract, liver, kidney, reproduction, development, and genetic material (ATSDR 2000). There is some evidence that DMA is a cancer promoter (Yamamoto et al. 1995).

Modifying Factors - The only data available regarding the effect of strontium on arsenic toxicity are from an *in vitro* test system that may not be representative of *in vivo* exposure (b).

Additional Uncertainties - Uncertainties have been addressed in the above discussion.

Table 7. Matrix of BINWOE Determinations for Simultaneous Exposure to Chemicals of Concern

		ON TOXICITY OF				
		Jet fuels	Hydrazines	Trichloroethylene	Arsenic	Strontium-90
EFFECT OF	Jet fuels		? (0)	**=IIIC (0)**[a] ? (0)[b]	? (0)	? (0)
	Hydrazines	? (0)		? (0)	? (0)	? (0)
	Trichloroethylene	**=IIIC (0)**[a] ? (0)[b]	? (0)		? (0)	? (0)
	Arsenic	? (0)	? (0)	? (0)		? (0)
	Strontium-90	? (0)	? (0)	? (0)	**>IIICb (+0.08)**	

[a] Neurological effects
[b] Effects on targets other than the nervous system

2.4 Recommendations for Data Needs

Neither *in vivo* data from human or animal studies nor *in vitro* data examining the toxicity of the 5-component mixture, or for 4- or 3-component submixtures, are available. Similarly, PBPK models describing the behavior of the 5-component mixture, or for 4- or 3-component submixtures, are not available. In the absence of direct interaction data, a component-based approach was utilized. However, data on the joint toxic action of the component pairs of the mixture are lacking, with no adequate joint action toxicity data available for any of the 10 component pairs of the mixture. Data on the potential mechanistic interactions between the component pairs are also scarce.

For the individual components, oral MRL/RfDs are available only for arsenic and trichloroethylene. Jet fuels, hydrazines, and strontium-90 are all known to produce noncancer effects by oral exposure, so the lack of oral health guidance values for these materials is problematic. Inhalation MRL/RfCs are available for all three of the chemicals in the mixture for which this route of exposure is expected to potentially contribute to exposure of offsite receptors at rocket launch sites (jet fuels, hydrazines, and trichloro-ethylene), although a chronic value is not available for hydrazines. Oral slope factors and inhalation unit risks are available for all of the mixture components, except jet fuels, for which there is no evidence of carcinogenicity.

3. Recommendation for Exposure-Based Assessment of Joint Toxic Action of the Mixture

As discussed by ATSDR (1992, 2001a), exposure-based health assessments are used, in conjunction with evaluation of community-specific health outcome data, consideration of community health concerns, and biomedical judgement, to assess the degree of public health hazard presented by mixtures of hazardous substances released into the environment.

Due to the lack of data regarding toxicity of the mixture of jet fuels, hydrazines, trichloroethylene, arsenic, and strontium-90, a component-based approach is recommended to assess potential public health effects associated with exposure to this mixture. Because of the extensive overlap of toxic endpoints for the five components of this mixture, the specific recommendation for this mixture is to assume additivity among the mixture components. BINWOE analysis of the joint toxic action of the component pairs was indeterminate for most pairs due to scarcity of data available regarding joint toxic action of the component pairs and insufficient understanding of toxic and pharmacokinetic mechanisms of the individual substances, but did support the assumption of additivity for depression of the central nervous system from exposure to jet fuels and trichloroethylene. Greater-than-additive effects were predicted for the effect of strontium-90 on general arsenic toxicity, due to inhibition of arsenic metabolic detoxification by strontium.

The hazard index is a component-based approach that assumes additivity for noncancer effects (ATSDR 2001a). In this approach, the ratio of exposure level to health guidance value (hazard quotient) for each substance affecting a particular endpoint is summed to provide a measure of hazard for the whole mixture. For cancer effects, the cancer risk for each substance (calculated from the lifetime average daily intake and the potency factor) is summed to provide an estimate of risk due to the whole mixture (ATSDR 2001a). These approaches incorporate the assumptions of dose addition for noncancer effects and response addition for cancer.

Because it assumes dose addition, the hazard index is most appropriately applied to components that cause the same effect by the same mechanism of action. However, the method is frequently applied to components with the same critical target organ or critical effect (effect that is the basis for the MRL, RfD, or other health guideline), without regard to mechanism of action, and may take into consideration other sensitive targets beside the critical target. Use of the dose-additivity assumption is likely to produce estimates of health hazard that range from appropriate to somewhat conservative, and which are therefore protective of public health (ATSDR 2001a).

Specific recommendations for implementing these approaches for noncancer and cancer effects are presented in the *Guidance Manual for the Assessment of the Joint Toxic Action of Chemical Mixtures* (ATSDR 2001a). Figure 2 of the guidance document shows that hazard indexes are only calculated if two

or more of the individual components have hazard quotients equaling or exceeding 0.1. If only one or if none of the components has a hazard quotient that equals or exceeds 0.1, then no further assessment of the joint toxic action is needed, since additivity and/or interactions are unlikely to result in a significant health hazard. Similarly, Figure 3 of the guidance document shows that cancer risks are summed only if estimated risks exceed $1x10^{-6}$ for at least two components.

Suggestive evidence that exposure to the mixture may constitute a hazard is provided when the hazard index for a particular exposure scenario exceeds 1. Although there is no direct quantitative relationship between hazard index and risk, concern for the possibility of a health hazard increases with increasing value of the hazard index above 1. An important point to note for the mixture of jet fuels, hydrazines, trichloroethylene, arsenic, and strontium-90, where exposure to some components may be by multiple pathways, is that the route-specific hazard indexes for a given duration and endpoint (or cancer risks) can be summed to account for exposure by multiple pathways (e.g., inhalation hazard index + oral hazard index = overall hazard index).

Critical endpoints for the health guidance values available for jet fuels, hydrazines, trichloroethylene, arsenic, and strontium-90 are shown in Table 1 in the Introduction. Other sensitive endpoints for these five substances are shown in Table 3 of the Introduction. In the absence of oral MRLs or RfDs for jet fuels, hydrazines, and strontium-90, oral hazard quotients cannot be calculated for these chemicals. This leaves only arsenic and trichloroethylene contributing to the hazard index for oral exposure to the mixture. Although the critical endpoints for these chemicals differ (neurological and hepatic effects for trichloroethylene, and dermal and gastrointestinal effects for arsenic), Table 3 shows that neurological, renal, and immunological endpoints are sensitive targets for both chemicals. Because these chemicals affect many of the same endpoints, it is recommended to calculate hazard indexes for oral exposure using both chemicals (ATSDR 2001a). Noncancer health guidance values for oral exposure to this mixture are shown in Table 8.

Inhalation MRLs or RfCs are available for all three chemicals for which this route is expected to potentially contribute to exposure to offsite receptors at rocket launch sites: jet fuels and hydrazines based on liver effects, and trichloroethylene based on neurological effects. Table 3 shows that the central nervous system and the liver are sensitive targets for all three chemicals. The immune system is also a sensitive target for jet fuels and trichloroethylene, and based on limited evidence, may also be a target for hydrazines (see Appendix B). Therefore, it is recommended that inhalation hazard indexes be calculated using all three chemicals together. The relevant health guidance values are shown in Table 9.

The target organ toxicity dose (TTD) modification of the hazard index method (ATSDR 2001a, 2001b) is not currently recommended for this mixture, due to weakness of the data and expected limited utility of the results. Lack of health guidance values for oral exposure to jet fuels and hydrazines is a major problem. These substances are known to produce liver and central nervous system effects by oral

exposure, as well as inhalation exposure. However, the oral data are insufficient for dose-response assessment (see Appendices A and B). As a result, the hazard index recommended above for oral exposure may significantly under-represent the health hazard associated with oral exposure to the mixture, and especially with regard to potential hepatotoxicity. In light of this major uncertainty, there is little justification for fine-tuning the oral hazard index of arsenic and trichloroethylene by developing TTDs based on endpoints other than liver toxicity (the chronic RfD for trichloroethylene is already based on liver effects and the liver is not a sensitive target for arsenic). Because the oral hazard index and inhalation hazard index are combined into an overall hazard index, and the liver and central nervous system effects of jet fuels and hydrazines by the oral route are not being taken into account in the oral hazard index, it seems reasonable to compensate by employing the most health protective form of the inhalation hazard index, using MRLs/RfCs rather than TTDs.

Cancer assessments available for jet fuels, hydrazines, trichloroethylene, arsenic, and strontium-90 are shown in Table 2 in the Introduction. By inhalation exposure, only hydrazine and trichloroethylene contribute to cancer risk, but by oral exposure, hydrazine, trichloroethylene, arsenic, and strontium-90 all may contribute. The slope factors and unit risks for these substances are presented in Table 10.

Table 8. Noncancer Health Guidance Values for Oral Exposure to Chemicals of Concern (See Appendices A, B, C, D, and E for Details)

Duration	Chemical				
	Trichloroethylene (mg/kg/day)	Arsenic (mg/kg/day)	Jet fuels (mg/kg/day)	Hydrazines (mg/kg/day)	Strontium-90 (mg/kg/day)
Acute	0.2	0.005	—	—	—
Intermediate	—	—	—	—	—
Chronic	$2x10^{-4}$	$3x10^{-4}$	—	—	—

Table 9. Noncancer Health Guidance Values for Inhalation Exposure to Chemicals of Concern (See Appendices A, B, and C for Details)

Duration	Chemical		
	Jet fuels (mg/m^3)	Hydrazines (mg/m^3)	Trichloroethylene (mg/m^3)
Acute	—	—	10
Intermediate	3[a]	0.005[b] $5x10^{-4}$[c]	0.5
Chronic	0.3	—	0.04

[a]assessment for JP-5/JP-8 recommended because (1) kerosene-type JP-5/JP-8 more representative of jet fuels as a group than wide-cut JP-4, and (2) less uncertainty in this assessment than in that for kerosene
[b]hydrazine
[c]1,1-dimethylhydrazine; reasonable default value for other hydrazines

Table 10. Cancer Health Guidance Values for Oral or Inhalation Exposure to Chemicals of Concern (See Appendices B, C, D, and E for Details)

Exposure	Chemical			
	Trichloroethylene	Arsenic	Hydrazines	Strontium-90[a]
Non radiation				
Oral $(mg/kg/day)^{-1}$	0.4[b]	1.5	3.0[c]	—
Inhalation $(\mu g/m^3)^{-1}$	$5x10^{-6}$	$4.3x10^{-3}$	$4.9x10^{-3}$[c]	—
Radiation				
Oral $(pCi)^{-1}$	—	—	—	$5.59x10^{-11}$
Inhalation $(pCi)^{-1}$	—	—	—	$6.93x10^{-11}$

[a]and disintegration products
[b]high end of range of central risk estimates with lowest uncertainty
[c]based on hydrazine; reasonable default value for other hydrazines

4. Conclusions

Due to the lack of data regarding toxicity of the mixture of jet fuels, hydrazines, trichloroethylene, arsenic, and strontium-90, a component-based approach is recommended to assess potential public health effects associated with exposure to this mixture. Because of the extensive overlap of toxic endpoints for the five components of this mixture, the specific recommendation for this mixture is to assume additivity among the mixture components. BINWOE analysis of the joint toxic action of the component pairs was indeterminate for most pairs due to scarcity of data available regarding joint toxic action of the component pairs, and insufficient understanding of toxic and pharmacokinetic mechanisms of the individual substances, but did support the assumption of additivity for depression of the central nervous system from exposure to jet fuels and trichloroethylene. Greater-than-additive effects were predicted for the effect of strontium-90 on general arsenic toxicity, due to inhibition of arsenic metabolic detoxification by strontium.

The hazard index approach is recommended as an additive component-based method for assessing possible health hazards from noncancer effects for mixtures of jet fuels, hydrazines, trichloroethylene, arsenic, and strontium-90. The hazard index approach allows for summing across routes of exposure to account for multiple pathways of exposure, which may be important for this mixture. For oral exposure, the lack of health guidance values is problematic and leaves only arsenic and trichloroethylene contributing to the hazard index for oral exposure to the mixture. Because these chemicals affect many of the same sensitive endpoints (neurological, renal, and immunological targets), it is recommended to calculate hazard indexes for oral exposure using both chemicals. For inhalation exposure, intermediate MRLs are available for all three chemicals for which this route is expected to potentially contribute to exposure to offsite receptors at rocket launch sites: jet fuels and hydrazines based on liver effects and trichloroethylene based on neurological effects. Because the central nervous system and the liver are sensitive targets for all three chemicals, it is recommended that inhalation hazard indexes be calculated using all three chemicals together. Application of the TTD modification of the hazard index method is not justified by the existing data set.

For cancer effects, the cancer risk for each substance (calculated from the lifetime average daily intake and the potency factor) is summed to provide an estimate of risk due to the whole mixture. Risk can be summed across routes to account for multiple pathways of exposure.

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Appendix A: Background Information for Jet Fuels

Jet fuels are complex mixtures of hydrocarbons produced by distillation of petroleum crude oil. Most jet fuels (e.g., JP-5, JP-7, JP-8) are middle distillates similar in composition to kerosene and containing primarily C_9–C_{16} hydrocarbons (approximately 80% aliphatic and 20% aromatic). JP-4 is a "wide-cut" fuel that is a blend of kerosene with lower boiling naphtha streams, like those used to produce gasoline, and therefore, containing a greater range of hydrocarbons (C_4–C_{16}). Although the composition of JP-4 is notably different from the kerosene-type fuels, and minor differences exist also among the latter, obvious differences in toxicity have not been reported in the published literature. Therefore, these fuels are discussed together below.

A.1 Toxicokinetics

Absorption following inhalation exposure to jet fuels and related substances can be inferred from the occurrence of systemic health effects in humans and research animals exposed by inhalation (ATSDR 1995a, 1995b, 1998). In addition, studies have demonstrated that a mixture of aliphatic hydrocarbons mostly in the C_{10}–C_{12} range (white spirit) was readily absorbed through the lungs in humans, that individual alkanes and cycloalkanes in the range of C_6–C_{10} were absorbed in rats exposed by inhalation, and that isopropylbenzene (cumene), which is representative of the aromatic C_9–C_{16} fraction, had a pulmonary retention percentage of approximately 50% in human volunteers (ATSDR 1999). Data on oral absorption of jet fuels and related substances are limited, but suggest that these substances are absorbed from the gastrointestinal tract (ATSDR 1995a, 1995b, 1998). Gastrointestinal absorption of aliphatic hydrocarbons is inversely proportional to length of the carbon chain; absorption is approximately 60% for C_{14} hydrocarbons (ATSDR 1999). Aromatic hydrocarbons in this size range are well absorbed from the gut when administered at low doses: 80–90% of ingested 2-methylnaphthalene and isopropylbenzene was recovered in the urine (ATSDR 1999). Oral exposure can also result in the fuels being aspirated into the lungs, leading to respiratory effects (ATSDR 1995a, 1998). Systemic health effects have been reported following dermal application of JP-5, kerosene, and several aromatic compounds of the appropriate size (isopropylbenzene, naphthalene, monomethylnaphthalenes), indicating that these substances are absorbed through the skin (ATSDR 1995a, 1995b, 1998, 1999). McDougal et al. (2000) studied skin absorption and penetration of JP-8 and its components in an *in vitro* system using rat skin. These researchers found that total flux of hydrocarbons across the skin was a relatively slow 20.3 $\mu g/cm^2$/hour. A total of 13 individual components were found to penetrate the skin, with fluxes ranging from a high of 51.5 $\mu g/cm^2$/hour for diethylene glycol monomethyl ether (an additive) to 0.334 $\mu g/cm^2$/hour for tridecane. In general, aromatic compounds penetrated skin more rapidly than aliphatics. Six compounds, all aliphatic, were absorbed into the skin; concentrations ranged from 0.055 µg/g skin (tetradecane) to 0.266 µg/g skin (undecane) after 3.5 hours.

Limited data suggest that jet fuels and related substances are widely distributed throughout the body after being absorbed (ATSDR 1995a, 1995b, 1998, 1999). Studies with white spirit (C_{10}–C_{12} aliphatic) and individual aliphatic hydrocarbons in the C_6–C_{10} range showed that these chemicals can accumulate in fat (ATSDR 1999). Following gastrointestinal absorption, the larger molecular weight aliphatics are transported primarily by the lymphatic system, while the smaller ones are transported by both the lymph and the blood. There is no information available on metabolism of jet fuels and related substances (ATSDR 1995a, 1995b, 1998), but data on C_9–C_{16} hydrocarbons suggest that metabolism of aliphatics in this range (primarily cytochrome P-450 mediated oxidation to fatty acids and alcohols) is slow, while the aromatics are metabolized faster (oxidation of alkyl site and/or ring, sometimes with formation of reactive intermediates, and conjugation with glutathione, glucuronic acid, or glycine) (ATSDR 1999). Data on elimination of jet fuels and related substances are not available (ATSDR 1995a, 1995b, 1998). It is noteworthy, however, that white spirit (C_{10}–C_{12} aliphatic) is only slowly eliminated from the fat, while aromatics in this size range are excreted rapidly as metabolites in the urine (ATSDR 1999).

A.2 Health Effects

Jet fuels can produce central nervous system impairment in humans by all routes of exposure, characterized by effects such as fatigue, coordination and concentration difficulties, headache, intoxication, anorexia, depressed mood, lack of initiative, dizziness, sleep disturbances, changes in posture, and reduced sensorimotor speed (ATSDR 1995b, 1998, 1999). Unconsciousness, coma, and convulsions have been observed after ingestion of kerosene by children. Similar symptoms of central nervous system depression have been observed in animal studies. Jet fuels and related substances can also produce respiratory, gastrointestinal, dermal, and ocular irritation in humans and animals (ATSDR 1995b, 1998, 1999). Respiratory effects may occur as a result of inhalation of jet fuel vapor, but in humans, have been more commonly and severely associated with aspiration into the lungs following oral exposure. Gastrointestinal effects have been noted after both inhalation and oral exposure. Dermal effects are usually a result of direct skin contact with the fuel, but have also been reported after oral exposure. Eye irritation has been reported as a consequence of exposure to jet fuel vapor in humans, although studies of direct ocular contact in animals have been negative.

Animal studies have also identified the liver, kidney, and immune system as targets for jet fuel toxicity. The liver is a sensitive and commonly affected endpoint in animal studies. MRLs for JP-4, JP-5, JP-7, and JP-8 are all based on liver effects (ATSDR 1995b, 1998). Observed effects in the liver include degenerative fatty change, hepatocellular necrosis, and hepatic inflammation. Hepatotoxicity was also indicated by increases in serum enzyme activities. In the kidney, animal studies have shown that jet fuels produce hyaline droplet nephropathy, which is unique to male rats and not predictive of renal effects in humans. Jet fuels and kerosine have also been found to produce immunosuppression (suppressed hyper-sensitivity reactions to antigens, suppressed ability of splenic T-cells to respond to mitogens, decreased number of viable immune cells, decreased immune organ weights) by inhalation and dermal exposure,

while also being weak dermal sensitizers themselves (ATSDR 1998; Stoica et al. 2001; Ullrich 1999). A developmental toxicity study of JP-8 found decreased fetal body weight associated with oral exposure during gestation, but only at doses that also produced significant decreases in maternal weight gain (ATSDR 1998). Data regarding reproductive toxicity are not available. Genotoxicity testing has been fairly extensive, and the results have been overwhelmingly negative (ATSDR 1995b, 1998). Skin painting studies with jet fuels and kerosene have produced evidence suggesting that chronic dermal application of these substances can produce skin tumors (ATSDR 1995b, 1998, 1999; Rosenthal et al. 2001). Dermal tumorigenesis or tumor promotion by these substances may be related to their ability to produce skin irritation and dermal cell toxicity (Rosenthal et al. 2001). Data regarding internal cancers in humans and animals are equivocal.

A.3 Mechanisms of Action

Central nervous system depression, as observed for jet fuels, is an effect common to many organic solvents. It is generally thought to occur when the lipophilic parent compound partitions into the nerve cell membranes and disrupts function of membrane proteins by disturbing their lipid environment or by directly altering protein conformation (ATSDR 1999). Oxidative metabolism of the parent compounds reduces their lipophilicity and counteracts their central nervous system depressive effects. The hydrocarbon parent compounds in jet fuels are also thought to be responsible for the respiratory irritation and pneumonitis that can result from inhalation or aspiration of these fuels. It has been hypothesized that the parent hydrocarbons interact with nerve cell membranes, resulting in bronchoconstriction, and dissolve into membranes of the lung parenchyma, resulting in hemorrhagic exudation of proteins, cells, and fibrin into the alveoli (ATSDR 1999). *In vitro* experiments have shown that JP-8 induces apoptotic cell death in rat lung epithelial cells, apparently by damaging mitochondria in the cells (Stoica et al. 2001). JP-8 also induced apoptosis in immune system cells (U-937 human monocytic cells, Jurkat T-cell leukemia cells, primary mouse thymocytes) *in vitro* (Stoica et al. 2001). In contrast, JP-8 produced necrotic cell death in primary and immortalized human keratinocytes and primary mouse skin fibroblasts in culture and when applied topically to immortalized human keratinocytes grafted onto nude mice (Rosenthal et al. 2001). While the central nervous system and irritant effects of jet fuels are apparently due to the parent hydrocarbons, effects on the liver and kidney are probably due to formation of reactive intermediates and metabolites during oxidative metabolism, and to subsequent binding of these reactive species to cellular macromolecules.

A.4 Health Guidelines

ATSDR (1995a) derived an intermediate-duration inhalation MRL of 9 mg/m^3 for JP-4 based on a lowest-observed-adverse-effect level (LOAEL) of 500 mg/m^3 for hepatotoxicity (hepatocellular fatty change) in female mice in a 90-day continuous exposure study, a human equivalent dose conversion factor of 5.7, and an uncertainty factor of 300 (10 for the use of a LOAEL, 3 for interspecies extrapolation, and 10 for

human variability). ATSDR (1995b) also derived a chronic inhalation MRL of 0.3 mg/m^3 for JP-7 based on a LOAEL of 150 mg/m^3 for hepatic inflammation in female mice exposed intermittently for 1 year ($LOAEL_{ADJ}$=26.8 mg/m^3), a human equivalent dose conversion factor of 3.3 (0.36 m^3/day/0.38 kg x 70 kg/20 m^3/day), and an uncertainty factor of 300 (10 for the use of a LOAEL, 3 for interspecies extrapolation, and 10 for human variability). ATSDR (1998) derived an intermediate inhalation MRL of 3 mg/m^3 for JP-5 and JP-8 based on a LOAEL of 150 mg/m^3 for hepato-cellular fatty change in mice exposed to JP-5 continuously for 90 days ($LOAEL_{HEC}$=150 mg/m^3 x 0.04 m^3/day/0.0246 kg x 70 kg/20 m^3/day=854 mg/m^3), and an uncertainty factor of 300 (10 for the use of a LOAEL, 3 for interspecies extrapolation, and 10 for human variability). ATSDR (1995a) derived an intermediate inhalation MRL for kerosene of 0.01 mg/m^3 based on decreased blood glucose levels (thought to be indicative of hepatic effects) in male rats intermittently exposed to 58 mg/m^3 for 14 weeks ($LOAEL_{ADJ}$=12.4 mg/m^3) and an uncertainty factor of 1,000 (10 for the use of a LOAEL, 10 for inter-species extrapolation, and 10 for human variability). ATSDR (1999) noted that the MRL for kerosene involves greater uncertainty regarding toxicological significance of the observed effect (decreased blood glucose) than the MRLs for JP-5, JP-8, and JP-7 (liver pathology), and chose the latter MRLs (and not the kerosene MRL) to be the appropriate surrogate values for the assessment of health effects due to exposure to the kerosene-like fraction (C_8–C_{16} aliphatics) of TPH (total petroleum hydrocarbons). Data on jet fuels and related substances were inadequate to support inhalation MRLs of other durations or oral MRLs. EPA does not list assessments for jet fuels or related substances on the Integrated Risk Information System (IRIS 2001) or in the Health Effects Assessment Summary Tables (HEAST) (EPA 1997). The International Agency for Research on Cancer (IARC 2001) placed jet fuels in cancer weight-of-evidence Group 3 (not classifiable as to human carcinogenicity). Jet fuels are not listed in National Toxicology Program's (NTP) 9th Report on Carcinogens (2001).

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Appendix B: Background Information for Hydrazine Compounds

The hydrazine compounds included in this Interaction Profile are hydrazine (diamine) and 1,1-dimethyl-hydrazine (unsymmetrical dimethylhydrazine), both of which have been used as rocket fuels. These chemicals are similar with regard to disposition in the body and health effects and will be discussed together below.

B.1 Toxicokinetics

Animal studies suggest that hydrazines are well absorbed following inhalation, oral, or dermal exposure, and are evenly distributed throughout the body without preferential accumulation in any specific tissues (ATSDR 1997a). Metabolism of hydrazines involves a number of enzymatic and non-enzymatic pathways, and differs somewhat for hydrazine and 1,1-dimethylhydrazine. *In vivo* studies in rats have shown that hydrazine undergoes acetylation and can react with cellular molecules. Metabolism of this compound is qualitatively similar by different routes of exposure. Observed metabolites include acetyl hydrazine, diacetyl hydrazine, pyruvate hydrazone, and urea in the urine, and nitrogen gas in the expired air. *In vitro* studies have shown that hydrazine is readily metabolized by cytochrome P-450 in rat liver and can also be a substrate for other enzyme systems (peroxidases) or nonenzymatic reactions (copper ion-mediated). Oxidative metabolism of hydrazine is accompanied by formation of free radicals, including acetyl, hydroxyl, and hydrogen radicals. The presence of acetyl radicals suggests that hydrazine is acetylated prior to radical formation. Metabolism of 1,1-dimethylhydrazine also results in generation of free radicals, but with this compound, methyl radicals are produced during oxidative demethylation (enzymatic or nonenzymatic) to formaldehyde. *In vivo* studies have found hydrazone derivatives of 1,1-dimethylhydrazine in the urine. Although both hydrazine compounds are readily metabolized, a fair amount of both is excreted unchanged in the urine. Elimination of metabolites and parent compound is rapid, with most of the absorbed dose being eliminated from the body within 24 hours.

B.2 Health Effects

The central nervous system is the most prominent target of hydrazines that has been identified in humans (ATSDR 1997a). Effects, which have been recorded after inhalation, oral, and dermal exposure, have included nausea, vomiting, dizziness, excitement, tremors, polyneuritis, impaired cognitive function, lethargy, narcosis, convulsions, and coma. Animal studies have confirmed that the central nervous system is an important target of hydrazine and 1,1-dimethylhydrazine. The effects that have been noted are similar to those observed in humans: behavioral changes, tremors, depression, lethargy, seizures, and convulsions. Very limited human data have also suggested that inhalation of hydrazines can affect the lungs (bronchitis, tracheitis, pneumonia, dyspnea, pulmonary edema), heart (atrial fibrillation, enlargement of the heart, degeneration of heart muscle fibers), liver (fatty degeneration, focal necrosis),

and kidney (tubular necrosis, hemorrhage, inflammation). Animal studies support these tissues as target organs for hydrazines. Hydrazine and 1,1-dimethylhydrazine have been reported to produce irritation, inflammation, hyperplasia, dysplasia, and cellular damage in the nasal mucosa and lungs of rodents by inhalation exposure. Studies have demonstrated multiple liver effects (hemosiderosis, degeneration, fatty change, elevated serum enzyme levels, hyperplasia, necrosis, hepatitis, fibrosis) due to hydrazine and 1,1-dimethylhydrazine in multiple species by inhalation, oral, and parenteral routes of exposure. Renal effects have also been observed in animal studies, although effects were mild in most cases. Injected hydrazine did produce more severe effects (nephritis) in studies in dogs and monkeys. Animal data on cardiovascular effects are inconsistent, but there are reports of angiectesis (dilated blood vessels) and altered blood pressure after exposure to 1,1-dimethylhydrazine and myocardial fat accumulation after injection with hydrazine.

Hydrazines can produce contact dermatitis in humans. Animal studies have also reported dermal and ocular irritant effects after direct contact. Animal studies have also shown that hydrazines can produce hematological effects (e.g., anemia) in dogs (but not in rodents or monkeys) and have presented limited evidence for effects on the immune system (decreased T-helper cells *in vivo*, immunomodulation in mouse splenocytes and lymphocytes *in vitro*), reproduction (ovarian and testicular atrophy, endometrial inflammation and cysts, aspermatogenesis, abnormal sperm), and development (reduced fetal body weights, perinatal mortality in one injection study but not in other studies). There is ample evidence that hydrazine and 1,1-dimethylhydrazine are genotoxic, producing methyl adducts in DNA and positive results in a series of assays for mutagenicity, micronucleus formation, sister chromatid exchange, unscheduled DNA synthesis, and cell transformation. Both hydrazine and 1,1-dimethylhydrazine are carcinogenic in rodents, producing multiple tumor types after inhalation, oral, and parenteral exposure.

B.3 Mechanisms of Action

Hydrazines may produce adverse effects by two different mechanisms (ATSDR 1997a). First, hydrazines that have a free amino group (including both hydrazine and 1,1-dimethylhydrazine) can bind directly to cellular molecules. For example, hydrazines can react with endogenous alpha-keto acids to form hydrazones. The consequences can be illustrated by the case of vitamin B6. Hydrazine and 1,1-dimethyl-hydrazine can form hydrazones with vitamin B6 derivatives, thereby inhibiting reactions that require vitamin B6 as a cofactor (e.g., transamination reactions, decarboxylation of amino acids, metabolism of lipids and nucleic acids, and glycogen phosphorylation) and inducing a functional deficiency of vitamin B6, which can lead to convulsions, anemia, and dermatitis. Convulsions and other neurological effects are known to be associated with exposure to hydrazines. Patients are commonly treated with a form of vitamin B6 (pyridoxine). Second, metabolism of hydrazines results in generation of reactive free radical intermediates. Binding of reactive intermediates may explain the genotoxic effects of hydrazines and may serve as the initiating event for cancers induced by hydrazines.

B.4 Health Guidelines

ATSDR (1997a) derived intermediate inhalation MRLs of 0.004 ppm (0.005 mg/m^3) for hydrazine and $2x10^{-4}$ ppm ($5x10^{-4}$ mg/m^3) for 1,1-dimethylhydrazine, based on LOAELs of 0.2 and 0.05 ppm, respectively, for liver effects in female mice exposed intermittently for 6 months (moderate fatty change for hydrazine, hyaline degeneration of the gall bladder for 1,1-dimethylhydrazine). Data were inadequate to support acute or chronic inhalation MRLs or oral MRLs. EPA has not derived RfD or RfC values for hydrazine or 1,1-dimethylhydrazine (EPA 1997; IRIS 2001). Both compounds are classified in IARC cancer Group 2B (possible human carcinogen) (IARC 2001) and listed as reasonably anticipated to be human carcinogens in NTP's 9th Report on Carcinogens (2001). Hydrazine is classified in EPA cancer Group B2 (probable human carcinogen) (IRIS 2001). EPA calculated for hydrazine an oral slope factor of 3.0 $(mg/kg\text{-}day)^{-1}$ based on hepatomas in male mice treated by gavage, and an inhalation unit risk of $4.9x10^{-3}$ $(\mu g/m^3)^{-1}$ based on nasal cavity adenomas or adenocarcinomas in male rats (IRIS 2001).

B.5 References

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Appendix C: Background Information for Trichloroethylene

C.1 Toxicokinetics

Trichloroethylene is rapidly and extensively absorbed following inhalation, oral, and dermal exposure (ATSDR 1997b; EPA 2001). Once absorbed, trichloroethylene is widely distributed to organs throughout the body (including the developing fetus) and, due to its lipophilic properties, accumulates in fat. Metabolism of trichloroethylene is extensive and occurs primarily in the liver, but also in the kidney, lungs, and other tissues. Biotransformation pathways in humans are thought to be qualitatively similar to those identified in animals. Two major pathways have been identified: (1) oxidation and (2) conjugation with glutathione. The initial, rate-limiting step in the oxidative pathway is oxidation by several isozymes of cytochrome P-450 to chloral hydrate. Chloral hydrate is then either oxidized to trichloroacetic acid via chloral hydrate dehydrogenase or reduced to trichloroethanol via alcohol dehydrogenase. Trichloro-ethanol undergoes conjugation with glucuronic acid to form trichloroethanol-glucuronide. The glucuronide can be eliminated in the urine or can be excreted to the bile and reabsorbed from the small intestine. This enterohepatic circulation is more prominent in humans than in rodents. Another metabolite that has been found in mice and humans is dichloroacetic acid, possibly formed by oxidation of trichloroacetic acid and/or trichloroethanol. The second pathway of trichloroethylene metabolism starts with glutathione conjugation in the liver to form S-(1,2-dichlorovinyl)glutathione, which is excreted in the bile and converted in the bile and intestines to S-(1,2-dichlorovinyl)-L-cysteine, which is reabsorbed by the body and concentrated in the kidney, where it can be detoxified by N-acetyltransferase and excreted in the urine or activated to a thioacetylating agent by β-lyase. This second pathway becomes especially important when high levels of trichloroethylene are present and the oxidative metabolism becomes saturated. Trichloroethylene is eliminated from the body predominately in the urine as metabolites (trichloroethanol, trichloroethanol-glucuronide, and trichloroacetic acid) and to a lesser degree in exhaled breath as the parent chemical or other volatile metabolites such as trichloroethanol and carbon dioxide.

Pharmacokinetic models have been developed for the disposition of trichloroethylene in mice, rats, and humans, including the prediction of target organ (e.g., liver, lung, brain, kidney) doses of biologically-active metabolites (Clewell et al. 2000; Fisher 2000; Fisher et al. 1998). These models have been used to investigate differences in metabolism among species. Such differences include higher peak blood levels of oxidative metabolites (e.g., trichloroacetic acid) in mice and rats than humans at equivalent doses, and longer duration of elevated blood levels in humans (ATSDR 1997b; EPA 2001). Species differences in enterohepatic circulation are thought to contribute to these differences. *In vitro* data suggest that the glutathione conjugation to form S-(1,2-dichlorovinyl)glutathione occurs more rapidly in mice than in rats or humans. However, it has not been established that subsequent steps in this pathway also occur more rapidly in mice.

C.2 Health Effects

Targets for trichloroethylene noncarcinogenic toxicity include the central nervous system (central nervous system depression, neurobehavioral deficits, hearing loss), liver (changes in serum cholesterol and bile acids, liver enlargement, cellular hypertrophy), kidneys (increased kidney weights, cytomegaly and karyomegaly in renal tubular epithelial cells), heart (decreased heart rate, cardiac arrhythmia), endocrine system (altered hormone levels), immune system (depressed immune function, autoimmune disease), male reproductive system (decreases in sperm count and motility), and developing fetus (cardiac and eye malformations, neurobehavioral alterations) (ATSDR 1997b; EPA 2001). The most sensitive endpoints following subchronic/chronic oral exposure were the liver, kidney, and developing fetus, with effects at doses down to 1–10 mg/kg/day. Following subchronic/chronic inhalation exposure, the most sensitive endpoints were the central nervous system, liver, and endocrine system, with effects at concentrations down to 1–100 ppm.

A recent article (Wartenberg et al. 2000) reviewed over 80 published papers and letters on the epidemiology of cancer in groups of people occupationally exposed to trichloroethylene. Based on analysis of the studies with the most rigorous exposure assessments, relative risks were elevated for kidney cancer (RR=1.7, 95% CI=1.1–2.7), liver cancer (RR=1.9, 95% CI=1.0–3.4), and non-Hodgkin's lymphoma (RR=1.5, 95% CI=0.9–2.3) in several cohorts of workers repeatedly exposed to high concentrations of trichloroethylene for years in workplace air. Workers in these studies, however, were also exposed to other solvents (e.g., tetrachloroethylene). Accurate adjustment for this and other confounding factors is not possible from the available data. Wartenberg et al. (2000) concluded that there is "moderate support" for a causative relationship between exposure to trichloroethylene and cancer using Hill's criteria of causation. Extensive testing in animals has shown mice developing liver and lung tumors and lymphomas, and rats developing kidney and testicular tumors (ATSDR 1997b; EPA 2001).

C.3 Mechanisms of Action

Nervous system effects from trichloroethylene, as for other lipophilic solvents, are thought to involve disruption of functions of neural membranes by the physical presence of the parent chemical in the neuronal membrane (ATSDR 1997b; EPA 2001). There is evidence to suggest that metabolites, such as trichloroethanol and dichloroacetic acid, may also contribute to the observed neurological effects. Trichloroethylene-induced cardiac arrhythmias are thought to involve parent-chemical sensitization of the heart to epinephrine-induced arrhythmias. In animals, chemicals that inhibited the metabolism of trichloroethylene increased the potency of trichloroethylene to induce cardiac arrhythmias, whereas chemicals enhancing trichloroethylene metabolism decreased its potency.

Carcinogenic and noncarcinogenic toxic effects of trichloroethylene in the liver and kidney are thought to be related to metabolism of the parent compound and production of reactive metabolites and

intermediates (ATSDR 1997b; EPA 2001; Goeptar et al. 1995). Reactive metabolites of the oxidative metabolic pathway that have been implicated in the production of liver effects include chloral hydrate, trichloroacetic acid, and dichloroacetic acid. Hypotheses that have been put forward for effects of these metabolites in the liver include peroxisome proliferation (oxidative damage caused by increases in free-radical generating enzymes and peroxisomal β-oxidation lead to tumor formation by an unknown mechanism; most closely associated with trichloroacetic acid; has not been observed in humans), responses mediated by the peroxisome proliferator-activated receptor (leads to promotion of gene transcription, including enzymes important in lipid metabolism; most closely associated with trichloro-acetic acid; qualitatively similar in humans and mice), disturbances in cell signaling (alterations in cell replication, selection, and apoptosis; affected in different ways by trichloroacetic acid and dichloroacetic acid), and effects on DNA (altered gene expression [e.g., hypomethylation of DNA leading to modified transcription of the gene] rather than induced mutation [trichloroethylene and its oxidative metabolites are weak genotoxicants]; associated with both trichloroacetic acid and dichloroacetic acid). EPA (2001) concluded that liver effects following trichloroethylene exposure may be due to both trichloroacetic acid and dichloroacetic acid acting by multiple modes of action.

Trichloroethylene-induced kidney damage has been proposed to involve conjugation products of trichloroethylene with glutathione. The conjugated products (e.g., dichlorovinyl-cysteine) can be hydrolyzed by β-lyase in the kidney, forming a reactive thiol group that can react with cellular macro-molecules and lead to cell damage. These cysteine intermediates have been shown to induce point mutations in bacteria. In support of this mechanistic hypothesis, chemical agents that inhibit β-lyase protected against dichlorovinyl-cysteine nephrotoxicity in rats.

Little is known about how trichloroethylene and/or its metabolites produce endocrine, immune, reproductive, and developmental effects, although some of the same mechanisms proposed for the liver, such as interference with cell signaling and activation of peroxisome proliferator-activated receptor, may be relevant to these other organ systems.

C.4 Health Guidelines

ATSDR (1997b) derived an acute inhalation MRL of 2 ppm (10 mg/m^3) for trichloroethylene based on a LOAEL of 200 ppm for subjective neurological symptoms such as fatigue and drowsiness in volunteers exposed 7 hours/day for 5 days. An uncertainty factor of 100 (10 for the use of a LOAEL and 10 to account for human variability) was used in the calculation. ATSDR (1997b) also derived an intermediate-duration inhalation MRL of 0.1 ppm (0.5 mg/m^3) for trichloroethylene based on a LOAEL of 50 ppm for decreased wakefulness during exposure, decreased postexposure heart rate, and slow-wave sleep in rats exposed for 8 hours/day, 5 days/week for 6 weeks. An uncertainty factor of 300 (10 for using a LOAEL, 3 for extrapolating from rats to humans, and 10 to account for human variability) was employed. ATSDR (1997b) did not derive a chronic inhalation MRL for trichloroethylene due to the lack of suitable data.

EPA (2001) derived a draft chronic inhalation RfC of 0.04 mg/m^3 for trichloroethylene based on central nervous system effects in two occupational studies with estimated exposure concentrations of 7 ppm (38 mg/m^3) and an uncertainty factor of 1,000 (10 for use of a LOAEL, 10 for subchronic exposure, and 10 for protection of sensitive individuals).

ATSDR (1997b) derived an acute oral MRL of 0.2 mg/kg/day for trichloroethylene based on a LOAEL of 50 mg/kg/day for neurobehavioral effects in mouse pups and an uncertainty factor of 300 (10 for the use of a LOAEL, 10 for extrapolating from animals to humans, and 3 for human variability [a full factor of 10 was not used because pups were taken to represent a sensitive population]). ATSDR (1997b) did not derive intermediate or chronic oral MRLs due to lack of appropriate data. EPA (2001) derived a draft chronic oral RfD of $2x10^{-4}$ mg/kg/day based on adverse liver effects at a human equivalent dose of 1 mg/kg/day in two species in subchronic studies and an uncertainty factor of 5,000 ($10^{½}$ for extrapolation from animals to humans, $10^{½}$ for use of a subchronic study, $10^{½}$ for use of a LOAEL, 50 to protect sensitive individuals, and a modifying factor of $10^{½}$ to reflect background exposure to trichloroethylene and its metabolites).

EPA (2001) characterized the weight of evidence for trichloroethylene as "highly likely to be carcinogenic to humans" under the proposed guidelines and "probable human carcinogen" (Group B1) under the current guidelines, based on limited human evidence from the joint analysis of epidemiology papers by Wartenberg et al. (2000), sufficient evidence in animals, and mechanistic information suggesting that trichloroethylene's mode of action may be relevant to humans. EPA (2001) calculated draft oral slope factors from data for a variety of tumors in humans and animals; after discounting the data showing the lowest risks (studies in rats, which appear to be less sensitive than humans or mice) and the highest risk (from a human inhalation epidemiology study based on a small number of cases and an uncertain exposure estimate), EPA concluded that confidence is greatest in the central risk estimates 0.02–0.4 per mg/kg/day (from human occupational inhalation data for kidney cancer, human oral environmental data for lymphoma, and mouse data for liver cancer). The corresponding inhalation unit risk from the human occupational data for kidney cancer was $5x10^{-6}$ per (μg/m^3). Similar conclusions regarding weight of evidence were reached in other recent assessments of trichloroethylene carcinogenicity. NTP (2001) listed trichloroethylene as reasonably anticipated to be a human carcinogen based on limited evidence of carcinogenicity from studies in humans, sufficient evidence of malignant tumor formation in experimental animals, and convincing relevant information that trichloroethylene acts through mechanisms indicating it would likely cause cancer in humans. IARC (1995) assigned trichloroethylene to Cancer Group 2A, *probably carcinogenic to humans*, based on limited evidence in humans and sufficient evidence in experimental animals.

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Appendix D: Background Information for Arsenic

D.1 Toxicokinetics

Arsenic, as water soluble arsenate or arsenite, is well-absorbed (≥80%) in both humans and animals exposed by the oral route (ATSDR 2000; NRC 1999). Judging from the oral toxicity data, arsenic trioxide also is well absorbed. Lower rates of absorption have been observed with insoluble or less soluble forms of arsenic, such as arsenic sulfide and lead arsenate. Absorption appears to occur by passive diffusion. Distribution occurs throughout the body. Concentrations in skin of humans exposed to background levels of arsenic are higher than in other tissues except blood. Arsenic accumulates in the skin of animals following long-term exposure. Concentrations in hair and nails tend to be higher than in live tissues. The rat tends to sequester arsenic in erythrocytes. Arsenates (As[V]) and arsenites (As[III]) are interconverted in the body by reduction/oxidation reactions. Reduction of arsenate to arsenite can be mediated by glutathione. Arsenite is methylated to yield the less toxic forms monomethylarsenite and dimethylarsenite. The liver is the major site for the methylation. Arsenic is promptly eliminated in the urine as a mixture of As(III), As(V), and the methylated forms. Smaller amounts are excreted in the feces.

D.2 Health Effects

Chronic oral exposure to arsenic has resulted in serious damage to the vascular system in humans, including Blackfoot disease (a progressive loss of circulation in the fingers and toes that may lead to gangrene), Raynaud's disease, and cyanosis of fingers and toes (ATSDR 2000; NRC 1999). The intima of the blood vessels appeared to have thickened. Direct irritation of the gastrointestinal mucosa can occur. Arsenic has caused anemia in humans exposed by the oral route. Increased hemolysis and a toxic effect on the erythropoietic cells of bone marrow may be factors in the development of anemia. Leukopenia has been reported in humans. Hepatic effects seen in humans were thought to be secondary to portal tract fibrosis and portal hypertension, which may have originated from damage to the blood vessels. Signs of renal damage generally are not seen or are mild in humans exposed to arsenic by the oral route. Characteristic dermal lesions caused by long-term oral exposure of humans to arsenic include hyperkeratinization (particularly on the palms and soles), formation of hyperkeratinized corns or warts, and hyperpigmentation of the skin with associated spots of hypopigmentation. A fraction of the hyperkeratinized corns may progress to squamous cell carcinoma of the skin. Signs of peripheral and/or central neuropathy are commonly seen in humans exposed to arsenic orally, with high-dose exposure producing central nervous system effects and low-dose exposure producing peripheral nervous system effects. The potential for arsenic to cause subtle neurological effects, such as neurobehavioral effects in children, has not been fully investigated. Studies of associations between hair arsenic concentrations (a biomarker of exposure) and neurobehavioral effects in children have observed an inverse association between hair arsenic and reading and spelling performance (Moon et al. 1985). Children may be

especially susceptible to arsenic because there is evidence that metabolism (i.e., detoxification) of arsenic may be less efficient in children and because arsenic's ability to inhibit cellular proliferation might be especially problematic in rapidly growing young children.

Effects on the skin, vascular system, and neurological system appear to be relatively sensitive effects of ingested arsenic; dermal effects are the best documented sensitive effect and the earliest observable sign of health effects from long-term exposure (ATSDR 2000; NRC 1999). The no-observed-adverse-effect level (NOAEL) and LOAEL for dermal effects in humans are $8x10^{-4}$ and 0.014 mg/kg/day, respectively. Hematological effects may be somewhat less sensitive, and renal effects even less sensitive and less common. Epidemiological studies provide convincing evidence that ingestion of arsenic causes cancer of the skin in humans. The lesions include squamous cell carcinomas, which develop from some of the hyperkeratotic warts or corns, and multiple basal cell carcinomas, arising from cells not associated with hyperkeratinization. Evidence is mounting that ingested arsenic may increase the risks of internal cancers as well (NRC 2001).

Some of the effects of arsenic seen in humans are supported by animal data, but animals do not develop dermal lesions and cancer as a result of oral arsenic exposure. Changes in vascular reactivity have been reported in rats given repeated oral arsenic doses of 11 mg/kg/day (ATSDR 2000). Hematological and hematopoietic effects, including decreased hematocrit and increased urinary excretion of porphyrins, have been observed in intermediate-duration dietary studies of arsenic in rats at doses of 2.5 mg/kg/day (Fowler and Mahaffey 1978; Mahaffey et al. 1981), and in chronic oral studies in dogs at 2.4 mg/kg/day (ATSDR 2000). Intermediate oral studies in rats demonstrated alterations in renal mitochondria at 2.5 and 4.7 mg/kg/day (ATSDR 2000; Mahaffey and Fowler 1977; Mahaffey et al. 1981). Mild proteinuria was observed in rats following a single oral dose of 10 mg/kg (ATSDR 2000). Repeated oral administration of arsenic to mice at 11 mg/kg/day altered neurotransmitter concentrations in some areas of the brain (Mejia et al. 1997). Developmental effects have been seen following high oral doses of arsenic in animals, but these are not sensitive effects (ATSDR 2000).

D.3 Mechanisms of Action

At relatively high oral exposures, methylation capacity may not be adequate to prevent cytotoxic levels of arsenic(III) from reaching tissues. Some of the effects of higher-dose oral exposure to arsenic are thought to be the result of direct cytotoxicity; these include gastrointestinal irritation, and dermal and neurological effects (ATSDR 2000). Arsenic(III) reacts with the sulfhydryl groups of proteins, inactivates enzymes, and interferes with mitochondrial function by inhibiting succinic dehydrogenase activity and uncoupling oxidative phosphorylation. It has been proposed that arsenic may compete with phosphate during oxidative phosphorylation and may inhibit energy-linked reduction of nicotinamide adenine dinucleotide (Goyer 1995). Chronic low-level exposure to arsenic is thought to stimulate keratinocyte secretion of growth factors; the resulting increase in cell division and DNA replication affords greater opportunities

for genetic damage. Arsenic induces metallothionein, a metal-binding protein. Only a small percentage of administered arsenic is bound to metallothionein, and the affinity of arsenic for metallothionein is much lower than that of cadmium or zinc (ATSDR 2000). It has been suggested that metallothionein may protect against arsenic toxicity by acting as an antioxidant against oxidative injury produced by arsenic (ATSDR 2000; NRC 1999).

D.4 Health Guidelines

ATSDR (2000) did not derive inhalation MRLs or an intermediate oral MRL for arsenic due to lack of suitable studies. ATSDR (2000) derived a provisional acute oral MRL of 0.005 mg/kg/day for arsenic based on a LOAEL of 0.05 mg/kg/day for facial (periorbital) edema and gastrointestinal irritation in poisoning cases from arsenic-contaminated soy sauce in Japan (Mizuta et al. 1956). These effects were the initial effects, and in some patients, were followed by dermal lesions, neuropathy (hypesthesia in legs, abnormal patellar reflex), mild anemia, mild degenerative liver lesions and hepatic dysfunction, and abnormal electrocardiogram. An uncertainty factor of 10 was applied to account for the use of a LOAEL. The MRL is considered provisional because the gastrointestinal effects were serious and because serious neurological and cardiovascular effects also occurred at the same dose. ATSDR (2000) derived a chronic oral MRL of $3x10^{-4}$ mg/kg/day for arsenic based on a NOAEL of $8x10^{-4}$ mg/kg/day for dermal lesions in male and female farmers exposed to high levels of arsenic in well water in Taiwan. An uncertainty factor of 3 was applied to account for human variability.

EPA has not derived an RfC for arsenic (IRIS 2001). EPA (IRIS 2001) derived a chronic RfD of $3x10^{-4}$ mg/kg/day for arsenic based on a NOAEL of $8x10^{-4}$ mg/kg/day for dermal lesions and possible vascular complications for farmers in Taiwan, which also was used as the basis for the ATSDR chronic oral MRL. An uncertainty factor of 3 was applied to account for the lack of reproductive data and to account for some uncertainty in which the NOAEL in the critical study accounts for all potentially sensitive individuals.

NTP (2001) has determined that inorganic arsenic compounds are known to be human carcinogens, based on sufficient evidence of carcinogenicity in humans. IARC (1987) concluded that there is sufficient evidence of a relationship between exposure to arsenic and human cancer, and classifies arsenic in Group 1. The American Conference of Governmental Industrial Hygienists (ACGIH) classifies arsenic (elemental and inorganic compound) as a confirmed human carcinogen; cancer category A1 (ACGIH 1998). EPA (IRIS 2001) has classified arsenic in Group A (human carcinogen), based on increased lung cancer mortality in several human populations exposed primarily through inhalation, increased mortality from internal organ cancers (liver, kidney, lung, and bladder), and increased incidences of skin cancer in populations exposed to arsenic through drinking water. An oral slope factor of 1.5 per (mg/kg)/day was derived based on analysis of the skin cancer data from a Taiwanese population exposed through drinking water. An inhalation unit risk of $4.3x10^{-3}$ per $\mu g/m^3$ was derived based on age-specific mortality from lung cancer in male smelter workers.

D.5 References

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Appendix E: Background Information for Strontium-90

^{90}Sr is a radioisotope of strontium. ^{90}Sr decays by emission of a beta-particle with a maximum energy of 0.546 millions of electron volts (MeV) and the creation of an yttrium-90 (^{90}Y) radioisotope, or daughter product. Unlike other radioactive isotopes that decay by beta-emission, ^{90}Sr does not directly release high energy photons or gamma-ray radiation (γ) (Brown 1997). However, the daughter product of ^{90}Sr, ^{90}Y, is both a beta-particle (2.28 MeV maximum energy) emitter, and to a minor degree for 0.012% of all disintegrations, a beta-particle and gamma-ray emitter. The decay product of ^{90}Y is ^{90}Zr, a stable isotope. The reaction is:

$$^{90}Sr\ (t_{1/2} = 29\ \text{years}) \rightarrow {}^{90}Y + \beta^{-}(0.546\ \text{MeV})$$

$$^{90}Y\ (t_{1/2} = 64\ \text{hours}) \rightarrow {}^{90}Zr\ (\text{stable}) + \beta^{-}(2.28\ \text{MeV})\ \ (99.989\%)$$
$$\rightarrow {}^{90}Zr\ (\text{stable}) + \beta^{-}(0.519\ \text{MeV}) + \gamma(1.76\ \text{MeV})\ \ (0.012\%)$$

E.1 Toxicokinetics

Stable strontium and radioactive strontium do not differ with regard to disposition in the body (ATSDR 2001c). Absorption following inhalation exposure depends on the chemical form of the inhaled strontium. Soluble compounds are rapidly absorbed from the lung (within hours), while more insoluble compounds may remain in the lung for extended periods of time (years). Absorption of ingested strontium (whether in the diet or administered as soluble strontium chloride [$SrCl_2$]) from the gastrointestinal tract is approximately 20% (range, 11–25%) in humans. Studies in rats suggest that absorption may be considerably higher in neonates. Within the gastrointestinal tract, absorption of strontium appears to occur in both the stomach and small intestine. Strontium is not well absorbed across intact skin, but passes much faster through scratched or abraded skin.

The distribution of absorbed strontium in the human body is similar to that of calcium, with approximately 99% of the total body burden in the skeleton. Strontium distributes relatively uniformly within the bone volume, where it exchanges with calcium in hydroxyapatite. Strontium is also found in the soft tissues, although at much lower concentrations than in bone. Strontium in the maternal skeleton can be transferred to the fetus during pregnancy. The distribution of strontium in the fetus at the end of gestation is similar to that of the mother, with most of the strontium burden in the skeleton. Strontium enters milk in humans and animals and can be transferred to newborns during breast feeding. Strontium is not metabolized in the body. However, strontium does bind with proteins and, based on its similarity to calcium, probably forms complex formation with various inorganic anions such as carbonate and phosphate, and carboxylic acids such as citrate and lactate.

Absorbed strontium is excreted primarily in the urine and feces. Urinary excretion is approximately 3-fold higher than fecal excretion. The observation of fecal excretion of radioactive strontium weeks to decades after an oral exposure or over shorter time periods after an intravenous exposure suggests the existence of a mechanism for transfer of absorbed strontium into the gastrointestinal tract, either from the bile or directly from the plasma. During lactation, absorbed strontium is also eliminated in breast milk. The terminal elimination half-time for strontium in humans has been estimated to be approximately 25 years. Estimates of the terminal elimination half-times of strontium reflect primarily the storage and release of strontium from bone. Over shorter time periods after exposure, faster elimination rates are observed that reflect soft-tissue elimination as well as elimination from a more rapidly exchangeable pool of strontium in bone.

E.2 Health Effects

The basis of the adverse effects of ionizing radiation on human or animal tissue is the direct interaction of free radicals with cellular macromolecules, including DNA (ATSDR 2001c). Low-level exposures are not necessarily harmful, as shown by the lack of discernable adverse effects in the general population from chronic low-level exposure to ^{90}Sr in fallout during the period of above ground weapons testing. Exposures to radioactive strontium become harmful when the amount of radiation damage exceeds the capacity of natural cellular repair mechanisms. External exposure to radioactive strontium has resulted in dermal and ocular effects in humans. Since absorbed radiostrontium is preferentially retained in bone, and therefore has a long biological half-life, all exposures leading to the presence of radiostrontium in the body, of whatever duration, will lead to chronic internal exposure to ionizing radiation. Consequently, the most significant effects of exposure to absorbed radioactive strontium are necrosis and cancers of bone, bone marrow, and tissues adjacent to bone. High level acute exposures can lead to acute radiation sickness resulting from destruction of the hematopoietic bone marrow. Dystrophic or osteolytic lesions have been described in humans and animals following intermediate or chronic exposures. At lower levels of exposure, chronic suppression of immune function has been observed in humans and animals. In animal studies, inhalation of insoluble particles of radioactive strontium led to retention in the lung and resulted in pulmonary necrosis and cancer. The young are more susceptible to adverse effects of absorbed radioactive strontium because of their higher rates of gastrointestinal absorption and of strontium retention in the immature skeleton. High prenatal exposure levels may cause major developmental anomalies in the skeleton and adjacent areas if critical tissues are destroyed. In addition, since children have a higher proportion of mitotic cells than adults, they may exhibit higher rates of cancer (genetic lesions become fixed mutations when mitosis occurs before genetic damage is repaired). Persons with Paget's disease (osteitis deformans) may be vulnerable to radioactive strontium because of their higher than normal rates of retention in focal sites of bone deposition.

E.3 Mechanisms of Action

The adverse health effects of radioactive strontium are related to its sequestration in bone, the high energy of its beta emissions, and in the case of ^{90}Sr, its long half-life (ATSDR 2001c). An extensive discussion of ionizing radiation and its health effects is found in the Toxicological Profile for Ionizing Radiation (ATSDR 1999). Beta emissions from radiostrontium bound to bone have resulted in various bone lesions (trabecular osteoporosis, sclerosis, osteolytic lesions), particularly in animals that were exposed chronically. In young rats and rabbits exposed orally to ^{90}Sr, necrotic effects on the vasculature of developing bone secondarily disrupted the process of osteogenesis. Disruption in the metaphyseal microvasculature disorganized the transformation of cartilage into bone, so that chondrocytes inappropriately resumed active proliferation. Severe reduction in hematopoietic tissue results from irradiation of the bone marrow by radiostrontium incorporated into bone. At high exposure levels, thrombocytopenia may lead to platelet loss severe enough to cause hemorrhaging; the resulting anemia will be exacerbated by destruction of erythropoietic tissue. Impaired immune function results from the genetic damage to lymphocytes.

Radioactive strontium is a genotoxic carcinogen. Following exposure *in vivo*, cytogenetic analysis has revealed aneuploidy, chromosomal breaks, gaps, rings, and exchanges, which are manifestations of unrepairable changes in DNA. It is generally understood that radiation-induced damage to genes that regulate cell growth is a major factor in the development of cancer in affected cells, and the observation of chromosomal breaks in leukemic cells of miniature swine following chronic oral exposure to $^{90}SrCl_2$ is consistent with this idea. However, the specific genes involved in radiostrontium-induced malignancies have not been identified. Because of strontium's chemical properties, which determine its distribution in the body, exposure to sufficient radiostrontium results in an increased risk of malignancy for particular tissues. In dogs, acute inhalation of insoluble ^{90}Sr particles that lodged in the lungs resulted in chronic radiation exposure to the lungs, leading to pulmonary hemangiomas and carcinomas of pulmonary epithelia. Other tissues were subsequently affected as the radioactive particles were cleared from the lungs. Following acute inhalation of soluble $^{90}SrCl_2$ aerosols, some dogs developed carcinomas of nasal airway tissues, probably resulting from irradiation of these tissues from the ^{90}Sr bound to the underlying bone. Following oral or inhalation exposures, absorbed ^{90}Sr was distributed to bone, from which it irradiated the surrounding tissues and induced various kinds of osteosarcomas, as well as malignancies of hematopoietic tissues in bone marrow.

E.4 Health Guidelines

No MRLs were derived for inhalation or oral exposures to radioactive strontium (ATSDR 2001c). The EPA has not derived an RfC or RfD for radioactive strontium (IRIS 2001). IARC has determined that all internally deposited beta emitters, including radioactive strontium, are carcinogenic to humans and has assigned them to Group 1 (IARC 2001). Radioactive strontium is not included in NTP's 9th Report on

Carcinogens (2001). The EPA has determined that all radionuclides, including radioactive strontium, are known human carcinogens, and has assigned them to Group A (EPA 1997). The EPA (1997) has calculated carcinogenicity slope factors (upper bound lifetime risk per pCi) for ^{90}Sr for ingestion ($4.09x10^{-11}$ for ^{90}Sr and $5.59x10^{-11}$ for ^{90}Sr plus disintegration products) and inhalation ($5.94x10^{-11}$ for ^{90}Sr and $6.93x10^{-11}$ for ^{90}Sr plus disintegration products).

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