Chapter 16
Consideration of Physical Stressors in Cumulative Risk Assessment

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Abstract Physical stressors represent an important class of factors that can affect the health of humans or ecosystems and should be considered in cumulative risk assessment. Physical stressors are defined here as biological agents (e.g., bacteria, viruses) or external forces (e.g., radiation, noise) that can modify exposure and/or elicit a physiological response from the exposed organism. Physical stressors can intersect with chemical stressors in at least three ways: (1) by directly interacting with chemicals to modify exposure (e.g., photoreactions of sunlight with air pollution), (2) by interacting with the same target system as a chemical stressor to elicit joint effects (e.g., noise and chemicals can both affect the physiological mechanism leading to hearing disorders), and (3) by interacting with the target system to alter its susceptibility or response to chemical exposure (e.g., virus-initiated disease leading to hyper-responsiveness to chemical insult). In this chapter, physical stressors will be discussed in terms of their actions on biological systems, modification of exposure or effects of chemical stressors, and suggestions for incorporation into cumulative risk assessment.

Keywords Sunlight · Heat · Pathogens · Noise · Nonchemical stressors · Biological stressor

Disclaimer The findings and conclusions in this report are those of the authors and do not necessarily represent the views of the U.S. National Institute of Occupational Safety and Health.

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16.1 Introduction

The inclusion of both chemical and nonchemical stressors in human health risk assessments is required to better reflect real-world exposures and protect vulnerable populations that face disproportionate exposure to multiple stressors (Gallagher et al. 2015; Sexton 2012). This represents a significant paradigm shift from previous human health risk assessments, which overwhelmingly addressed single chemicals and only occasionally included clearly defined classes of chemicals (e.g., organophosphates (U.S. EPA 2002a)). Although there are an increasing number of examples of cumulative risk assessment efforts that include nonchemical stressors (Fox et al. 2017), challenges remain in their successful incorporation. In contrast, ecological risk assessments often include consideration of nonchemical stressors in addition to chemical stressors, albeit with a focus on protection at a population level, not an individual level (Heugens et al. 2001; U.S. EPA 1984).

The term "nonchemical stressors" is broad and includes physical factors (biological agents or external forces), as well as psychosocial influences that can modify exposure, susceptibility, or response to chemical stressors. Whereas physical stressors are often amenable to quantitative measurement (e.g., virus titers, radiation intensity), animal and mechanistic studies, and tend to have a direct impact on chemical exposure or biological response; psychosocial stressors involve perception and often have a more nuanced role in influencing biological responses to chemical stressors. Therefore, physical stressors could offer a promising starting place for including nonchemical stressors in cumulative risk assessment. Cumulative risk assessment refers to the process of estimating the risk associated with exposure to more than one stressor. In this chapter, the focus is on incorporating physical stressors into component-based cumulative risk assessment approaches (see Chap. 14). Despite the complexity involved, psychosocial stressors should also be accounted for in cumulative risk assessment and will be covered in detail in later chapters (see Chaps. 17 and 18).

Although physical stressors have not routinely been included in cumulative risk assessments, there are some important historical examples of the consideration of physical and chemical stressor interactions and their impact on human health. One such example can be found in combined radiation and chemotherapy (Rubin 1977). In a 1977 paper reviewing the topic, Rubin suggests that quantitative dose-response relationships in animal models are needed for radiation in order to protect against unnecessary long-term harm from combined radiation/chemotherapeutic treatment (Rubin 1977). A second classic example illustrating the need for considering physical stressors in cumulative risk is that of radon and smoking and their combined effects on lung cancer (Reif and Heeren 1999).

There are many, often complicated, ways for physical stressors to modify the toxicity of chemical stressors (Fig. 16.1), and vice versa. As illustrated in Fig. 16.1, physical stressors can interact with chemical stressors at the level of exposure, target, or biological response. Indeed, a single physical stressor can interact at multiple levels. For example, sunlight can modify a chemical prior to inhalation

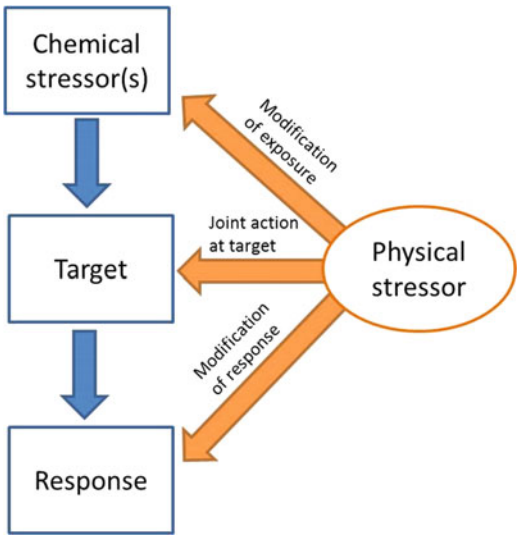

Fig. 16.1 Graphic depiction of the different mechanisms by which physical stressors can impact chemical stressor exposures and responses

exposure (see discussion below), contribute to phototoxicity of an orally administered chemical at a dermal target site, and elicit damage to skin, which could, in turn, alter absorption of or response to concurrent dermal chemical exposures. It is important to emphasize that chemicals can also modify exposure and/or response to physical stressors.

This chapter will discuss a range of physical stressors in terms of quantifying their toxicity, identifying interactions with chemical stressors, and incorporating them into cumulative risk assessment. Some examples of physical stressors are provided in Table 16.1. They fall into two major categories – biological agents and external forces. In general, the biological agents exhibit indirect effects by influencing the health status of individuals and thereby altering their response to chemical stressors (e.g., bacteria and viruses interacting with chemical stressors). The second major category is forces, which can interact directly and indirectly at both the exposure and response levels. Therefore, several different forces (e.g., sunlight, heat, noise) will be discussed to demonstrate potential issues to consider in accounting for the impact of these stressors on cumulative risk. Although there is a history of including some physical stressors in ecological risk assessment, the focus of this chapter will be on human health.

Table 16.1 Examples of potential physical stressors to human health

Biological agents/factors	Forces
Viruses	Electromagnetic radiation
	Radio waves
	Visible light
	Ultraviolet light (ionizing)
	X-rays (ionizing)
Pathogenic bacteria	Particle radiation
	α-Radiation
	β-Radiation
	γ-Radiation (ionizing)
	Neutron radiation
Allergens – pollen, dander, dust mites	Acoustic radiation
	Ultrasound
	Sound
Physical health status	Thermal radiation (heat)
Nutritional status	
Disease status	
Microbiome	
Lack of sleep	
Hypothermia	

16.2 Biological Agents: Microbial Disease and Chemical Stressors

As mentioned in the introduction, biological agents or factors interact indirectly with chemicals by changing the health status of the target subject and potentially increasing their susceptibility to chemical injury. Alternatively, chemical exposures can affect health status leading to a modified response following exposure to the biological agent. The diseases induced by different biological stressors (e.g., lack of sleep versus allergens) are varied. However, many biological stressors ultimately involve immune system modulation (Jain and Walker 2015). Microbial diseases are perhaps the most well-studied biological agents within the physical stressor spectrum and will be used to illustrate the process of exploring potential interactions with chemical stressors.

There are at least four types of mechanisms that underlie potential interactions between chemical stressors and infectious disease. The best understood is suppression of immune responses, resulting in increased incidence and/or severity of infectious disease. Alternatively, certain immune/inflammatory mediators that are activated during infection affect metabolic enzymes and transporters, resulting in increased chemical toxicity. Chemical exposure may enhance inflammation and immune pathology associated with an infection. Conversely, infection may enhance chemical-induced lesions (e.g., p53 mutations, inflammation, cell proliferation).

These mechanisms are not necessarily comprehensive, distinct, or mutually exclusive. However, they provide a starting place for evaluating interactions between infectious disease and chemical stressors. Each mechanism will be discussed in more detail below with the goal of illustrating options for incorporating biological stressors into cumulative risk assessments.

Substantial data from animal models, and more limited data in humans, suggest that a number of chemicals suppress a variety of immune responses (Selgrade 2010), which can lead to increased risk of bacterial infection. For example, select air pollutants (e.g., chloroform, toluene, ozone) have been demonstrated to decrease the function of alveolar macrophages, which are the first line of defense against some bacterial species (e.g., *Streptococcus zooepidemicus*) (Selgrade and Gilmour 2006). Research on this model provides both qualitative and quantitative approaches to describe the associated risk. Data comparing human and murine alveolar macrophages exposed to similar doses of ozone in vitro and in vivo indicate that cells from both species respond almost identically as measured by macrophage phagocytic capability. Thus, these data suggest that (1) the effects of ozone exposure on murine alveolar macrophage function are predictive of effects on human alveolar macrophage function and (2) effects of in vitro exposure of macrophages to ozone are predictive of effects that result from in vivo exposure (Selgrade et al. 1995). Application of inhalation dosimetry methods eliminates the need for the uncertainty factor that is typically applied to account for animal to human extrapolation in the absence of toxicokinetic data (U.S. EPA 2012). Furthermore, the lack of difference in sensitivity observed in studies with mice and humans eliminates the need to apply a factor for toxicodynamics. In addition to air pollution and alveolar macrophage function that demonstrate at least a qualitative relationship between immune function and disease, developmental exposures to arsenic, polychlorinated biphenyls, and cigarette smoke also have been linked to immune suppression. In the cases of arsenic (Soto-Pena et al. 2006) and PCBs (Heilmann et al. 2010), a quantitative relationship exists between exposure to the chemical and suppression of the immune system in humans. However, predicting the impact of immune suppression on the incidence or severity of infection in a population (i.e., risk) is difficult. Immunocompetence (i.e., the ability to mount a normal immune response) in a population may be represented as a bell-shaped curve including individuals with little or no immune reserve (response capacity) available (e.g., the very young and old and those who are immunocompromised by disease or medications) and very robust individuals. The proportion of the population at risk of infection depends on the level of immune competence, as well as the dose and virulence of infectious agents, with increasing risk of infection corresponding to increasing dose or virulence. In populations exposed to immunosuppressive agents, the distribution curve would be expected to shift, putting a larger portion of population at risk for disease development.

Although the effects of toxicants on host defenses against infection have received the most attention, it is also possible for infections to affect host defenses against toxicants, by interfering with metabolic enzymes and transporters. There are many examples of increased chemical/drug toxicity with infections or other

inflammation-related diseases (Morgan et al. 2008). For example, an influenza epidemic resulted in decreased clearance of theophylline, an asthma medication with a narrow therapeutic window, resulting in toxicity in children (Kraemer et al. 1982). Interestingly, many other factors can influence theophylline dose, including smoking, drugs (e.g., phenobarbital, erythromycin), and disease (e.g., heart failure, liver disease) (Kraemer et al. 1982). Other examples involve murine cytomegalovirus infection increasing the toxicity of parathion (Selgrade et al. 1984) and sodium pentobarbital-induced sleeping time and decreasing cytochrome P450 (CYP) levels in liver microsomes in mice (Catignani et al. 1989). In multiple tissue types, infection and inflammatory diseases have demonstrated downregulation of ATP binding-cassette (ABC) drug transporters involved in cellular efflux of xenobiotics (Petrovic et al. 2007). The proposed pathways underlying these effects begin when infections and other inflammatory stimuli cause the release of inflammatory cytokines from monocytes, macrophages, and stromal cells (the acute phase response), resulting in the modulation of transcription factor activities in the liver. These changes ultimately lead to a downregulation of CYP and ABC transporter genes. The production of cytokines also activates nitric oxide synthase 2 to form nitric oxide that inhibits CYP enzyme activities directly and/or leads to the downregulation of CYP proteins via destabilization (Morgan et al. 2008). Risk assessment procedures account for these enzyme-related changes by applying a tenfold intraspecies uncertainty factor (U.S. EPA 2002b). This uncertainty factor is meant to capture differences in individual susceptibility within the population. However, use of a default uncertainty factor does not accurately reflect what is known about the effects of infection/inflammation on chemical toxicity. As risk assessments begin to use adverse outcome pathways to characterize the risk associated with multicomponent mixtures (see Chap. 7), infection and inflammation may be incorporated into that process.

The third type of interaction between chemicals and infection involves chemical-mediated exacerbation of inflammation and pathology resulting from infection. Examples include effects on influenza infection by ozone (Selgrade et al. 1988), ultraviolet radiation (Ryan et al. 2000, 2002), TCDD (Burleson et al. 1996; Lawrence and Vorderstrasse 2004; Warren et al. 2000), and acrolein (Ong et al. 2012). In all cases, mortality is enhanced by exposure to the toxicant without increased viral load. Although all of these chemicals have immunosuppressive potential, reduced viral clearance resulting from immune suppression does not appear to be responsible for the increase in mortality. Instead, morbidity and mortality occur very early in infection before involvement of adaptive immunity, and surviving mice develop protective immunity that prevents subsequent reinfection (Lawrence and Vorderstrasse 2004; Ryan et al. 2000). Increased inflammatory responses appear to be responsible for observed mortality (Head and Lawrence 2009). Both pathogens and tissue damage trigger similar receptors and signaling pathways that lead to innate inflammatory responses (Kono and Rock 2008; Tolle and Standiford 2013). A systems biology approach that integrates these triggers at the pathway level is needed to account for the joint effects of these immune system modulators.

The fourth, and final, type of interaction involves enhancement of chemical induced lesions (e.g., p53 mutations, inflammation, and cell proliferation) by infection. This interaction might explain the joint effects of hepatitis B virus infection and aflatoxin on liver cancer (Kensler et al. 2010). In nested, case-control data within a cohort study of 18,000 men in Shanghai, Qian et al. (1994) demonstrated a statistically significant increase in the relative risk (95% confidence limits) of 3.4 (1.1, 10) for hepatocellular carcinoma cases with detectable urinary biomarkers for aflatoxin, 7.3 (2.2, 24.4) for individuals without evidence of aflatoxin exposure but seropositive for hepatitis B antigen, and 59.4 (16.6, 212.0) for individuals exhibiting both urinary aflatoxin markers and positive hepatitis B status. The results strongly suggest an interaction between aflatoxin and hepatitis B in the development of hepatocellular carcinoma. Hepatitis B infection and the resulting chronic inflammation may promote DNA lesions leading to P53 mutations and may promote cell proliferation, contributing to chronic hepatitis and/or cirrhosis and ultimately carcinoma. It is plausible that similar interactions may exist between other infections and toxicants that target the liver. Again, as we begin to use systems biology to work through cumulative risk, an understanding of the pathways underlying this interaction could be applied. However, in this instance, decisions to decrease risk by promoting public health interventions such as limiting exposure to aflatoxin and vaccinating against hepatitis B are recommended.

In summary, the chemical/infection interactions described here involve joint action of chemicals and immune/inflammatory responses (the biological forces). Superimposed on all of this are genetic differences which affect both susceptibility to infection and toxicity. Existing information regarding molecular pathways involved in immune activation and inflammation should be applied using a systems approach to understand the cumulative risk that results from exposure to chemicals and infectious agents.

16.3 Forces: Modification of Exposure by Sunlight

As discussed in the previous section exploring microbial disease, there are multiple pathways by which a physical force can interact with chemicals to affect health. In the case of sunlight, there is a clear primary target – the skin – which is subject to direct damage, leading to aging and cancer of the skin or photodermatosis (immune reaction to sunlight). It follows that chemicals that elicit skin toxicity could interact with sunlight to increase skin damage. While acknowledging that there are many opportunities for sunlight to interact with chemicals at a common target site, the focus of this section is not on interaction of sunlight and chemicals at the adverse outcome level but at the exposure level.

Sunlight, along with other climatic characteristics (e.g., temperature, humidity), has the potential to modify both the concentration and form of chemicals present in the air. The criteria air pollutants (ozone, particulate matter, carbon monoxide, nitrogen oxides, sulfur dioxide, and lead) are of particular interest, as these are the

six common air pollutants for which EPA is mandated by the Clean Air Act to set National Ambient Air Quality Standards. Sunlight together with temperature can trigger photochemical reactions of air pollutants. Examples of these reactions include production of ozone from hydrocarbons and nitrogen oxides, nitrogen dioxide from oxidation of nitrogen oxide, carbon monoxide from oxidation of hydrocarbons, and many well-known toxic compounds such as formaldehyde, acetaldehyde, acrolein, and other carbonyl and nitrate-containing products from oxidation of hydrocarbons and nitrogen oxides, as well as reduction of primary emitted pollutant concentrations (Finlayson-Pitts and Pitts 1999). In addition, these reactions contribute to air pollution in the form of secondary organic aerosols (SOA), which are present in fine particulate matter (PM).

Atmospheric transformation processes affect the relative composition and resulting cumulative health effects of contaminants, including criteria pollutants. It is important to understand how atmospheric transformations affect air pollution mixtures and PM composition and resulting toxicological risk. Evidence of the importance of these issues can be found in the ongoing efforts at the U.S. Environmental Protection Agency to develop a framework for addressing multipollutant risk assessment (Johns et al. 2012).

In terms of research into atmospheric pollutant mixtures, smog chambers have been used to prepare consistent, controlled mixtures of various primary pollutants to study how different conditions (e.g., natural or simulated sunlight, different temperatures, or humidity levels) change mixture composition. Smog chambers facilitate the study of sunlight and temperature effects in the absence of interference from changing weather patterns and unexpected emissions from nontarget sources. They can interface with in vitro or in vivo models to provide direct exposure in toxicity studies (e.g., direct air-to-tissue or air-liquid-interface inhalation exposures) (Lichtveld et al. 2012). This method avoids pre-collection with filters and liquids, thereby offering a significant advantage over methods that require sample preparation, which can alter component concentration ratios and toxicological responses (Lichtveld et al. 2012). There are many examples of photochemical experiments using smog chambers, such as industrial compounds and nitrogen oxide mixtures and complex mixtures of motor vehicle exhaust in urban atmospheres. These experiments often demonstrate enhanced toxicity following photochemical reactions, as measured by markers of inflammation and other biological endpoints (e.g., cytotoxicity) (Doyle et al. 2007). Confirmatory experiments can be conducted with observed secondary products to link particular species with effect. More recently, Gas In Vitro Exposure Systems (GIVES) have been used in the field to expose cells directly to ambient air (Vizuete et al. 2015). Following field exposure, cells can be evaluated for cytotoxicity and gene expression changes (Vizuete et al. 2015).

Modification of various parameters (e.g., pollutant sources, mixture composition, component concentrations, or atmospheric conditions) can also aid in interpretation of the mechanism or mode of action of air pollution determined through toxicity testing. For example, a primary pollutant mixture representing the average volatile organic compounds (VOCs) observed in 40 U.S. cities can significantly

change after 1 day of "aging," which could affect ozone concentrations, presence of co-pollutants, and toxicity. The degree of aging can be influenced by meteorological conditions (e.g., cloud cover, sunlight intensity) (Sexton et al. 2004). Based on this type of study, models can be developed to predict ozone concentration (Sexton et al. 1988). Information gained from these approaches can be used to identify toxic secondary products, which can be included in air quality simulation models for multipollutant risk assessments.

Cytotoxicity and inflammation markers are endpoints that have been used to explore the effects of sunlight and temperature on atmospheric transformations of air pollutants (Lichtveld et al. 2012; Sexton et al. 2004). However, numerous biological models and endpoints could be used in the same context. For example, novel genomic analyses of cell-based exposure to an urban mixture demonstrated transcriptional changes in a subset of genes, with increased expression alterations resulting from mixture irradiation (19–709 following a 1-day sunlight irradiation) (Rager et al. 2011). This type of study offers promise for elucidating the effects of atmospheric conditions on complex mixtures and health. Additionally, biomarkers identified in in vitro studies could be explored for their utility in an epidemiological context.

Sunlight and temperature can influence the toxicity of air pollutants. Fortunately, both of these physical stressors can be easily quantified (Jeffries et al. 1989) and incorporated into air quality simulation models to estimate air pollution exposure concentrations and distribution in a target area (Vizuete et al. 2010). Results from these simulation models can be used to estimate total exposure within a population and integrated with health information (e.g., excess deaths) (Li et al. 2010). In order to better characterize risk associated with ambient exposures, studies that incorporate sunlight and temperature should be considered. These studies capture potential transformations of primary pollutants and resulting changes in their toxicity. Without consideration of these processes, there is the potential to underestimate risk from exposure to air pollution mixtures.

16.4 Forces: Heat and Chemicals

Increasing global temperatures associated with climate change have focused attention on potential health effects associated with heat (Spector and Sheffield 2014; Patz et al. 2014; Kovats and Hajat 2008). Thermal stress encompasses temperatures that fall above (heat stress) or below (cold stress) the normal range and require a physiological response in order to maintain homeostasis of the internal body temperature (Wilson et al. 2014). While both heat stress and cold stress can impact health, heat stress will be used as an illustrative example. Heat is a complex actor – it can have a direct effect on health (e.g., heat strain, heat stroke) and exacerbate existing disease conditions both alone and in concert with chemicals, and it can interact with chemicals by modifying their absorption or effect.

A complicating factor in the study of heat stress is the differences in thermoregulation strategies between small rodents typically used in toxicity studies and humans (Gordon et al. 2014). For example, mice and rats rely on a metabolic strategy to balance heat loss and production, while humans depend more on regulation of peripheral blood flow (Gordon et al. 2014). Furthermore, laboratory animal studies are typically conducted at temperatures that are below ideal ambient temperatures for rodents, causing a mild hypothermic response that may distort their response to chemical exposure (Gordon et al. 2014). Fortunately, many epidemiology studies have addressed the role of heat in disease and will be the focus of this section.

A wide array of diseases are associated with heat stress including mental health disorders (Berry et al. 2010), reproductive system dysfunction (Strand et al. 2012), kidney disease (Tawatsupa et al. 2012; Garcia-Trabanino et al. 2015), cardiovascular disease (Braga et al. 2002; Schwartz et al. 2004), and respiratory disease (Braga et al. 2002; Michelozzi et al. 2009). It follows that heat stress has the potential to act together with chemicals and other nonchemical stressors to disrupt normal function or exacerbate disease. Incorporating heat stress into cumulative risk assessment could be motivated by co-occurrence, as in the case of agricultural workers that are occupationally exposed to both heat stress and pesticides (see case study below). Alternatively, for an assessment aimed at evaluating the cumulative risk of exposures contributing to an observed disease (effects-based risk assessment or disease-based risk assessment), heat stress could be included when it is identified as a risk factor. If heat stress is identified as a risk factor that should be included in a cumulative risk assessment, quantifying the contribution of heat stress is the next goal.

The two internationally recognized methods for rating the level of heat stress are the Wet Bulb Globe Temperature (WBGT) index (ISO 7243) and the Predicted Heat Strain (PHS) model (Alfano et al. 2014). The method for assessing the WBGT index is currently undergoing revision, but generally a combination of air temperature, black globe temperature, and natural wet bulb temperature is used to approximate heat stress. For a detailed review of the history and limitations of this method, see Budd (2008). As recommended in Budd (2008), plotting the WBGT against an adverse effect to generate a "dose"-response relationship would allow for an assessment of the heat stress dose response. Examples of heat stress dose response include heat exhaustion (Yaglou and Minard 1957) (Fig. 16.2) and heat stroke (Schickele 1947). Unfortunately, this type of dose-response data will not be available for every endpoint of interest. However, data in the literature for a general heat-related adverse outcome (e.g., heat exhaustion) could be used to identify situations where heat stress may play a significant role in shaping cumulative risk. Furthermore, it is well known that certain populations (e.g., pre-existing conditions such as asthma, elderly) are more acutely affected by heat stress. Therefore, in cumulative risk assessments where heat stress has been identified as an important risk factor, an additional uncertainty value could be applied to account for increased risk to vulnerable populations.

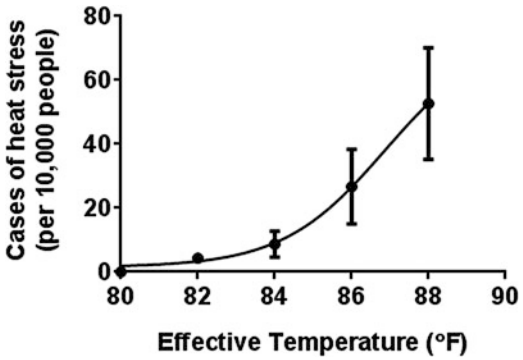

Fig. 16.2 Example of a dose-response relationship for heat stress. Heat stress data from military personnel was used to generate the dose-response relationship. Circles and bars represent the average and standard error values from three populations: Junior Platoon Leader (PLC) candidates on 6-week training, new reservists on 2-week training, and recruit trainees on 12-week training. The raw data used in the calculations described above were extracted roughly from Yaglou and Minard (1957). The solid line represents a four-parameter logistic fit to the data using GraphPad Prism

16.4.1 Case Study for Estimating Cumulative Risk from Occupational Exposure to Heat Stress and Pesticides

This case study is meant to illustrate potential considerations and decision points for conducting a cumulative risk assessment that includes both chemical and physical stressors and does not represent an accurate analysis of risk from the model stressors. Consider an agricultural community in North Carolina. The community is concerned about the combination of heat stress and pesticide exposure and would like to better understand relative contributions to overall risk in order to decide how to focus advocacy efforts. For example, they could advocate for more personal protective clothing or greater pesticide use oversight, if pesticides are driving risk, or they could advocate for implementation of cooling measures (longer breaks, greater availability of shaded areas and personal cooling devices, etc.), if heat is driving risk.

Step 1: Scoping

The first step of assessing risk from stressors present in a community is to scope the problem. This is necessarily an iterative process, as it involves identifying potential stressors and health endpoints that are of concern and exploring availability of data on exposures and outcomes. Although it would be ideal to include all relevant stressors for a particular health outcome, exposure data may limit the number and type of stressors that can be included. Additionally, the questions can be interrelated – a decision about which stressors to include could influence which risk assessment method is selected. Table 16.2 provides examples of the types of questions and potential answers that could be considered in scoping efforts related to heat and

Table 16.2 Scoping questions and answers for a risk assessment of heat stress and pesticide exposure in an agricultural community

Questions	Examples of potential answers
Is there a particular health outcome of concern?	Yes (e.g., cardiovascular disease, asthma)
	No (e.g., hospital visits, morbidity – any cause)
Who is in the population of interest?	Agricultural workers only
	Agricultural workers and their families
	Rural community in proximity to agricultural activities
What pesticides should be included?	All pesticides to which the population is exposed (exposure-based decision)
	Pesticides that are of concern based on toxicity information (disease-based decision)
	Pesticides with a particular mechanism of action (chemical class-based decision)
What other stressors should be included?	Heat only
	Heat and psychosocial stressors (e.g., socioeconomic status, exposure to violence)
	All stressors potentially linked to disease of interest
What risk assessment method is appropriate?	Dose-addition model (e.g., Hazard Index approach)
	Independent-action model

pesticide exposure. For this case study, stressors will be limited to heat and select pesticides (diazinon, parathion, chlorpyrifos, and permethrin) known to be applied on crops in the area. Three of the pesticides selected (diazinon, parathion, and chlorpyrifos) have the same mechanism of action – acetylcholinesterase inhibition – while permethrin has a different mechanism of action. All four pesticides target the nervous system and represent common agricultural exposures, while heat stress targets cardiovascular function. The set of stressors were selected based on their co-occurrence, not a common mechanism of action or target.

Step 2. Evaluating Exposure

Evaluating chemical exposures is fairly straightforward and is the same as in a single chemical risk assessment. In this example, biomonitoring data from the published literature will be used to characterize exposure to pesticides using the following equation:

$$I_C = u_C \times \frac{e_{\text{creatinine}}}{u_{\text{creatinine}} \times \text{bw}} \times \frac{1\text{mg}}{1000\mu g}, \tag{16.1}$$

where I_C is the intake of chemical C with units of μg-g/kg body weight/day, u_C is the measured concentration of chemical C (μg-g/L urine), $u_{\text{creatinine}}$ is the measured concentration of creatinine in the urine (g creatinine/L urine), $e_{\text{creatinine}}$ is the daily creatinine excretion (g/day), and bw is body weight (kg). (See Table 16.3 for values of the four pesticides measured in urine.) For the purposes of this example, a daily creatinine excretion rate of 1.5 g/day, a concentration of creatinine in the urine of 1 g/L, and a body weight of 70 kg were used.

Table 16.3 Biomonitoring data on pesticide exposure of farmworkers in North Carolina

Pesticide	Mean ± SD (ng/ml)[a]	Max (ng/ml)[a]	Estimated mean intake (μg/kg/d) [b]	Estimated max intake (μg/kg/d) [b]	Reference dose (μg/kg/d)
Diazinon	2.76 ± 6.72	7.16	0.06	0.15	0.09[c]
Parathion	7.67 ± 42.03	457.00	0.16	9.79	6[d]
Chlorpyrifos	5.37 ± 3.70	20.70	0.12	0.44	10[e]
Permethrin	3.34 ± 4.49	30.70	0.07	0.66	50[f]

[a]Urinary data from (Raymer et al. 2014)
[b]Mean and max intake values calculated using Eq. 16.1 in text
[c]Reference dose for diazinon from Teuschler et al. (1999)
[d]Provisional reference dose calculated by EPA (https://www.epa.gov/sites/production/files/2016-09/documents/parathion.pdf)
[e]Reference dose for chlorpyrifos from Zhao et al. (2006)
[f]Reference dose for permethrin from IRIS (2017)

Table 16.4 Heat stress data for summer months in North Carolina

Week (2015)	Heat stress average over week (WGBT[a])	Heat stress max temp (WGBT[a])
June 28–July 4	86 °F	92 °F
July 5–July 11	91 °F	94 °F
July 12–July 18	88 °F	93 °F
July 19–July 25	89 °F	93 °F
July 26–Aug 1	91 °F	94 °F
Aug 2–Aug 8	90 °F	97 °F
Aug 9–Aug 15	88 °F	91 °F
Aug 16–Aug 22	89 °F	95 °F
Aug 23–Aug 29	87 °F	92 °F

[a]Temperature was used as an estimate for WGBT. Source: weather history for Raleigh-Durham International Airport (weatherunderground.com)

Physical stressors present greater challenges. As discussed throughout this chapter, there are not widely accepted measurement tools and methods for characterizing exposure to physical stressors. In this example, we will use the WBGT index as a measure of effective temperature. Next, consideration of how to capture the data is required. For example, temperatures could be presented as weekly or monthly averages. Alternatively, lowest and highest temperatures could be used to present a range of possible risk values. Finally, temperature variability has recently been shown to play a role in increased risk from heat and cold (Shi et al. 2015). (See Table 16.4 for sample data on heat stress exposure in North Carolina.) The exposure options used in the risk characterization step should be selected based on the goals of the risk assessment, i.e., capturing a worst-case exposure or the most common (e.g., average) exposure level.

Step 3: Dose-Response Analysis

Dose-response analyses for the pesticides used in this example have been conducted elsewhere (see example in Teuschler et al. 1999). These analyses are used to estimate reference values, which represent a daily exposure to a human population that is not likely to be associated with appreciable risk of harmful effects over a lifetime (Table 16.3; see Chap. 14 for more on Risk Assessment). The oral reference dose for diazinon is based on cholinesterase inhibition in the plasma of rats fed diazinon (Teuschler et al. 1999), which represents the mechanism of action involved in nervous system effects of organophosphate pesticides. The two other organophosphate pesticides, parathion (https://www.epa.gov/sites/production/files/2016-09/documents/parathion.pdf) and chlorpyrifos (Zhao et al. 2006), reference dose estimates were both based on erythrocyte cholinesterase inhibition in humans. Finally, the permethrin reference dose was based on increased liver weight in rats (IRIS 2016). A dose-response relationship for heat stress is presented in Fig. 16.2. The determination of a reference dose for a chemical differs in many ways from determining a "safe temperature" for use in a cumulative risk assessment that includes heat as a stressor. The application of uncertainty, for example, would differ. In this example, the OSHA guidelines for permissible heat exposure threshold limit values (TLVs) are used as a reference point. The OSHA TLVs for heat stress also include different work levels – continuous, 75% work to 25% rest, 50% work to 50% rest, and 25% work to 75% rest per hour. In addition, different TLVs are provided depending on the work load (light, moderate, and heavy). Assuming a 75% work to 25% rest level per hour and a heavy work load for the agricultural setting, the TLV is 78 °F (OSHA 2016). This TLV agrees with the dose-response data in Fig. 16.2 showing a lack of heat stress at temperatures of 80 °F and below.

Step 4: Risk Characterization

A Hazard Index approach can be used to combine the individual hazard quotients for the different stressors (see chapter on risk assessment for a detailed discussion of the Hazard Index). As mentioned in the exposure section, this step also involves many decision points as to which exposure data should be used. For comparison, a Hazard Index can be calculated for the lowest weekly average temperature and highest recorded temperature over the period. The Hazard Index is calculated by summing the hazard quotients for the individual stressors. The hazard quotient for each stressor is calculated by dividing the exposure by the acceptable limit (i.e., reference dose for pesticides and TLV for heat stress). A Hazard Index less than 1 indicates no expectation of adverse health effects. As the Hazard Index increases above one, there is increasing concern for adverse health effects. In this case, the Hazard Index calculation would be:

$$HI = HQ_{Heat} + HQ_{Diazinon} + HQ_{Parathion} + HQ_{Chlorpyrifos} + HQ_{Permethrin}$$

For the lowest temperature and mean pesticide exposure case, the Hazard Index would be:

$$\mathrm{HI} = \frac{86}{78} + \frac{0.06}{0.09} + \frac{0.16}{6} + \frac{0.12}{10} + \frac{0.07}{50}$$
$$\mathrm{HI} = 1.10 + 0.67 + 0.03 + 0.012 + 0.001$$
$$\mathrm{HI} = 1.81$$

In this low exposure case, it is apparent that the pesticides are contributing less to the overall Hazard Index score, and heat stress is driving the value above 1.

Alternatively, the Hazard Index for the worst-case scenario would be:

$$\mathrm{HI} = \frac{97}{78} + \frac{0.15}{0.09} + \frac{9.79}{6} + \frac{0.44}{10} + \frac{0.66}{50}$$
$$\mathrm{HI} = 1.24 + 1.67 + 1.63 + 0.04 + 0.01$$
$$\mathrm{HI} = 4.60$$

In the high-exposure case, heat stress continues to contribute to the >1 Hazard Index, but the values of diazinon and parathion represent larger contributions.

Step 5: Risk Management
The case study provided above illustrates how incorporating heat stress into an evaluation of risk can help in understanding relative contributions of different stressors. These exercises can help communities and policymakers in deciding between different risk mitigation strategies. It is interesting to note that whereas pesticide exposure can vary widely from average exposure to maximum, temperature has a less dynamic range. As more physical (and other nonchemical stressors) are incorporated into cumulative risk assessments, a better understanding of their impact will be gained.

16.5 Forces: Noise and Chemicals

Noise presents another multifaceted physical stressor that can enhance chemical toxicity indirectly via activation of stress hormones and directly by acting jointly at a common target. Diseases associated with co-occurring exposure to noise and chemicals include hearing loss, as well as respiratory and cardiovascular diseases. The indirect pathway is often referred to as "noise annoyance" because it involves perception of noise as a stress (Babisch et al. 2013). Both acute and chronic studies with noise exposure have demonstrated stress hormone responses. For example, a study with 3950 middle-aged men exploring factors contributing to incidence of ischemic heart disease found that participants who were highly annoyed by noise had a higher chance of developing heart disease (odds ratios = 1.7–3.0) (Babisch et al. 2003). Less information is available on the interaction of noise with respiratory tract disease, which represents an emerging area of study (Recio et al. 2016). Noise pollution and air pollution tend to co-occur in urban environments. Frequently heard in urban settings, noises associated with danger (e.g., sirens) have the potential to trigger a stress response even during sleep. Commonly, studies

exploring road traffic pollution have focused on either air or noise pollution, but not the combination. However, many diseases (e.g., asthma, bronchitis, skin disease) display increased incidence in areas with high noise pollution (Ising et al. 2003, 2004).

Occupational settings (e.g., construction, machine operation) can have particularly elevated and prolonged noise exposures. There is also the potential for relatively high chemical exposures in occupational settings (e.g., chemical manufacturing, oil and gas industry, agriculture). Noise exposure can cause both auditory and nonauditory effects alone and in combination with other factors. There are common features in auditory dysfunction caused by noise and some ototoxic chemicals (Fechter 1995; Johnson and Morata 2010). Degeneration of the sensory hairs in the cochlea is one of the most common findings in sensorineural hearing loss. Animal studies have demonstrated loss of hair cells from exposure to both noise and solvents with reactive oxygen species hypothesized to play a role in the hair cell damage (Henderson et al. 2006; Chen et al. 2007). Other chemicals such as metals (e.g., lead, mercury) may affect both the cochlea (Rice 1997) and the central auditory pathways (Discalzi et al. 1993; Lasky et al. 1995; Otto and Fox 1993) depending on the substance. Le Prell et al. (2007) reported that the formation of free radicals after noise trauma continued up to 10 days after cessation of the exposure, which could explain why the loss of hair cells worsens after exposure. Toxic insults on the cochlea have also been shown to continue after cessation of exposure to solvents (Johnson and Canlon 1994).

More recently, it has been reported that some aromatic solvents reduce the protective role played by the middle-ear acoustic reflex (Venet et al. 2011). A dysfunction of this reflex would increase risks to hearing by allowing higher acoustic energy levels to penetrate the inner ear (Maguin et al. 2009; Campo et al. 2007). This would make co-exposure more dangerous than exposure to noise or to styrene alone. Other chemicals such as metals (e.g., lead, mercury) and pesticides may affect the hearing function (Choi et al. 2012; Shargorodsky et al. 2011) by acting on both the cochlea (Rice 1997) and the central auditory pathways (Discalzi et al. 1993; Lasky et al. 1995; Otto and Fox 1993) depending on the substance.

Solvent exposures have the potential to affect hearing in the absence of exposure to occupational noise, or they can enhance the effects of noise on hearing loss. Carbon monoxide (CO) exposure has also been shown to potentiate noise-induced hearing loss (Rao and Fechter 2000). Fechter et al. (2000) characterized the joint effects of CO and noise on hearing loss using a benchmark-dose approach for risk assessment (U.S. EPA BMDS version 1.3). They found that an exposure of 194 ppm CO represented the lower bound of the benchmark dose that would yield a 10% increase in noise-induced hearing loss. Notably, these levels of CO are less than one order of magnitude higher than the permissible exposure level (PEL) of 50 ppm set by the Occupational Safety and Health Administration (OSHA). It is also important to note that periods of recovery following exposure did not abrogate the effects (i.e., changes from co-exposure to noise and CO were permanent) (Chen and Fechter 1999; Rao and Fechter 2000). Furthermore, the dose response was not monotonic

for noise, with the smaller noise exposure resulting in maximal hearing loss in combination with CO. In more recent work, co-exposure to styrene and different types of noise (6-h continuous noise of 85 dB Sound Pressure Level (SPL) or impulse noise of 80 dB) was evaluated, and impulse noise was found to elicit greater damage (Venet et al. 2015; Campo et al. 2014). The characteristics of the noise exposure and solvents like toluene and styrene can disrupt the natural protective mechanisms of the ear such as the middle-ear acoustic reflex. This was a demonstration of a second mechanism of solvent-induced damage, beyond the cochleotoxic mechanism described earlier. It consists of a rapid pharmacological impact on the central nervous system by the inhibition of the protective reflex (Campo et al. 2001). Finally, studies have also demonstrated that as the number of stress factors increase, the lowest observable adverse effect level (LOAEL) for hearing loss decreases.

The evidence presented above represents a small portion of the literature describing the link between occupational chemical exposure and hearing loss. To address this concern, OSHA and other groups have published comprehensive evaluations of ototoxic substances, as well as documented hazards associated with workplace exposure to noise and ototoxic chemical substances (EU-OSHA 2009; Johnson and Morata 2010). These references include qualitative information on noise and chemical interactions and highlight policies from specific countries and multinational agencies.

In an example of incorporating physical stressors into cumulative risk assessment in a quantitative manner, Evans et al. (2014) developed a case study characterizing risk for hearing impairment from combined exposures to noise and volatile organic compounds (VOCs). They used data from the 1999–2000 U.S. National Health and Nutrition Examination Survey (NHANES) to estimate VOC exposures and modeled street-level noise data (i.e., a noise map) to estimate block group-level noise categories (45–60 dB, 61–65 dB, 66–70 dB, and 71–75 dB). The cumulative risk for potential hearing loss from co-exposure to noise and VOCs was calculated using a Hazard Index approach (see the heat and pesticide case study above and Chap. 14 for a discussion of HI). Hazard Indices ranged from 0.8 (lowest noise category and 10th percentile for total VOCs) to 1.7 (highest noise category and 90th percentile for total VOCs). Although the authors noted limitations of the approach (e.g., issues with combining heterogeneous data), it did demonstrate the feasibility of combining chemical and nonchemical stressors using an established cumulative risk assessment approach. Furthermore, it identified noise as the driver of risk in the case study, which could help inform decision-makers in how to invest limited resources to provide the greatest impact to public health.

Currently, the French Institut National de Recherche et de Sécurité (INRS) is working to incorporate information on noise damage risk-criteria into the web-tool *Mixie*. The original web-tool was created by the University of Montreal and the Institut de Recherche Robert-Sauvé en Santé et en Sécurité du Travail (Vyskocil et al., 2007) to assess the risks associated with exposure to a mixture of airborne chemical substances in the workplace (http://www.irsst.qc.ca/en/publications-tools/tool/i/100037/n/mixie-mixtures-of-substances-in-the-workplace-computer-

based-tool-for-evaluating-the-chemical-risk-calculation-of-the-rm). Toxicological effects are considered additive and the multiple exposure index is used for assessing the risk encountered by people exposed to several substances present in the workplace. The sum of the fractions of measured individual exposure concentrations and their Time-Weighted Average Exposure Value (TWAEV) for each substance results in a percentage of the recommended dose of the mixture. A percentage of 100 indicated that exposures are at their recommended exposure limit according to Canadian Occupational Exposure Limits (OELs). The planned additions would incorporate information on the interaction of chemicals with noise, to alert those conducting risk assessment.

16.6 Challenges and Recommendations for Incorporating Physical Stressors into Cumulative Risk Assessment

Although it is an accepted fact that physical stressors can impact human health, they have not typically been included in cumulative risk assessment efforts, which have focused almost exclusively on chemicals. There are multiple factors that present obstacles to the inclusion of physical stressors into cumulative risk assessment. However, there are also options for working around or overcoming these challenges using available tools.

16.6.1 Challenge: Deciding Which Physical Stressors to Include in Cumulative Risk Assessments

There are numerous potential physical stressors available for consideration. For example, a risk assessment in an urban environment could reasonably include traffic noise, heat stress, microbial load, and ultraviolet radiation, among other risk-contributing factors. Determining which physical stressors to include can increase the complexity of the scoping phase. Another challenge may be that data are not available for all of the potential physical stressors that are relevant.

16.6.1.1 Recommendations

There are two considerations that could help to guide inclusion of physical stressors into cumulative risk assessment. The first consideration is which physical stressors are relevant to the goals of the risk assessment. For example, if the risk assessment is targeted toward understanding relative contributions of stressors to a particular disease outcome, only physical stressors that could plausibly contribute to the disease should be included. The same types of methods that are useful in

prioritizing chemicals for study and assessment (see Part II on prioritizing mixtures) can also be applied to physical stressors. The second consideration that will drive inclusion decisions is data availability. It should be noted that exposure to some physical stressors (e.g., radio waves, microbes) may be more difficult to measure in populations than others (e.g., temperature). However, the data needs for these physical stressors will become more apparent as researchers and risk assessors continue to work through examples.

16.6.2 Challenge: Lack of Physical Stressor Data

The lack of available "dose"-response data for physical stressors is often cited as an impediment to their incorporation into cumulative risk assessments. Typical toxicity studies with physical stressors or combinations of physical and chemical stressors tend to include one level of exposure (e.g., with or without a particular microbe). However, many of the available cumulative risk assessment approaches that have been applied to multiple chemicals require dose-response information. Additionally, there is a lack of data on interactions between physical and chemical stressors. Although examples of interaction data have been presented in this chapter, joint action of the vast majority of physical and chemical stressor combinations has not been evaluated.

16.6.2.1 Recommendations

To overcome the real deficit in traditional toxicological (i.e., dose-response) data for physical stressors and data on physical and chemical stressor interactions, research in these areas needs to be prioritized. This is likely to be an iterative process. As more case studies are developed and cumulative risk assessments that include physical stressors are performed, researchers will gain a better understanding of the types of physical stressor data that will be most useful to risk assessors, which in turn will guide study design. In addition to generating more toxicological data on physical stressors, making better use of existing databases (e.g., NHANES) and creatively using data from nontraditional sources (e.g., meteorological data combined with hospital visits for respiratory disease) are recommended. The case study by Evans et al. (2014) exploring the use of secondary data on exposure to noise and VOCs in a cumulative risk assessment for hearing loss provides an excellent template for replication with other physical/chemical stressor combinations and health outcomes.

16.6.3 Challenge: Heterogeneity of Data on Physical Versus Chemical Stressors

While doses of chemicals are typically presented in route-specific standard format (e.g., mg/kg for oral or dermal exposure and ppm for food-based or inhalation exposure), there is no consensus on "dose" units for physical stressors (e.g., for heat, the WBGT index is preferred; however, ambient temperature could be more widely available), and physical stressor "dose" units are not likely to align with chemical dose units (e.g., temperature versus chemical dose).

16.6.3.1 Recommendations

Although there is potential for confusion from combining factors across different data types, this does not pose a significant obstacle in calculating risk. Common dose units are not required in the HI approach (see Evans et al., (2014) example and the case study on heat and pesticides presented in this chapter) or the independent action approach. However, increased research attention on physical stressors and their dose-response relationships should inform selection of an appropriate "dose" measure.

16.6.4 Challenge: Categorical Differences

A common concern in cumulative risk assessment discussions is how to deal with stressors that fall under different regulatory jurisdictions. There are multiple arguments for including only stressors that fall under a single regulatory umbrella. There is the possibility of a legislative mandate guiding this decision (e.g., the 1996 Food Quality Protection Act [FQPA] charges the U.S. EPA's Office of Pesticide Programs to address pesticide mixtures). Due to the specificity of the FQPA, there is an assumption that stressors outside the scope of the legislation should be excluded from cumulative risk assessments addressing the mandate. Alternatively, the argument can be made on pragmatic grounds. For example, because there is not a clear path toward exposure reduction for some physical stressors, they should not be included in cumulative risk assessments. Finally, it could be argued that including stressors outside of the regulatory scope of an agency could result in the unintended consequence of decreasing action on chemical exposure. For example, if a cumulative risk assessment concludes that the risk from specific chemical exposures is dwarfed by a nonchemical factor such as low socioeconomic status, it might decrease political will to mitigate the chemical exposures.

16.6.4.1 Recommendations

The argument for limiting the scope of cumulative risk assessment based on legislative mandate or practical considerations brings up an often-cited goal of conducting cumulative risk assessments tailored to the question at hand. For example, if the goal is to make decisions about chemical remediation (i.e., determine which chemicals require resource-intensive cleanup efforts), limiting the risk assessment to chemicals may be in order. However, the inclusion of a broad range of potential stressors (both chemical and nonchemical) is recommended in addressing more global public health issues, such as determining stressors that are most likely to impact public health in communities-of-concern and using cumulative risk assessment information to determine how to direct limited resources to best protect public health. Furthermore, inclusion of both chemical and nonchemical stressors allows for comparison of relative contribution of the various stressors. In other words, the stressors that are likely to drive the adverse outcome of interest could be identified and targeted for intervention.

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