Regulatory Toxicology and Pharmacology 54 (2009) 264–271

Contents lists available at ScienceDirect

Regulatory Toxicology and Pharmacology

journal homepage: www.elsevier.com/locate/yrtph

Binary weight-of-evidence evaluations of chemical interactions—15 years of experience

Hana R. Pohl *, Moiz M. Mumtaz, Franco Scinicariello, Hugh Hansen

Agency for Toxic Substances and Disease Registry, U.S. Department of Health and Human Services, Division of Toxicology and Environmental Medicine, 1600 Clifton Road, F-32, Atlanta, GA 30333, USA

ARTICLE INFO

Article history:
Received 14 November 2008
Available online 13 May 2009

Keywords:
Risk assessment
Chemical mixtures
Interactions
Additivity
Synergism
Antagonism

ABSTRACT

The paper reflects on the last 15 years of experience in the field of mixtures risk assessment. It summarizes results found in various documents developed by the Agency for Toxic Substances and Disease Registry (ATSDR) of the weight-of-evidence (WOE) approach applied to 380 binary combinations of chemicals. Of these evaluations, 156 assessments indicated possible additivity of effects [=], 76 indicated synergism (greater-than-additive effects [>]), and 57 indicated antagonism (less-than-additive effects [<]). However, 91 combinations lacked the minimum information needed for making any assessments and, hence, were undetermined.

The paper provides examples of the rationale behind some of the WOE decisions and discusses the importance of expert judgments in risk assessment evaluations. Examples are given regarding the importance of human variability in mixtures' ability to affect human health and regarding the dose versus effect relationships.

Published by Elsevier Inc.

1. Introduction

The Agency for Toxic Substances and Disease Registry (ATSDR) has developed a program for chemical mixtures that includes mixtures research and mixtures risk assessment (De Rosa et al., 2004). Relevant to the risk assessment part of the program is ATSDR's release to the public of a document titled *Guidance Manual for the Assessment of Joint Toxic Action of Chemical Mixtures*. This manual provides guidelines for evaluating the toxicity of chemical mixtures encountered at hazardous waste sites (http://www.atsdr.cdc.gov/interactionprofiles/). An overview of this guidance was published previously (Wilbur et al., 2004). The guidance builds on two integral parts of ATSDR's mixtures program: strategically targeted applied research and assessment of the weight-of-evidence (WOE) for interactions (Mumtaz and Durkin, 1992). Also, this guidance serves as the basis of ATSDR's interaction profile program that develops a series of documents, or interaction profiles, that summarize pertinent toxicity data on specific mixtures of concern, evaluate potential interactions, and provide conclusions that are relevant to public health (Pohl and Abadin, 2008; Pohl et al., 2003, 2004; Roney and Colman, 2004). Similar approaches are used or recommended by a number of agencies (EPA, 1986, 1989; National Academy of Sciences [NAS], 1974; National Research Council [NRC], 1989; OSHA, 1993). The basic methodology that drives this guidance was applied in some of ATSDR's site consultations (ATSDR, 2005, 2006; Pohl et al., 1999) and in several internal documents developed prior to the interaction profiles program. As a result, 380 binary evaluations of interactions between chemicals related to end points of concern are currently available. An overview and the relevance to future research of chemical mixtures are presented in this paper.

2. Methods

2.1. Mixtures assessment

As illustrated in Fig. 1, mixtures can be evaluated as complete entities if data on a particular mixture are available (ATSDR, 2001, 2004a).

When available, data on one mixture often can be used on a similar mixture. A similar mixture is one that has the same chemicals as the mixture of concern but in slightly different proportions or one that has most of the same components in highly similar proportions. Similar mixtures are expected to have similar fate, transport and health effects (e.g., the jet fuel JP-5 from different sources). If no data are available, other approaches to evaluate the toxicity of the components of the mixture are commonly used. For most mixtures, component-based approaches such as the hazard index (HI) are recommended. The hazard index approach assumes dose additivity to assess the health effects of a chemical mixture from the available data on the mixture's components.

* Corresponding author. Fax: +1 770 488 3470.
E-mail address: hpohl@cdc.gov (H.R. Pohl).

0273-2300/$ - see front matter Published by Elsevier Inc.
doi:10.1016/j.yrtph.2009.05.003

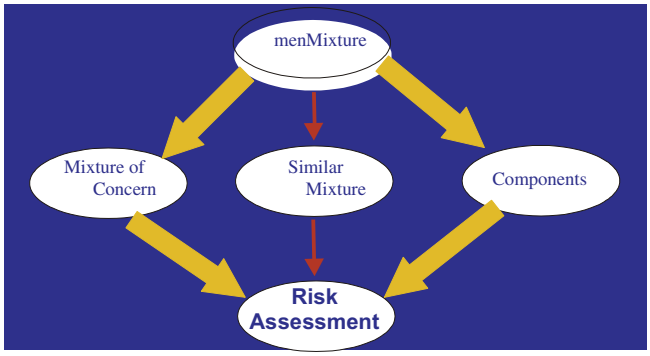

Fig. 1. Principles of mixtures evaluation.

Exposures or doses for the various components of the mixture are scaled by a defined level of exposure generally regarded as acceptable or safe (i.e., health-based guidance value) by the agency performing the assessment.

The general equation for the hazard index (HI) calculation is:

$$\mathrm{HI} = \frac{\mathrm{ChemExposure}_1}{\mathrm{DR}_1} + \frac{\mathrm{ChemExposure}_2}{\mathrm{DR}_2} + \frac{\mathrm{ChemExposure}_3}{\mathrm{DR}_n}, \quad (1)$$

$\mathrm{ChemExposure}_1$ is defined as the level of exposure to the first chemical in the mixture and DR_1 is some defined level of exposure to the first chemical (i.e., health-based guidance value),

$\mathrm{Exposure}_2$ and DR_2 are the corresponding levels for chemical 2, and the summation can extend to any number of chemicals, signified by the *n*.

When the hazard index for a mixture exceeds unity (1), a concern for the potential hazard of the mixture increases. Separate hazard indexes are estimated for each pathway and exposure duration of concern. For a given duration, hazard indexes are summed across pathways that affect the same receptor population. The target-organ toxicity dose (TTD) method, which is a refinement of the HI method, was introduced to accommodate the assessment of mixtures whose components do not all have the same critical effect. TTDs are derived similarly to other health-based guidance values (ATSDR, 2004a). For a TTD analysis, a combined WOE score is computed for each effect of concern for the mixture.

The exposure-based assessment of potential health hazard (i.e., HI) is a screening approach, to be used in conjunction with biomedical judgment, community-specific health outcome data, and community health concerns to assess the degree of public health hazard. ATSDR does not use the approach to (nor has the authority to) establish clean-up levels. The results of assessments may trigger recommendations for potential public health actions, including surveillance, health studies, community education, exposure investigations and research.

Component-based approaches are most useful when augmented with a weight-of-evidence evaluation of the potential for non-additive interactions among the components in the mixture. The WOE evaluations of the mixtures' components are used to qualitatively adjust the HI. For example, if the component-based analyses indicate that several binary combinations will have more than additive joint toxic action, the HI may underestimate the actual toxicity of the mixture. Conversely, if the component-based analyses indicate that several binary combinations will have less than additive joint toxic action, the HI may overestimate the actual hazard presented by the exposure scenario.

2.2. Binary weight-of-evidence

The assessment of binary weight-of-evidence (BINWOE) enables the direction of interactions in the mixture to be judged when information about the toxicity of the entire mixture is unavailable. As defined, "WOE is a qualitative judgment, based on empirical observations and mechanistic considerations, which categorizes the most plausible nature of any potential influence of one compound on the toxicity of another for a given exposure scenario" (Mumtaz and Durkin, 1992). In brief, this approach systematically evaluates data relevant to joint action for each possible pair of chemicals in the mixture to make qualitative BINWOE determinations for the effect of each chemical on the toxicity of every other chemical (Table 1). The importance of expert judgment will be discussed later in the manuscript.

The BINWOE determination is an end-point-specific classification that indicates the expected direction of an interaction (greater-than-additive, less-than-additive, additive, or indeterminate) and scores the data qualitatively by using an alphanumeric scheme that considers mechanistic understanding, toxicological significance, and relevance of the exposure duration, sequence, bioassay (*in vitro* versus *in vivo*), and route of exposure (ATSDR, 2004a). The mechanistic understanding scores are for groups that indicate direct data of the mechanism of the interaction (I), that infer the mechanism of the interaction from similar chemicals (II), or that indicate that reasons for the observed interaction or additivity are not known (III). Similarly groups for toxicological significance indicate that: the interaction was observed directly and is linked to a toxicologically significant end point (A) or that the toxicological significance of the interaction can be inferred (B) or is unclear (C). For each pair of chemicals, a minimum of two BINWOEs is derived: one for the effect of Chemical A on Chemical B and one for the effect of Chemical B on Chemical A. Some binary combinations of chemicals may have multiple BINWOEs developed for target organ-specific information.

2.3. Mixtures evaluated

The BINWOE determinations for the 15 mixtures presented in Table 2 are summarized in this paper.

Eleven mixtures were evaluated in ATSDR's interaction profiles during the years of 2000–2007. The other mixtures were evaluated in internal documents during the years of 1995–1996, shortly after the Mumtaz and Durkin (1992) methodology was introduced.

Most mixtures were selected based on information in ATSDR's database on chemicals at hazardous waste sites. Of the 1706

Table 1
BINWOE classification.

Direction of interaction
=additivity
>greater-than-additivity
<less-than-additivity
Mechanistic understanding
I. Direct and unambiguous mechanistic data: The mechanism(s) by which the interactions could occur has been well characterized and leads to an unambiguous interpretation of the direction of the interaction
II. Mechanistic data on related compounds: The mechanism(s) by which the interactions could occur has not been well characterized for the chemicals of concern, but structure–activity relationships, either quantitative or informal, can be used to infer the likely mechanisms(s) and the direction of the interaction
III. Inadequate or ambiguous mechanistic data: The mechanism(s) by which the interactions could occur has not been well characterized or information on the mechanism(s) does not clearly indicate the direction that the interaction will have
Toxicological significance
A. The toxicological significance of the interaction has been directly demonstrated
B. The toxicological significance of the interaction can be inferred or has been demonstrated for related chemicals
C. The toxicological significance of the interaction is unclear