Public Health Challenges Posed by Chemical Mixtures

Hugh Hansen, Christopher T. De Rosa, Hana Pohl, Michael Fay, and Moiz M. Mumtaz

Agency for Toxic Substances and Disease Registry, Division of Toxicology, Atlanta, Georgia

Approximately 40 million people live within a 4-mile radius of waste sites that the Agency for Toxic Substances and Disease Registry (ATSDR) has assessed to date. Human populations living in the vicinity of such sites are often subjected to complex chemical exposures that may contribute to the total body burden of oxogenous chemicals. Apart from the contaminants found at waste sites, exposure may also include environmental, occupational, and personal agents. Concurrent exposure to chemicals such as welding fumes, indoor air pollutants, tobacco smoke, alcohol, and prescription and nonprescription drugs makes the health assessment of exposure to waste site chemicals a more complex task. Voluntary exposures such as these frequently entail exposures to relatively high chemical concentrations and can usually be well defined and quantified. Conversely, involuntary exposures from waste sites may be at low concentrations and hence difficult to characterize and quantify. Of the approximately 1450 waste sites evaluated by the ATSDR, 530 (37%) had either completed or potentially completed exposure pathways. Results of public health assessments conducted at 167 sites during 1993 to 1995 show that about 1.5 million people have been exposed to site-specific contaminants. At 10% or more of the sites that had either completed or potentially completed exposure pathways, 56 substances were identified. Of these, 19 are either known or anticipated human carcinogens, and 9 are associated with reproductive or endocrine-disrupting effects. In this paper we present important concerns regarding hazardous waste sites including the impact on human health, ecology, and quality of life. To address such human-health related issues, the ATSDR has established a mixtures program that consists of three components: trend analysis to identify combinations of chemicals of concern, experimental studies to identify data that would be useful in the development and implementation of predictive decision support methodologies, and development of assessment methodologies and guidance to provide health assessors with the tools to incorporate the evaluation of multiple-chemical exposure into site assessments. — *Environ Health Perspect* 106(Suppl 6):1271–1280 (1998). *http://ehpnet1.niehs.nih.gov/docs/1998/Suppl-6/1271-1280hansen/abstract.html*

Key words: chemical mixtures, hazardous waste, exposure, multiple chemical exposure, occupational, alcohol, drugs, tobacco

A growing number of chemicals are introduced into the marketplace on a weekly basis. Of approximately 7 million chemicals in existence, 70,000 are in current use with more than 1000 added each year worldwide (*1*). However, the U.S. Environmental Protection Agency (U.S. EPA) reports that in 1995 U.S. manufacturing facilities released more than 2.2 billion pounds of toxic chemicals into the environment, a decline in releases from previous years. Although this decline in releases is a welcome success of primary prevention, in certain instances body burdens of key pollutants are noted with little insight into the potential for joint toxic actions of such chemicals at environmental levels. In sharp contrast to the controlled conditions of laboratory investigations, humans are typically exposed to such chemicals through means that vary widely. A typical laboratory study involves the controlled exposure of experimental animals to a single chemical by a single route for a specified period of time (*2*). However, exposures normally experienced by people in the vicinity of hazardous waste sites (HWS) may be characterized as complex exposures—involving multiple agents and multiple pathways and patterns of exposure dramatically different from those typically studied in toxicologic research (*3*). Such exposures may affect at-risk populations because of elevated concentrations, enhanced toxicity resulting from interactions of different chemicals complex exposures, and possibly intrinsic physiologic sensitivity as well. For example, because the developing fetus is exquisitely sensitive to the effects of such chemicals at certain time-critical windows of exposure, certain adverse effects on the fetus may subsequently produce transgenerational effects (*4*).

Section 104(i)(5)A of the Comprehensive Environmental Response, Compensation, and Liability Act (CERCLA) (*5*) directs the Administrator of the Agency for Toxic Substances and Disease Registry (ATSDR) (in consultation with the Administrator of the U.S. EPA and agencies and programs of the U.S. Public Health Service) to assess whether adequate information on the health effects of [profile] substances is available. Where adequate information is not available, ATSDR, in cooperation with the U.S. National Toxicology Program, is required to assure the initiation of a program of research designed to determine these health effects. The statute further directs the ATSDR: Where feasible, develop methods to determine the health effects of substances in combination with other substances with which they are commonly found (*5*). This directive requires that the ATSDR encourage, initiate, coordinate, and conduct chemical mixtures research that would advance methods development for chemical mixtures assessment.

The human health effects and assessments of environmental chemicals found at HWS are documented in public health assessments prepared by the agency. Other primary concerns that impact human heath at HWS also need to be addressed: clean-up costs, property values, ecology, and quality of life.

This paper is based on a presentation at the Conference on Current Issues on Chemical Mixtures held 11–13 August 1997 in Fort Collins, Colorado. Manuscript received at *EHP* 26 May 1998; accepted 28 July 1998.

Address correspondence to H. Hansen, Agency for Toxic Substances and Disease Registry, Division of Toxicology, Mail Stop E-29, 1600 Clifton Road NE, Atlanta, GA 30333. Telephone: (404) 639-5293. Fax: (404) 296-4653. E-mail: hjh1@cdc.gov

Abbreviations used: ATSDR, Agency for Toxic Substances and Disease Registry; CERCLA, Comprehensive Environmental Response, Compensation, and Liability Act; CEPs, completed exposure pathways; HazDat, Hazardous Substance Release/Health Effects Database; HWS, hazardous waste sites; MRL, minimal risk level; NPL, national priorities list; OR, odds ratio; PBPK, physiologically based pharmacokinetic; PEL, permissible exposure limit; SAR, structure–activity relationships; SARA, Superfund Amendments and Reauthorization Act; TCE, trichloroethylene; TSP, total suspended particles; U.S. EPA, U.S. Environmental Protection Agency.

Primary Concerns

Human Health

One of the ATSDR's primary goals is to identify people at health risk because of their exposure to environmental chemicals. Although this goal can be accomplished through the establishment of a cause-and-effect relationship between environmental pollutants and public health impairment (*6*), causality in the face of complex exposures may be difficult to establish. In such cases a more realistic approach using weight-of-evidence evaluation as a surrogate for causality has been suggested (*7*). This approach allows for a broader consideration of studies that investigate human health effects at HWS that may not establish a direct causal link to adverse health effects in exposed populations. Such a characterization of data should be kept in mind when considering studies of health effects at HWS. Earlier epidemiologic studies that were conducted to investigate health effects in communities around HWS failed to report statistically significant increases in adverse health effects (*8*). However, recent investigations using meta-analysis techniques studied 593 HWS in 339 counties of 49 states, where the sole source of water supply was contaminated groundwater (*9*). For each identified county, age-adjusted site-specific cancer mortality rates for 13 major cancer sites (anatomic) were extracted from U.S. cancer mortality and trends during 1950 to 1979 for white males and during 1970 to 1979 for white females (*9*). Significant associations were found for cancers of the lung, bladder, esophagus, stomach, large intestine, and rectum for white males and for cancers of the lung, breast, bladder, stomach, large intestine, and rectum for white females. Because there were no exposure data on populations at risk of exposure to HWS contaminant releases, these findings should be interpreted with care.

Clearly, the array of complex exposures on which these generalizations are based compel a more in-depth consideration of the health effects for populations living in the vicinity of HWS. Such exposures may be responsible not only for obvious adverse health effects but also for compromised physiologic defenses that are rendered less effective in maintaining homeostasis. Cases of immunosuppression in both humans and animals for common agents such as asbestos, tobacco smoke, benzene, toluene, and various metals such as zinc and lead have been well documented. Exposures to such agents are usually concurrent. Injury, especially to the lung, by one agent will produce greater vulnerability to a second agent, such as cadmium chloride followed by inhalation of oxygen (*10*).

Figure 1 represents the spectrum of biologic response to pollutant exposure (*11*). At low-level exposures human populations do not show any observable health effects; thus the chemicals remain as body burdens having no discernable effect on the overall health of the individuals. Physiologically, the body adjusts to the presence of chemicals at this level through adaptive mechanisms. As the pollutant exposure levels increase, some effects may be observed; but these effects, such as enzyme induction and certain biochemical and subcellular changes, may be of uncertain significance. The body may have compensatory mechanisms at this level of pollutant exposure (*12*). However, as the levels of the pollutants increase, significant readily observable adverse effects may ensue. At these pollutant levels the body has exhausted its adaptive and compensatory mechanisms and its functioning could be compromised. Such adverse effects could lead to organ function impairment through compromise of physiologic processes, leading to pathophysiologic changes such as fatty changes and necrosis resulting in significant organ function impairment. Exposure to higher levels of pollutants than these levels could thus lead to morbidity and ultimately to death. Exposures from the multiple sources may cause some individuals to cross the threshold for adverse health effects in this continuum of effects. Considering that the human population is normally distributed and thus lacks homogeneity in biochemical characteristics, some fraction of the population will be more susceptible than others and some fraction could be hypersensitive. For these reasons a specific smaller fraction of the population may be hypersensitive to pollutant burdens and exhibit adverse responses to levels of exposure that may otherwise be considered low.

The contribution of hazardous waste to exposures of populations living near HWS may be for short periods at low levels but may constitute a significant contribution to overall body burdens when such exposures are concurrent with occupational and personal exposures. The capacity for compromised physiologic systems is even greater in sensitive subpopulations [i.e., children, women of child-bearing age, and the elderly (*5*)]. In such populations pollutant burdens can initiate pathophysiologic

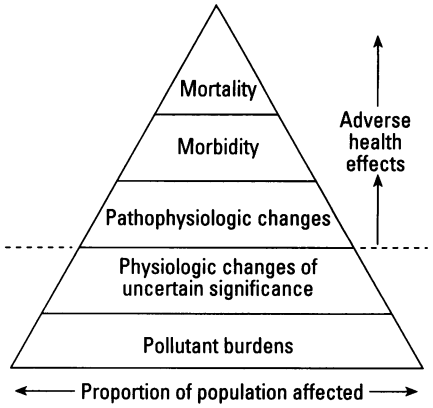

Figure 1. Spectrum of biologic response to environmental pollutant exposure. Adapted from the American Thoracic Society (*10*).

changes at lower levels in comparison to the normal population. For example, human infants and children differ from adults in size, immaturity of biochemical and physiologic functions in major body systems and body composition in terms of proportions of water, fat, protein, mineral mass, and chemical constituents (*13,14*). During the first 2 months of life, rapid development occurs in the brain (cell migration, neuron myelination, and creation of neuron synapses), lungs (developing alveoli), and bones (rapid growth and handling). Development of the brain and lungs continues until age 12, at which time gonad maturation, ova and sperm maturations, and breast development occur (*15*). Depending on the chemical, the stage of growth and development may be a critical variable in evaluating the toxicity of HWS chemicals (*16*).

Of particular concern are findings of recent studies that show associations between low-level exposures to hazardous chemicals and developmental effects or birth defects (*17–19*). Broadly defined, developmental toxicity is any adverse effect on the developing organism from implantation through prenatal development or postnatally to the time of sexual maturation (*20*). Developmental effects can be categorized as structural abnormalities, altered growth, functional deficiencies, congenital neoplasia, and death of the developing organism. Exposure to a chemical causing developmental toxicity can occur to either parent before conception, to the mother during pregnancy, or directly to the developing organism postnatally (e.g., via breast feeding) (*21*). Many chemicals can cross the placenta and concentrate in the fetus. Thus, the developing fetus is extremely

vulnerable to chemical exposure. In fact, the actual concept of testing for developmental toxicity requires observation of developmental effects in a fetus without apparent maternal toxicity.

Structural abnormalities (birth defects) and altered growth play an important role in developmental toxicology for several reasons. They appear to be sensitive end points to detect chemical toxicity, and they are easier to recognize than some other effects (e.g., subtle functional deficiencies). The ease of recognition, however, does not necessarily mean that birth defects are more important than other end points such as lowered intelligence quotients or chronic alterations in the endocrine systems or behavior. Many birth defects can be linked to a specific period of development, thus enabling researchers to pinpoint the window of opportunity for toxic exposure. In laboratory settings, various patterns of exposure timing and dosing can be tested to obtain a whole spectrum of developmental effects such as delayed ossifications at low doses and skeletal malformations at high doses. In contrast, human exposure to environmental chemicals usually represents a combination of exposure pathways, timing patterns, and doses.

Susceptibility of human fetuses and young infants to adverse health effects caused by high-dose exposures to environmental chemicals has been well documented in epidemiologic studies. For example, mothers who consumed rice oil accidentally contaminated by a heat exchange fluid containing chemical mixtures of polychlorinated biphenyls in Japan in 1968 (Yusho incident) and Taiwan in 1979 (Yu-Cheng incident) had children with developmental health effects (*22–26*). Effects such as permanent abnormalities in neurobehavioral function, skin hyperpigmentation, decreased birth weight, and porphyrinuria were seen in infants exposed *in utero* and/or via breast feeding (*27*).

Similarly, several outbreaks of mercury poisoning have been described around the world. In 1956 and 1960, bread made from grain treated with fungicides containing mercury was consumed by pregnant women in Iraq (*28*). Infants were born with severe brain damage. A similar outbreak was reported again in 1971 to 1972 (*29*). High mercury concentrations were associated with the more severe effects such as mental retardation and seizures; lower mercury concentrations correlated with delays in walking and talking (*30,31*). In Minamata, Japan, infants exposed *in utero* had severe brain damage resulting from their mothers' consumption of methylmercury-contaminated fish during pregnancy (*32*).

Besides effects observed following high-dose exposures to chemicals in the environment, low-dose exposures may also be harmful. For example, a number of recent epidemiologic studies have demonstrated neurobehavioral impairment at low-dose exposure levels of lead that were once thought to be safe. Many of these studies have been reviewed elsewhere (*17,33,34*). Reproductive effects of hazardous substances released from HWS have been extensively documented by health investigations. Several studies have examined associations between maternal residence near HWS and the occurrence of birth defects, low birth weight, and other reproductive outcomes (*35–38*).

It is important to realize that humans are exposed daily to many chemicals from multiple sources. Low-dose exposure to chemical mixtures may play an important role in developmental toxicology because of possible interactions among the components of the mixture. The ATSDR recognized these concerns and conducted or supported several studies that investigated low-level exposures and their correlation with developmental effects. Some of these studies reported on public drinking water contamination and birth outcomes (*39,40*), whereas others evaluated associations between HWS and birth defects (*18,37*).

Increased risk has been reported for neural tube defects (odds ratio [OR] = 2.1) and heart defects (OR = 4.2) for mothers living within one-quarter mile of a national priorities list (NPL) site in California (*37*). The authors further found increased ORs for developmental effects in association with maternal residence within 1 mile of NPL sites containing selected chemical contaminants. For example, exposure to cyanides and pesticides was associated with neural tube defects; exposure to lead, arsenic, chromium, and 1,1,-dichloroethylene was associated with heart defects. The authors did not find any correlation between development of oral cleft defects and waste site exposures. No direct measurement of waste site exposure was performed for the study. The risk reported for single chemicals may be biased because of actual exposure (i.e., to a mixture of compounds) from the waste sites. Further, the study did not control for other sources of exposure to chemicals (e.g., industrial emissions, agricultural pesticides, occupational exposure, etc.).

Birth weight reduction (OR = 5.1) associated with parental residence near a hazardous waste landfill in New Jersey has been reported (Table 1) (*18*). Another finding was an increased risk for prematurity (OR = 2.1) in the exposed population (Figure 2). The results pertain to a narrow time period (1971 to 1975) when it is believed the landfill exposure was the greatest (the whole study period was from 1961 to 1985). The authors related the magnitude of the effect to that of birth weight reduction due to cigarette smoking during pregnancy. The study did not control for other potential risk factors for low birth weight such as maternal health, cigarette and alcohol consumption during pregnancy, parental occupation, and parental socioeconomic status.

In a study investigating adverse pregnancy outcomes in relation to exposure to volatile organic compounds from drinking water at a U.S. Marine Corps base in North Carolina, no effects were seen in the exposed group as a whole (*39*). In association with perchloroethylene exposure, two susceptible subgroups were identified: Small-for-gestational-age category was reported in mothers over 35 years old (OR = 3.9) and in mothers with histories of fetal death (OR = 1.6). In association with trichloroethylene (TCE) exposure, small-for-gestational-age results were observed in male offspring (OR = 3.9). However, the

Table 1. Evidence of low birth weight from studies.[a]

Study (reference)	Exposure description	Small for gestational age (risk)[b]	Birthweight deficit, avg
NJ 94 (*38*)	Residential proximity to Lipari landfill	5.1	–192 g
NC 97 (*39*)	U.S. Marine Base Camp Lejeune drinking water contaminants		
	Trichloroethylene	1.5	–139 g
	Tetrachloroethylene	3.9[c]	–209 g[c]
		1.6[d]	–51 g[c]

[a]Adapted from Berry and Bove (*38*). [b]Odds ratio. [c]Mothers 35 years of age and older. [d]Mothers with previous fetal deaths.

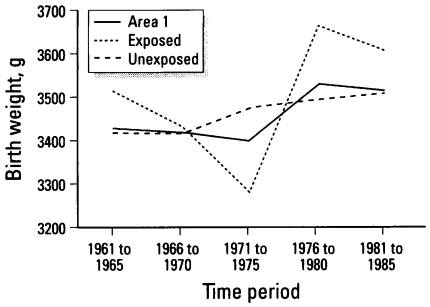

Figure 2. Average adjusted birth weight (grams) by 5-year time period for the years 1961 to 1985, Lipari landfill, New Jersey. Adapted from Berry and Bove (*38*). Gestational age 37 to 44 weeks.

sample size was too small and the sex-based difference has no biologic explanation based on known mechanisms of TCE toxicity.

Birth outcomes in relation to drinking water contamination in New Jersey during 1985 to 1988 have been reported by Bove et al. (*40*). Exposure to several chemicals present in drinking water was related to developmental effects in offspring of exposed mothers (Table 2). The strongest relations to adverse birth outcomes were noted for total trihalomethanes and carbon tetrachloride. The major bias in the study comes from possible exposure misclassification. The study did not control for maternal occupational exposure, smoking and alcohol or drug consumption, or any other source of chemical exposure. The authors realize that the study, by itself, cannot resolve whether the drinking water contaminants caused the adverse birth outcomes.

In summary, these studies conducted or supported by the ATSDR are suggestive of the following: Elevations in the rates of neural tube defects and major cardiac defects have been found in populations residing in the proximity of toxic waste sites and that consumed contaminated public drinking water. Also, elevations in the rate of oral cleft defects were linked to contaminated public drinking water. Substantial exposures to toxic waste site contaminants and drinking water contaminants are related to the small-for-gestational-age category. The possibility of adverse developmental outcomes in connection with low-dose exposure to chemical mixtures is real. However, further studies are needed to better define the plausible risk to future generations. Based on the findings of the preceding studies the impact of the evidence on cancer and adverse reproductive outcomes was summarized to Congress as follows: "Although epidemiologic findings are still unfolding, when evaluated in aggregate (i.e., by combining health data from many Superfund sites), proximity to hazardous waste sites seems to be associated with a small to moderate increased risk of some kinds of birth defects and, less well documented, some specific cancers" (*41*).

Quality of Life

The ATSDR mission is "to prevent exposure and adverse human health effects and the diminished quality of life associated with exposure to hazardous substances from waste sites, unplanned releases, and other sources of pollution present in the environment" (*6*). With respect to the ATSDR mission, the quality of life of persons exposed or potentially exposed to hazardous substances in the environment may be diminished at either end of the quality of life continuum (*42–44*). The quality of life of people living in the vicinity of HWS may be diminished directly and concretely as a consequence of actual disease caused by exposure to hazardous substances, or indirectly as a consequence of the psychologic stress caused by individuals' perception of their (or their family's or community's) increased risk for disease occasioned by the exposure. Health assessors are asked to be aware of the importance the agency places on a site's impact on quality of life. In a public health context, sensory impairments, neurobehavioral concerns, and emotional well-being all contribute to the issue of quality of life. Factors affecting the quality of life for populations living near HWS have been addressed in an ATSDR report (*45*).

Ecology

As a part of its health-related responsibilities in regard to HWS, the ATSDR requested that the National Academy of Sciences review and evaluate whether animal epidemiologic studies would be useful for human health risk assessment (*46*). This effort revealed a wealth of literature that documents the presence of environmental contaminants in the tissues of organisms throughout the food chain (*47*). In the exposure assessment process, ATSDR health assessors evaluate food production as a potential source when food production is used as a subsistence food source. In certain areas biologic magnification via wild plants, animals, and fish may contaminate a food source that constitutes a significant portion of the diet of local residents (*48*). The U.S. EPA is required to protect human health and the environment with respect to releases or potential releases of contaminants from abandoned HWS (*49*). The national contingency plan calls for identification and mitigation of the environmental impacts of these sites and the selection of remedial actions that are "protective of environmental organisms and ecosystems" (*49*). In response the U.S. EPA has developed a framework for ecologic risk assessment (*50*).

Exposures in Context

Human populations living in the vicinity of HWS are often subjected to complex chemical exposures that may contribute to the total body burden. Apart from the contaminants found at waste sites, exposure may also include environmental, occupational, and personal agents. Concurrent exposure to chemicals such as welding fumes, indoor air pollutants, tobacco smoke, alcohol, and prescription and nonprescription drugs makes the health assessment of exposure to involuntary waste site chemicals a more complex task. Voluntary exposures such as these frequently entail exposures to relatively high chemical concentrations and can usually be well defined and quantified whereas involuntary exposures from waste sites may be at low concentrations and hence be difficult to characterize and quantify (Figure 3). The figure represents a control gradient with decreased individual control over exposures proceeding from personal, occupational, and environmental to hazardous waste.

Table 2. Evidence of structural birth defects from studies in New Jersey in 1995.[a]

	Risk, odds ratio			
Exposure description	Central nervous system	Neural tube defects	Major cardiac defects	Oral cleft defects
Carbon tetrachloride	3.8	5.4	1.0	3.6
Trichloroethylene	1.7	2.5	1.2	2.2
Tetrachloroethylene	<1.0	1.2	1.1	3.5
Dichloroethylene	2.5	2.6	<1.0	1.7
1,2-Dichloroethane	<1.0	1.0	2.1	<1.0
Benzene	<1.0	2.1	1.8	<1.0

[a]Adapted from Bove et al. (*37*).

Figure 3. Human exposure near hazardous waste sites: a control gradient with decreased individual control over exposures proceeding from personal, occupational, and environmental to hazardous waste.

Personal exposures such as to first-hand tobacco smoke or alcohol are voluntary. Occupational exposure is voluntary but the individual has less control over exposures. The individual has few options for controlling environmental exposures under ordinary circumstances. In most cases there are no clear options for individual control over hazardous waste exposures. Sometimes an individual may not even be aware of the site, the exposure pathways, or the nature of the exposure. In terms of concentrations, for normal populations personal exposures are usually at higher levels than hazardous waste exposures. Added to these personal exposures are increments from occupational, environmental, and hazardous waste exposures.

The relative magnitude of exposures is reflected in governmental regulations and guidelines for occupational verses environmental exposures (Table 3) (*51*). For example, for methylene chloride the permissible exposure limit (PEL) is 500 ppm whereas the minimal risk level (MRL) is 0.4 ppm. Thus, the ratio for the PEL to the MRL is 1250. The PEL to MRL ratio for benzene is 20 and for TCE is 50. Finkel et al. (*52*) state, "People are both biologically unique in their sensitivity to environmental pollutants and also environmentally unique in the exposures they face due to geography, occupation, lifestyle, mobility patterns, and activities."

Although the U.S. EPA reports almost a 50% decline in releases of toxic chemicals in the past 8 years, the *Toxics Release Inventory* (*53*) shows the number of those chemicals classified as toxic wastes continues to rise. Recent studies on the health effects of air pollution point to the increased awareness that air pollution as a mixture of chemicals must be addressed. The components of this complex mixture may include ozone, sulfur dioxide, and total suspended particles (TSP), which may include brake dust, road dust, gasoline, diesel engine exhaust, incinerator emissions, and coal fly ash. Recent findings show a consistent association between air pollution and mortality. Of particular health significance is the TSP component of air pollution, which is itself a complex mixture (*54*). Acute adverse respiratory effects of ozone exposure are also well documented in animal and human populations (*55–57*).

The National Institute for Occupational Safety and Health has recently developed a national occupational research agenda that characterizes the national workforce (*58*). Currently, about 125 million people are in the U.S. workforce. In less than a decade, this number will grow to 147 million and the workforce will be older and more racially diverse. Further, more than half of the U.S. workforce is employed indoors and office and indoor job sectors continue to expand. Along with the trend toward an indoor nonindustrial workforce has been the increase in reports of symptoms and signs related to indoor air environments. These range from allergic and infectious diseases to nonspecific symptoms including headaches and eye irritations. Although the majority of health problems reported cannot be attributed to specific exposures, evidence suggests that multiple factors are involved, including microbiologic, chemical, physical, and psychologic/social stressors (*58*). The exposures to personal agents in the environment are well known: Tobacco, prescription and nonprescription drugs, alcohol, herbal remedies, vitamins, and cosmetics add to daily incremental exposures to complex mixtures.

Completed Exposure Pathways

Although numerous chemicals might occur in the environment and be frequently encountered at HWS, not all are of actual public health concern (*59,60*). A method to identify chemical mixtures of actual public health concern has been linked to the concept of completed exposure pathways (CEPs) at various HWS (*61*). A CEP evaluation consists of identifying and characterizing the following five elements: source of contamination, environmental medium, point of exposure, route(s) of exposure, and a receptor population (*62*). A CEP occurs when the five elements of an exposure pathway link the contaminant source to receptor populations. Should a CEP exist in the past, present, or future, the population is considered exposed. A potential exposure pathway exists when one or more of the five elements are missing or if modeling is performed to replace real sampling data (e.g., modeled groundwater data using soil or other groundwater data levels). For fiscal year 1996 ATSDR investigators conducted health assessments at sites at which more than 111,550 people were exposed to site contaminants and more than 59,900 people were potentially exposed (*6*).

Completed Exposure Pathways at National Priority List Sites

An analysis conducted using the ATSDR comprehensive on-line hazardous substance release/health effects database (HazDat) (*63*) has shown that of approximately 1450 NPL sites evaluated, 530 (37%) had chemicals identified in CEPs. The total number of CEP incidents for the sites was 7244, or about 14 incidents per site. The exposure routes for these chemicals were as follows: 91% through groundwater, 46% through contaminated soil, and 14% through contaminated biota. These numbers are larger when potential CEPs are included in the analysis. The top five chemicals on the CEP site count report were TCE, lead, tetrachloroethylene, arsenic, and benzene (*64*).

The ATSDR uses CEP data to develop toxicologic information databases, initiate health investigations, focus toxicologic research, and refine the agency's priority substance list. The ATSDR lists the chemicals to which people have been exposed at HWS; the agency included with the CERCLA *Priority List of*

Table 3. Comparison and ratio of permissible exposure limits to minimal risk levels.

Chemical	PEL,[a] ppm, TWA	MRL,[b] ppm, acute	Ratio, PEL/MRL
Trichloroethylene	100	2	50
Benzene	1	0.05	20
Mercury, mg/m^3	0.1	0.0002	500
Toluene	200	3	67
Methylene chloride	500	0.4	1250
Vinyl chloride	1	0.5	2
Carbon tetrachloride	10	0.2	50

TWA, time-weighted average. [a]Occupational Safety and Health Administration (*49*). [b]Chou et al. (*71*).

Hazardous Substances (*64*) a CEP site count report. The CEP ranking presented in the list is based on a site frequency count and thus lists the number of sites at which a substance has been found in a CEP. This information is derived from ATSDR's public health assessments and consultations. Because this CEP report focuses on documented exposure, it provides an important prioritization based on substances to which people are known to have been exposed (*64*).

Once the chemicals and their mixtures have been identified in the CEP, the significance of exposure to such chemical mixtures needs to be determined. This determination is based on a multistep public health assessment process that consists of integrating input and conclusions of four major components, namely, toxicity and exposure assessment, environmental monitoring data, health outcome data, and community health concerns (Figure 4). To evaluate the exposure and the toxicity of chemicals that have been identified and quantitated at a site, agency-derived MRLs are used as an initial screen. Because such evaluations could be from exposure through multiple media, namely, water, soil, and air, the MRLs for more than one exposure route are used in determining the significance of such exposures.

Agency health assessors usually conduct a site visit to collect pertinent information. Such site visits also involve, at times, meetings with one or more of the following: the individual members of a community living in the vicinity of the site, community representatives, local health officials, or representatives of the local news media. This information may give the health assessor a basis to evaluate the conclusions of the toxicity assessment of the chemical mixtures in light of the community health concerns. Such exhaustive and complex evaluations lead to a consideration of a full range of potential follow-up activities and appropriate recommendations, including health studies, health education, research, or establishment of registries to monitor potentially exposed human populations. The challenges posed by the many areas of uncertainty involved in these evaluations and the limited time and financial resources available are considerable.

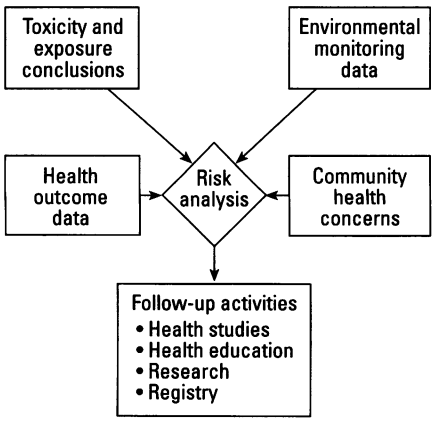

Figure 4. Contributions to risk analysis in the determination of significant human exposure.

The ATSDR Approach to Chemical Mixtures

Because all the possible chemical mixtures to which human beings are potentially exposed cannot be experimentally tested, the ATSDR has developed a research program in chemical mixtures. The primary objectives of this program are to perform a critical synthesis of relevant data and identify generalizable rules that can be used in site-specific assessments of health risk following exposure to mixtures of environmental chemicals. This research program allows scientists to pursue various aspects of chemical mixtures research such as to identify environmental chemical mixtures that affect public health; evaluate the potential for exposure of human populations to chemical mixtures; study the pharmacokinetic behavior of chemical mixtures; identify various end points that would be affected; evaluate target organs that would be affected; study the mechanisms of action, progression, and repair; identify biomarkers (specific and generic) that would allow the determination of the health of an organism; and develop qualitative and quantitative health assessment methods for assessments of multiple health effects.

Trend Analysis

HazDat contains environmental contamination and other data from more than 3500 HWS or events with ATSDR activities for which ATSDR has conducted public health assessments, prepared health consultations, or provided responses to emergencies involving releases of toxic substances into community environments (*63*). HazDat also contains information abstracted from the toxicologic profiles on more than 200 substances most frequently encountered at HWS. Different types of sites are encountered in the environment, such as landfills, municipal sites, incinerators, battery recyclers, solvent recovery, wood treatment, manufacturing, mining, and smelting. Thus, HazDat can be queried for information such as the kinds of contaminants released from HWS into various environmental media, the environmental media that are most affected by these contaminants, and the pathways by which people are exposed to the contaminants. The content and scope of HazDat allow for the development of different approaches to the listing of hazardous chemicals. A trend analysis has been performed to generate a frequency listing for the chemicals found at HWS. Using trend analysis, a comprehensive frequencies list has been generated that used data on all NPL sites in HazDat (*63*). This initial list is being used as an indicator mixtures listing. This list provides chemicals most often found in various environmental media such as water, soil, or air. The chemicals are listed within each medium as two-, three-, and four-component mixtures. Plans are to create specific lists for various types of sites. Such lists can be refined through input on toxicity, concentration, and pathway completion to produce a priority mixtures list. Before such lists are finalized, potential for interaction can also be factored. Priority mixtures can be used for further research to understand the toxicology of chemical mixtures, including quantitative modeling of health effects and further experimental testing.

Assessment

Inherent in the data analysis component of the mixtures program is the process of hypothesis generation. When information is available on component chemicals and chemical mixtures, this systematic evaluation of data would permit the generation of hypotheses that can be tested and the identification of research needs. For individual chemicals the available literature on toxicity, mechanism of action, and interactions with other chemicals is critically reviewed, evaluated, and summarized in the toxicologic profiles. Likewise it is important to conduct similar reviews, evaluations, assessments, and summaries for priority mixtures, if data are available. As numerous studies have demonstrated, the evaluation of the joint toxicity of chemical mixtures is a formidable challenge and not a superficial modification of the single chemical methods (*65*). Those challenges to the investigator are to confront the complexities involving chemical interactions, patterns of exposure, and toxicity with well-conceived research proposals; to the clinician, to identify appropriate

biomarkers of exposure and disease resulting from low exposures to multiple chemicals; and to the health assessor, to apply biomedical judgment to the exposure scenarios at specific HWS.

Clearly, data gaps present the most immediate challenge to the health assessment and evaluation of chemical mixtures. These data gaps could exist in exposure, toxicity, or dose–response assessment aspects of the overall evaluation. In addition to performing critical reviews of available literature, hypotheses may also be formulated through the use of computational techniques such as structure–activity relationships (SAR), physiologically based pharmacokinetic (PBPK) modeling, and progressive integration of information, that is, binary-to-ternary-to-quaternary chemical mixtures (if possible).

These hypotheses will be subjected to experimental testing using *in vitro* or limited *in vivo* studies. The results of such investigations may lead to further testing, revisions in hypotheses, and/or the formulation of new hypotheses. When verified, such hypotheses would allow the development of rules that can be generalized and applied to mixtures of interest that have not been subjected to testing, cannot be subjected to testing, or used until testing data become available.

Weight-of-Evidence Methodology

The chemical mixtures toxicity assessment typically uses the hazard index approach. The intent of this approach is to approximate the toxicity index that would have been calculated had the mixture itself been tested. The critical effect for the whole mixture is rarely known. Because of resource limitations, a majority of the mixtures encountered in the environment cannot be tested.

Hence, all plausible candidates for the critical effects of the mixture are usually considered by calculating, if possible, organ-specific hazard indices. When conducting such an exercise, it is pertinent to consider the role of potential interactions that could modify the expected outcome when toxic chemicals occur together. Integration of the knowledge and insights gained about chemical interactions and the actions of the chemical components of a mixture become part of the weight of evidence for interactions (*66*).

Quantitative Modeling

Development of alternative risk assessment procedures and models is a complex data-intensive task; paucity of data is frequently the bottleneck to developing potential hazard-assessment methods or models of risk assessment of chemical mixtures such as interaction prediction. Thus, experimental research is sponsored to obtain data to elucidate toxicologic mechanisms, to better understand the molecular toxicology of chemicals, particularly their mechanisms of interaction, and to establish quantitative models. From the outset, such research should identify the following elements that could contribute to the toxicity of the chemical mixture(s): identification of multiple target organs; defining internal doses through toxicokinetics (absorption, distribution, metabolism, receptor binding, and elimination); considering all information on assessments, biomonitoring, biomarkers, adducts, and metabolites; and evaluating mechanisms of interaction, the levels at which they occur, and their significance at environmental levels. It is also important that the laboratory investigator, the model developer, and the risk assessor work in a collaborative relationship to ensure that the animals or test system, doses and dosing regimen, and other variables of the experimental procedure have been selected based on existing data and experimental design that will address existing data gaps.

Experimental Testing

In the current environment of austere resource allocations and heightened awareness of animal use in toxicologic research, more pragmatic experimental testing methods must be used without compromising the sensitivity or specificity obtained through classical methods. Efforts should take into consideration all options available, including recently developed innovative techniques. To this extent significant advances have been made in alternative toxicologic testing methods such as *in vitro* testing, PBPK modeling, and biologically based dose–response modeling. Positive correlations have been noted between *in vitro* activity and *in vivo* potency. Thus, several *in vitro* assays validated with *in vivo* studies are available to conduct toxicant interaction studies. Even though most of these tests are still in various investigatory phases, they have been studied enough to obtain initial estimates of dose–response relationships for mixtures of chemicals.

For certain specific end points of concern, it may be feasible to develop a screen of tests to study interactions using these assays. The underlying assumption is that if biologic activity in these tests is well correlated with *in vivo* toxic potency, interaction coefficients measured using such screening tests may be similarly correlated.

The plausibility of such assumptions must be further established by highly focused parallel studies involving the results from *in vitro* testing, limited animal testing, and theoretical studies to assure the inclusions of biologic and pharmacokinetic processes that are important to the *in vivo* expression of toxicity. Because many of the examples of chemical compound interactions are pharmacokinetically based, this problem may be substantially complicated for mixtures.

Nevertheless, *in vitro* test systems—particularly those that use *in vitro* metabolic activating systems—may permit screening many important combinations of chemicals, which would not otherwise be possible, especially for receptor-based or other toxicodynamic interactions. Coupled with pharmacokinetic modeling, these systems may serve as the best practical approach for detecting interactions and using this information in the health assessment process.

A selection of mixtures for the *in vitro* assays to identify a broad range of interaction coefficients could prove beneficial to the ultimate goal of predicting interactions. Based on the *in vitro* results, *in vivo* measurements could be made in a subsequent *in vivo* validation study for such mixtures that cover the same range of observed interaction coefficients. Before the results of subsequent *in vitro* assays in estimating or predicting joint effects of chemical mixtures can be used with confidence, a strong link must be established between *in vitro* and *in vivo* assay correlations and between *in vitro* and *in vivo* measured interaction coefficients.

Based on the advances in our understanding of the mechanisms of toxicity, an experimental parallelogram design has been recently advanced that allows carefully planned and goal-oriented research to be conducted (Figure 5). This design allows multispecies comparisons as well as extrapolation to human populations. As a first step, *in vitro* rodent species bioassays (rats, mice, or hamsters) are conducted to test the toxic effects of chemicals. Results from such studies are validated in *in vivo* rodent studies. These studies are then followed with *in vitro* studies with human systems (e.g., human cell lines) to evaluate the findings of the animal studies. Thus, the *in vitro* rodent studies are used to confirm the initial *in vivo* rodent findings and these results are subsequently confirmed *in vitro*

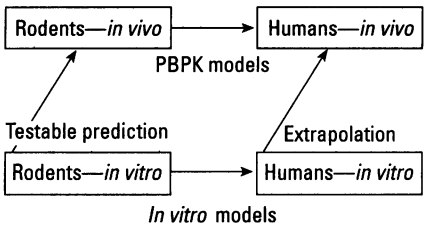

Figure 5. Parallelogram for *in vitro–in vivo* extrapolation between animals and humans. Adapted from Himmelstein et al. (*64*).

in human systems. The information thus generated is used to extrapolate the potential effects of the chemicals to human populations. Using this design, humans have been shown to be more like rats and less like mice regarding metabolism of 1,3-butadiene (*67*). Based on this comparative data, Himmelstein et al. (*67*) suggest that because the concentration of butadiene epoxide, the active moiety, will be low in humans, the human cancer risk following exposure to butadiene will be similarly low. However, if this assessment was based only on mouse data, the risk would be incorrectly estimated as higher than the current estimate. This experimental design is also useful for the study of toxicity of chemical mixtures under various conditions of exposure.

With the intent to help support development of chemical mixtures assessment methods, the ATSDR has initiated a range of toxicity testing and research efforts in cooperation with the National Institute of Environmental Health Sciences, the private sector, and academic institutions. These activities are geared toward limited *in vivo* and *in vitro* studies using various currently available testing assays and methods. Similarly, SAR can be used to extend our range of plausible inference and extrapolation within and across chemical classes. Although no single assay screen or method is likely to provide all necessary information, such approaches in aggregate can provide significant insights about the overall public health implications of exposure to multiple chemicals.

Conclusion

Because site interventions ranging from education of people regarding their risks to site remediation require extensive time and human and monetary resources, it is imperative that the risks at HWS be accurately characterized. Agencies are increasingly using a decision framework that involves risk-based priorities with the objective of implementing prevention measures to protect the people potentially exposed to hazardous materials (*68*). However, once the hazards have been accurately characterized, it is equally important that risks to public health be skillfully communicated. This phase of the intervention process is pivotal because the perception of risk is often not consistent with the real risks to public health (*69*).

A critical element in producing accurate health assessments is the availability of relevant data. Often the public health assessment process is forced to rely on limited availability of adequate scientific information. Thus, in the public health assessment process an evaluation of complex exposures may devolve into an evaluation based on data derived from studies entailing time-weighted average exposures to single chemicals by single routes and short-term exposure scenarios (*70*). Such evaluations are not able to address the potential for chemical interactions and the aggregate contribution to body burdens. Hence, ATSDR research is focused on the reduction of uncertainties resulting from the absence of critical data. An array of innovative tools that have been used successfully in pharmacology and medicine are increasingly finding acceptance in the health risk assessment community. These tools may allow for accurate assessments of the potential for adverse health effects from exposure to environmental chemicals and their mixtures, thereby leading to a reduction of uncertainty in the risk characterization process. Tools such as SAR modeling and PBPK/pharmacodynamic modeling can find utility in the decision-making process and the performance of risk characterization. The ATSDR is supporting work at a number of research and educational institutions as well as collaborating with industry to develop these methods into useful tools in the assessment of chemical mixtures and to facilitate their use in the mainstream of public health practice.

REFERENCES AND NOTES

1. Newill VA. Keynote address: significance of risk assessment in the management of environmental exposures of chemical mixtures. Toxicol Ind Health 5(5):635 (1989).
2. Mumtaz MM, McKean DL, Bruins RJ, Schoeny RS, De Rosa CT. Research strategy for risk characterization of complex exposures. In: Proceedings of the Fourth International Conference on the Combined Effects of Environmental Factors. Baltimore, MD:John Hopkins University Press, 1991;15–21.
3. De Rosa CT, Mumtaz MM, Choudhury H, McKean DL. An integrated approach to risk characterization of multiple pathway chemical exposures. In: Comparative Environmental Risk Analysis (Cothern CR, ed). Boca Raton, FL:Lewis Publishers, 1991;165–175.
4. Johnson BL, De Rosa CT. Human health implications of hazardous wastes. Eur J Oncol (in press).
5. U.S. Congress. Comprehensive Environmental Response, Compensation, and Liability Act. 42 U.S.C. 9604 (104)(i)(5)A. Washington:U.S. Government Printing Office, 1996.
6. ATSDR. Agency Profile and Annual Report. Atlanta, GA:Agency for Toxic Substances and Disease Registry, 1996.
7. Gilbertson M. Guest commentary. Great Lakes Res Rev 1(2):3–4 (1995).
8. Grisham J, ed. Health Aspects of the Disposal of Waste Chemicals. New York:Pergamon Press, 1986.
9. Griffith J, Duncan R, Riggan W, Pellom A. Cancer mortality in U.S. counties with hazardous waste sites and ground water pollution. Arch Environ Health 44:69 (1989).
10. Haschek WM, Witschi HR. Respiratory system. In: Handbook of Toxicologic Pathology (Haschek WM, Rousseaux CG, eds). New York:Academic Press, 1991;790.
11. American Thoracic Society. Guidelines as to what constitutes an adverse respiratory health effect, with special reference to epidemiologic studies of air pollution. Am Rev Respir Dis 131:666–668 (1985).
12. De Rosa CT, Dourson ML, Osborne R. Risk assessment initiatives for noncancer endpoints: implications for risk characterization of chemical mixtures. Toxicol Ind Health 5(5):805–824 (1989).
13. National Research Council. Pesticides in the Diets of Infants and Children. Washington:National Academy Press, 1993;42.
14. WHO. Principles for Evaluating Health Risks from Chemicals During Infancy and Early Childhood: The Need for a Special Approach. Environmental Health Criteria 59. Geneva:World Health Organization, 1986.
15. Bearer CE. Environmental health hazards: how children are different from adults. Crit Issues Children Youths 5(2):11–26 (1995).

16. National Research Council. Pesticides in the Diets of Infants and Children. Washington:National Academy Press, 1993;43.
17. Mushak P, Davis JM, Crocetti AF, Grant LD. Prenatal and postnatal effects of low-level exposure: integrated summary of a report to the U.S. Congress on childhood lead poisoning. Environ Res 50:11–36 (1989).
18. Berry M, Bove F. Birth weight reduction associated with residence near a hazardous waste landfill. Environ Health Perspect 105(8):856–861 (1997).
19. Weisglas-Kuperus N, Sas TCJ, Koopman-Esseboom C, van der Zwan CW, De Ridder MA, Beishuizen A, Hooijkaas H, Sauer PJ. Immunologic effects of background prenatal and postnatal exposure to dioxins and polychlorinated biphenyls in Dutch infants. Pediatr Res 38:404–410 (1995).
20. ATSDR. Guidance for Developing Toxicological Profiles. Atlanta, GA:Agency for Toxic Substances and Disease Registry, 1995.
21. Manson JM, Wise LD. Teratogens. In: Casarett and Doull's Toxicology. The Basic Science of Poisons (Amdur MO, Doull J, Klaassen CD, eds). New York:Pergamon Press, 1991.
22. Chen PH, Hsu ST. PCB poisoning from toxic rice-bran oil in Taiwan. In: PCBs and the Environment. Vol 3 (Waid JS, ed). Boca Raton, FL:CRC Press, 1986;27–38.
23. Kashimoto T, Miyata H. Differences between Yusho and other kinds of poisoning involving only PCBs. In: PCBs and the Environment. Vol 3 (Waid JS, ed). Boca Raton, FL:CRC Press, 1986;2–26.
24. Kuratsune M. Yusho, with reference to Yu-Cheng. In: Halogenated Biphenyls, Terphenyls, Naphthalene, Dibenzodioxins and Related Products. 2nd ed (Kimbrough RD, Jensen AA, eds). Amsterdam:Elsevier Science Publishers, 1989;381–400.
25. Okumura M. Past and current medical states of Yusho patients. Am J Ind Med 5:13–18 (1984).
26. Rogan WJ. Yu-Cheng. In: Halogenated Biphenyls, Terphenyls, Naphthalenes, Dibenzodioxins and Related Products. 2nd ed (Kimbrough RD, Jensen AA, eds). Amsterdam:Elsevier Science Publishers, 1989; 401–415.
27. Yoshimura T. Epidemiological study on Yusho babies born to mothers who had consumed oil contaminated with PCB. Fukuoka Igaku Zasshi 65:74–80 (1974).
28. Bakir F, Damluji SF, Amin-Zaki L, Murtadha M, Khalidi A, al-Rawi NY, Tikriti S, Dalahir HI, Clarkson TW, Smith JC, et al. Methylmercury poisoning in Iraq. Science 1181(96): 230–241 (1973).
29. Amin-Zaki L, Elhassani SB, Majeed MA, Clarkson TW, Doherty RA, Greenwood M. Intra-uterine methyl mercury poisoning in Iraq. Pediatrics 84:587–595 (1974).
30. Marsh DO, Myers GJ, Clarkson TW, Amin-Zaki L, Tikriti S, Majeed MA, Dabbagh AR. Dose-response relationship for human fetal exposure to methylmercury. Clin Toxicol 10:1311–1318 (1981).
31. Marsh DO, Clarkson TW, Cox C, Myers GJ, Amin-Zaki L, Tikriti S. Fetal methylmercury poisoning: relationship between concentration in single strands of hair and child effects. Arch Neurol 44:1017–1022 (1987).
32. Harada H. Congenital Minamata disease: intrauterine methylmercury poisoning. Teratology 18:285–288 (1978).
33. ATSDR. Toxicological Profile for Lead. Atlanta, GA:Agency for Toxic Substances and Disease Registry, 1993.
34. U.S. EPA. Air Quality Criteria for Lead. EPA 600/8–83-028F. Research Triangle Park, NC:U.S. Environmental Protection Agency, 1986.
35. Geschwind S, Stolwijk J, Bracken M, Fitzgerald E, Stark A, Olsen C, Melius J. Risk of congenital malformations associated with proximity to hazardous waste sites. Am J Epidemiol 135:1197–1207 (1992).
36. Marshall EG, Gensburg LJ, Greary NS, Deres DA, Cayo MR. Analytic Study To Evaluate Associations Between Hazardous Waste Sites and Birth Defects. Albany, NY:New York State Health Department, 1995.
37. Croen LA, Shaw GM, Sanbonmatsu L, Selvin S, Buffler PA. Maternal residential proximity to hazardous waste sites and risk for selected congenital malformations. Epidemiology 8(4):347–354 (1997).
38. Berry M. Lapari Landfill Birth Weight Study: A 25-year Trend Analysis. Trenton, NJ:New Jersey Department of Health, 1994.
39. ATSDR. Volatile Organic Compounds in Drinking Water and Adverse Pregnancy Outcomes (U.S. Marine Corps Base Camp Lejeune, North Carolina). Atlanta, GA:Agency for Toxic Substances and Disease Registry, 1997.
40. Bove FJ, Fulcomer MC, Klotz JB, Esmart J, Dufficy EM, Savrin JE. Public drinking water contamination and birth outcomes. Am J Epidemiol 141(9):850–862 (1995).
41. Johnson BL. Present to Subcommittee on Commerce, Trade, and Hazardous Materials. U.S. House of Representatives, Committee on Commerce, 104th Congress. 23 May 1995.
42. Campbell A, Converse PE, Rodgers WL. The Quality of American Life: Perceptions, Evaluations, and Satisfactions. New York:Russell Sage Foundation, 1976.
43. Katz S. The science of quality of life. J Chron Dis 40:459–463 (1987).
44. Miettinen. OS. Quality of life from the epidemiologic perspective. J Chron Dis 40:641–643 (1987).
45. ATSDR. Unpublished data.
46. National Research Council. Animals as Sentinels of Environmental Health Hazards. Washington:National Academy Press, 1991;19.
47. National Research Council. Animals as Sentinels of Environmental Health Hazards. Washington:National Academy Press, 1991;80.
48. ATSDR. Public Health Assessment Guidance Manual. Atlanta, GA:Agency for Toxic Substances and Disease Registry, 1992;6–29.
49. U.S. EPA. Risk Assessment Guidance for Superfund. Vol II: Environmental Evaluation Manual, Interim Final. EPA 540/1-89/001. Washington:U.S. Environmental Protection Agency, 1989.
50. U.S. EPA. Framework for Ecological Risk Assessment. EPA/630/R-92/001 Washington:U.S. Environmental Protection Agency, 1992.
51. OSHA. General Industry Air Contaminants Standard (29 CFR 1910.1000). Washington:Occupational Safety and Health Administration, 1993.
52. Finkel A, Thomas R, Rodricks J. Science and judgement in risk assessment: an overview of the National Academy of Sciences' committee on risk assessment for hazardous air pollutants report. In: Proceedings ILSI Risk Science Institute, Brookings Institute Seminar. Fairfax, VA:CASET Associates, 1994;17.
53. U.S. EPA. Toxics Release Inventory. Washington:U.S. Environmental Protection Agency, (1995).
54. Loomis DP, Borja-Aburto VH, Bangdiwala SI, Shy CM. Ozone Exposure and Daily Mortality in Mexico City: A Time-Series Analysis. Research Rpt No 75. Cambridge, MA:Health Effects Institute, 1996.
55. Folinsbee LJ, McDonnell WF, Horstman DH. Pulmonary function and symptom responses after 6.6 hour exposure to 0.12 ppm ozone with moderate exercise. J Air Pollut Control Assoc 38(1):28–35 (1988).
56. Fouke JM, Delemos ER, McFadden ER. Airway response to ultra short-term exposure to ozone. Am Rev Respir Dis 137:326–330 (1988).
57. McDonnell WF, Horstman DH, Hazucha MJ, Haak SE. Respiratory response of humans exposed to low levels of ozone for 6.6 hours. Arch Environ Health 46:145–150 (1991).
58. NIOSH. National Occupational Research Agenda. NIOSH Publ No 96-115. Washington:National Institute for Occupational Safety and Health, 1996.
59. Fay RM, Mumtaz MM. Development of a priority list of chemical mixtures occurring at 1188 hazardous waste sites, using HazDat database. Food Chem Toxicol 34:1163–1165 (1996).

60. De Rosa CT, Johnson BL, Fay M, Hansen H, Mumtaz MM. Public health implications of hazardous waste sites: findings, assessment and research. Food Chem Toxicol 34:1131–1138 (1996).
61. Mumtaz MM, Neft NE, Lewis CR, Lichtveld MY. The public health impact of chemicals and chemical mixture by-products at hazardous waste sites. In: The Proceeding of the International Congress on the Health Effects of Hazardous Waste. Princeton, NJ:Princeton Scientific Publishing, 1994;508–516.
62. ATSDR. Public Health Assessment Guidance Manual. Atlanta, GA:Agency for Toxic Substances and Disease Registry, 1992;6–31.
63. ATSDR. HazDat. Hazardous Substances Database. Alanta, GA:Agency for Toxic Substances and Disease Registry, 1997.
64. ATSDR. 1997 CERCLA Priority List of Hazardous Substances That Will Be the Subject of Toxicological Profiles and Support Document. Atlanta, GA:Agency for Toxic Substances and Disease Registry, 1997.
65. Yang RSH. Introduction to the toxicology of chemical mixtures. In: Toxicology of Chemical Mixtures (Yang RSH, ed). New York:Academic Press, 1994;1–10.
66. Mumtaz MM, Durkin PR. A weight-of-evidence approach for assessing interactions in chemical mixtures. Toxicol Ind Health 8(6):377–406 (1992).
67. Himmelstein MW, Bahman A, Turner MJ, Medinsky MA, Bond JA. Interspecies differences in 1,3-butadiene metabolism. CIIT Act 14(8):1–8 (1994).
68. U.S. EPA. Reducing Risks: Safety Priorities and Strategies for Environmental Protection. Washington:U.S. Environmental Protection Agency, 1990;13–14.
69. Carnegie Commission. Risk and the Environment, Improving Regulatory Decision Making: A Report of the Carnegie Commission on Science, Technology, and Government. New York:Carnegie Commission, 1993;92.
70. Mumtaz MM, Cibulas W, De Rosa CT. An integrated framework to identify significant human exposures (SHELs). Chemosphere 31(1):2485–2498 (1995).
71. Chou SJ, Holler J, De Rosa CT. Minimal risk levels (MRLs) for hazardous substances. J Clean Technol Occup Med 7(1):1–24 (1998).