Regulatory Toxicology and Pharmacology 73 (2015) 712–725

Contents lists available at ScienceDirect

Regulatory Toxicology and Pharmacology

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

Development of an inhalation unit risk factor for isoprene

Joseph T. Haney Jr. [a, *], Tracie Phillips [a], Robert L. Sielken Jr. [b], Ciriaco Valdez-Flores [c]

[a] Texas Commission on Environmental Quality (TCEQ), Toxicology Division, MC-168, P.O. Box 13087, Austin, TX 78711, United States
[b] Sielken & Associates Consulting, Inc., 3833 Texas Avenue, Bryan, TX 77802, United States
[c] Texas A&M University, 4073 Emerging Technologies Building, 3131 TAMU, College Station, TX 77843, United States

ARTICLE INFO

Article history:
Received 14 October 2015
Received in revised form
28 October 2015
Accepted 29 October 2015
Available online 3 November 2015

Keywords:
Isoprene
Carcinogenicity
Tumorigenesis
Regulatory
Cancer
Liver
Toxicity
Inhalation

ABSTRACT

A unit risk factor (URF) was developed for isoprene based on evaluation of three animal studies with adequate data to perform dose–response modeling (NTP, 1994, 1999; Placke et al., 1996). Ultimately, the URF of 6.2E-08 per ppb (2.2E-08 per μg/m^3) was based on the 95% lower confidence limit on the effective concentration corresponding to 10% extra risk for liver carcinoma in male B6C3F$_1$ mice after incorporating appropriate adjustment factors for species differences in target tissue metabolite concentrations and inhalation dosimetry. The corresponding lifetime air concentration at the 1 in 100,000 no significant excess risk level is 160 ppb (450 μg/m^3). This concentration is almost 4400 times lower than the lowest exposure level associated with statistically increased liver carcinoma in B6C3F$_1$ mice in the key study (700 ppm in Placke et al., 1996) and is above typical isoprene breath concentrations reported in the scientific literature. Continuous lifetime environmental exposure to the 1 in 100,000 excess risk level of 160 ppb would be expected to raise the human blood isoprene area under the curve (AUC) less than one-third of the standard deviation of the endogenous mean blood AUC. The mean for ambient air monitoring sites in Texas (2005–2014) is approximately 0.13 ppb.

1. Introduction

Isoprene is the 2-methyl analog of 1,3-butadiene. It is used largely in the manufacturing of synthetic rubber (e.g., for vehicle tires). It is also used in the manufacturing of styrene-isoprene-styrene block co-polymers and butyl rubber, in the production of hydrocarbon resins, and for the synthesis of terpenes (BG Chemie, 2000; Melnick et al., 1996; Sharkey, 1996). Anthropogenic sources of isoprene include: petroleum cracking, ethylene production (by-product), wood pulp production, oil fires, tobacco smoke, and automobile exhaust (Hurst, 2007; Melnick et al., 1996; Sharkey, 1996).

Isoprene is also produced naturally by plants (isoprene biosynthesis is associated with photosynthesis), animals, and bacteria. The amount of isoprene produced naturally far exceeds that which is produced synthetically. It is the underlying structure of isoprenoid biochemicals such as cholesterol, carotenoids, and vitamin A (Hurst, 2007; Song et al., 2004). Greater than 200 different plant species, especially trees, emit isoprene (Loreto, 1997). Isoprene accounts for more than half of natural volatile organic compound (VOC) emissions. U.S. woodlands produce an estimated 3 mg/m^2 compared to about 5 mg/m^2 per hour total VOC, with the south central and southeastern areas of the U.S. having the highest biogenic emissions. Emissions are seasonal (highest in the summer) since isoprene is primarily emitted by deciduous trees (Guenther et al., 1994, 1995 and Fuentes and Wang, 1999 as cited by NTP, 2014). The tree species with the highest isoprene emissions are generally in the genera *Quercus* (oaks) and *Populus* (poplars), with *Picea* (spruces) being the only conifer isoprene emitters (Logan et al., 2000). In addition to emissions from trees, foods are expected to be a daily source of exposure since agricultural crops emit isoprene and it is the basic structural unit in many natural products found in consumed foods (e.g., terpenes, vitamins A and K, carrots, coffee, essential oil of oranges) (NTP, 2014).

Humans produce isoprene endogenously at a rate of 0.15 μmol/kg per hour, which is equivalent to 2–4 mg/kg-day, with blood concentrations ranging from 1.0 to 4.8 μg/L (Taalman, 1996 and Cailleux et al., 1992 as cited by NTP, 2014). In human breath, isoprene has been found to be one of the main endogenous compounds, accounting for up to 70% of exhaled hydrocarbons (Gelmont et al., 1981 as cited by NTP, 2014). For example, MAK

* Corresponding author.
E-mail addresses: Joseph.Haney@tceq.texas.gov (J.T. Haney), Tracie.Phillips@tceq.texas.gov (T. Phillips), SielkenAssoc@aol.com (R.L. Sielken), Ciriacov@tamu.edu (C. Valdez-Flores).

http://dx.doi.org/10.1016/j.yrtph.2015.10.030

(2012) reports a weighted multiple-study mean of 64 ± 49 ppb in 337 volunteers. By comparison, annual averages at ambient air monitoring sites in Texas range from not detected to 0.84 ppb, with an approximate statewide mean and median of 0.13 and 0.07 ppb, respectively (Texas Air Monitoring Information System (TAMIS) data for 2005–2014). Generally, the major sources of isoprene in ambient air appear to be biogenic emissions in rural areas and vehicle emissions in urban areas (Borbon et al., 2001 and So and Wang, 2004 as cited by NTP, 2014).

As stated previously, isoprene is the 2-methyl analog of 1,3-butadiene, an industrial chemical that has been identified as an animal and human carcinogen. According to the National Toxicity Program's 13th *Report on Carcinogens* (NTP, 2014), isoprene is "reasonably anticipated to be a human carcinogen" based on sufficient evidence of carcinogenicity from studies in experimental animals (i.e., tumors at several different tissue sites in mice and rats). For example, inhalation exposure to isoprene induced increased incidences of neoplasms of the liver, lung, and hematopoietic system in mice (Placke et al., 1996). It is important for the Texas Commission on Environmental Quality (TCEQ) to conduct an inhalation carcinogenic dose–response assessment for isoprene since:

- A carcinogenic dose–response assessment for inhalation exposure to isoprene has not been conducted by human health assessment programs such as the Integrated Risk Information System (IRIS) of the U.S. Environmental Protection Agency (USEPA) or the Office of Environmental Health Hazard Assessment (OEHHA) of the California Environmental Protection Agency (CalEPA);
- The TCEQ performs carcinogenic dose–response assessments for chemicals considered "likely to be carcinogenic to humans", particularly when a suitable dose–response assessment conducted by another agency is not available for adoption (TCEQ, 2015);
- Isoprene is detected in ambient air and there are industrial point sources of isoprene emissions in Texas that may increase naturally-occurring ambient air concentrations in neighboring communities; and
- A unit risk factor (URF) may be needed to help ensure the protection of public health.

Accordingly, a URF for isoprene has been developed based on the evaluation of three laboratory animal studies with adequate data to perform dose–response modeling (NTP, 1994, 1999; Placke et al., 1996). The purpose of this paper is to present the procedures used in the carcinogenic assessment of isoprene and the derivation of the URF. The URF is then used to calculate the environmental air concentration associated with the no significant excess risk level of 1 in 100,000 assuming lifetime exposure (TCEQ, 2015).

2. Materials and methods

The TCEQ (2015) guidelines for carcinogenic assessment employ the four-step risk assessment process formalized by the National Research Council (NRC, 1983, 1994) and the procedures recommended in the most recent USEPA cancer guidelines (USEPA, 2005a, 2005b) and scientific literature. For chronic adverse effects determined or assumed (e.g., by default, due to a lack of sufficient carcinogenic MOA data to justify an alternative approach) to be associated with linear dose–response relationships in the low-dose region (typically cancer), the TCEQ adopts or derives URFs. In such cases, a linear extrapolation is performed to estimate excess lifetime risk at lower doses, for example, through the calculation of a point of departure (POD) using USEPA benchmark dose (BMD) software (version 3.4) to fit data to a dose–response model. A common POD for calculation of a URF is the 95% lower confidence limit on the effective concentration (EC) corresponding to 10% extra risk (LEC_{10}). The slope of the line from zero excess risk at zero exposure to this POD is the inhalation URF (e.g., $0.1/LEC_{10} = URF$), which may be described as the excess risk estimated to result from continuous lifetime exposure to an agent on a per ppb or $\mu g/m^3$ in air basis (i.e., excess risk per ppb or $\mu g/m^3$ assuming continuous lifetime exposure).

While human studies are preferred for URF derivation under the TCEQ (2015) guidelines, no human studies have evaluated the relationship between human cancer and inhalation exposure to isoprene specifically (NTP, 2014). Although there are currently no human studies that indicate isoprene exposure may increase the risk of cancer, the USEPA Cancer Guidelines (USEPA, 2005a) indicate as a matter of public health-protective policy that positive effects in animal cancer studies are a basis for assessing the carcinogenic hazard to humans (in the absence of human data), and laboratory animal studies are available to quantify the relationship between animal tumors and isoprene exposure via inhalation. More specifically, three animal studies contain the data necessary to perform dose–response modeling for tumors induced by inhalation exposure to isoprene and are considered below (NTP, 1994, 1999; Placke et al., 1996). Additional details on these animal studies and methods utilized for the dose–response assessment (e.g., BMD modeling) are provided below.

3. Carcinogenic assessment

The following sections discuss key steps in the carcinogenic assessment of isoprene and development of the URF. Consistent with Fig. 1-2a of TCEQ (2015), the key steps are generally as follows:

- Conduct literature review and solicit information from interested parties.
- Perform carcinogenic weight of evidence (WOE) and mode of action (MOA) analyses (linear low-dose extrapolation is the default for a mutagenic or unknown MOA).
- Identify key studies with sufficient information to conduct dose–response analyses (only animal study cancer data are sufficient and available for isoprene inhalation exposure).
- Conduct dose–response modeling with the best methods available to derive a POD (e.g., LEC_{10} from BMD modeling).
- Calculate the URF (e.g., $0.1/LEC_{10} = URF$).

The first two steps shown above (i.e., literature search, carcinogenic WOE and MOA analyses) are inherently part of the process, but need not be discussed in detail here since the focus of this paper is on documentation of the dose–response analyses and methods used in the URF derivation process. The first step was initially conducted by the TCEQ in 2012, and a new scientific literature search (through July 2015) did not reveal additional studies for dose–response modeling. In regard to the second step, a carcinogenic WOE analysis has recently been conducted by NTP (2014) and concluded that isoprene is "reasonably anticipated to be a human carcinogen", and an MOA analysis would not likely result in a departure from the linear, low-dose extrapolation approach employed in this study (e.g., although Placke et al., 1996 indicated that a threshold effect level appeared to exist for tumor development, like 1,3-butadiene, the diepoxide intermediates of isoprene have the ability to cause mutations and there is a lack of sufficient carcinogenic MOA data to justify an alternative approach). Consequently, the following sections focus on the last three steps shown above.