MICHIGAN DEPARTMENT OF ENVIRONMENTAL QUALITY

INTEROFFICE COMMUNICATION

TO: File for Hexachloroethane (CAS No. 67-72-1)

FROM: Cathy Simon, Toxics Unit, Air Quality Division

SUBJECT: Updated Screening Levels for Hexachloroethane

DATE: September 19, 2012

The initial threshold screening level (ITSL) for hexachloroethane has been revised from 3.5 µg/m^3 (24-hour averaging time) to 30 µg/m^3 based on an annual averaging time. The new acute ITSL is 1,600 µg/m^3 based on an 8-hour averaging time. The initial risk screening level (IRSL) for hexachloroethane has been changed from 0.3 µg/m^3 to 0.1 µg/m^3, and the secondary risk screening level (SRSL) has been established at 1 µg/m^3. Both the IRSL and SRSL are based on an annual averaging time.

Background

In August 1992, the Michigan Department of Environmental Quality (MDEQ) Air Quality Division (AQD) established an ITSL for hexachloroethane of 3.5 µg/m^3 (24-hour averaging time (MDEQ, 1992). This ITSL was derived from an oral reference dose (RfD) of 1 µg/kg/day listed in the U.S. Environmental Protection Agency's (US EPA's) Integrated Risk Information System (IRIS) database. No inhalation reference concentration (RfC) was available in IRIS at this time. Also in August 1992, an IRSL of 0.3 µg/m^3 (annual averaging time) was established based on an inhalation unit risk value of 4 x 10^{-6} (µg/m^3)$^{-1}$ listed in the IRIS database (MDEQ, 1992).

In September 2011, the US EPA updated the IRIS database for hexachloroethane, establishing a revised oral RfD and revised oral unit risk value, adding an inhalation reference concentration (RfC) for the first time, and withdrawing the previous inhalation cancer potency value (EPA, 2012a).

The focus of this evaluation was to review the new RfC and revised unit risk value listed in the updated IRIS database, and determine the appropriateness of using these values to derive an ITSL, IRSL, and SRSL for hexachloroethane. This review relied primarily on the scientific information presented in the IRIS database and supporting documentation, *Toxicological Review of Hexachloroethane* (EPA, 2011), which were completed in September 2011.

Evaluation of the RfC and ITSL

In 1992 when the ITSL was first established, an oral RfD was available on IRIS, but no inhalation RfC. At that time, the RfD was identified as the best available data to use in deriving the ITSL. Typically, an inhalation RfC is considered the first choice in the hierarchy of methods used to establish an ITSL. When an RfC is available, the ITSL is set at the same value as the RfC.

The RfC listed on the current IRIS database is 30 µg/m^3, and is derived from a six week animal inhalation study by Weeks et al (1979). No other subchronic or chronic inhalation studies in

animals were available for hexachloroethane, nor were any human data available for derivation of an RfC. In the study by Weeks et al (1979), groups of male and female Sprague-Dawley rats (25 per group), male Hartley guinea pigs (10 per group), male beagle dogs (4 per group), and male and female quail (20 per group) were exposed to 0, 15, 48, or 260 ppm hexachloroethane (0, 145, 465, or 2,517 mg/m^3, respectively) for 6 hours/day, 5 days/week for 6 weeks. Half of the animals were sacrificed and necropsied at the end of the 6 week exposure period, while the remaining animals were observed for an additional 12 weeks prior to sacrifice. Other tests included in the study by Weeks et al (1979) included a teratology study and a behavioral study. In the teratology study, pregnant female Sprague-Dawley rats (22 per group) were exposed to the same concentrations as above on days 6 – 16 of gestation. In the behavioral study, changes in conditioned avoidance and spontaneous motor activity were measured in groups of 15 male Sprague-Dawley rats, also exposed to the same concentrations.

The US EPA (2011) identified a NOAEL of 465 mg/m^3 and a LOAEL of 2517 mg/m^3 for rats, beagle dogs, and guinea pigs from the study by Weeks et al (1979). No developmental effects were observed at any exposure concentrations in fetuses from exposed pregnant Sprague-Dawley rats. The following table, modified from Table 4-20 in *Toxicological Review of Hexachloroethane* (EPA, 2011), summarizes the results from the study by Weeks et al (1979):

Table 1: Summary of data from the Weeks et al (1979) inhalation toxicity study with hexachloroethane

Species	NOAEL (mg/m^3)	LOAEL (mg/m^3)	Effect
Male beagle dogs	465	2517	Tremors, ataxia, hypersalivation, head bobbing, facial muscular fasciculations.
Male Hartley guinea pigs	465	2517	Reduced body weight, increased relative liver weight, 40% mortality by week 5.
Sprague-Dawley rats	465	2517	Both sexes (all animals): tremors, ruffled pelt, red exudate around eyes. Males: reduced body weight gain, increased relative kidney, spleen, and testes weights. Females: increased relative liver weight.
C. Japonica (Japanese quail)	2517	Not Established	No effects.
Pregnant Sprague-Dawley rats	Maternal: 465 Developmental: 2517	Maternal: 2517 Developmental: Not established	Maternal: tremors[a], decreased body weight gain. Fetal: no effects.
Male Sprague-Dawley rats	465	2517	No effect for avoidance latency and spontaneous motor activity tests. Reduced body weight.
[a] Incidence data on tremors not reported by the study authors. Note: Above table modified from Table 4-20 (EPA, 2011).			

In addition to the effects noted above in the table, Weeks et al (1979) found an increased incidence of mucopurulent nasal exudate in pregnant rats exposed to the two highest doses of hexachloroethane. This inflammatory exudate was found in 85% of the animals exposed to 48 ppm, and 100% of those exposed to 260 ppm. Similar lesions as well as lymphoid hyperplasia in the lamina propria of the trachea and pneumonitis were also observed in the male and non-

pregnant female rats exposed to 260 ppm and sacrificed at 6 weeks. The authors attributed these lesions to "potentiation of an endemic mycoplasia infection". Lastly, it should also be noted that body weight gain of pregnant rats was significantly decreased in animals exposed to 260 ppm starting at day 8 of gestation, and at day 14 in the 48 ppm dose group.

Unfortunately, very little quantitative data were provided by Weeks et al (1979) for the results of the subchronic inhalation study. Some of the limitations include: no incidence data for histopathological effects were provided, no organ weight data were provided for any of the study groups, and the only body weight data provided were for the male rats in the behavioral study groups.

The US EPA identified neurological effects as the key effect for derivation of the inhalation RfC, as stated in the IRIS documentation (EPA, 2012a):

> Neurological effects were observed in male and non-pregnant female Sprague-Dawley rats, male Beagle dogs, and pregnant Sprague-Dawley rats only at the highest dose tested. Incidence data were not reported, which precluded application of BMD modeling. Therefore, the NOAEL of 465 mg/m^3 identified in Weeks et al. (1979) was selected as the POD for the derivation of the RfC based on effects in male and non-pregnant female rats and male dogs exposed to hexachloroethane for 6 weeks and pregnant rats exposed for 11 days, on GD 6–GD16.

While identifying neurological effects as the key effect for deriving the RfC, the US EPA also considered both the respiratory effects and decreased body weight observed in exposed animals in this determination. With regard to the respiratory effects, the US EPA appeared to agree with Weeks et al (1979) that the data suggested these effects were due to a potentiation of an underlying infection rather than a result of hexachloroethane exposure. The US EPA also noted that the reduced weight gain in the rats could be related to mycoplasma, since infected rodents generally gain less weight or lose weight compared with non-infected rodents. However, the US EPA concluded that the respiratory tract effects could not be excluded from consideration as a potential effect because no data were presented by Weeks et al (1979) to demonstrate the presence of mycoplasma in the lungs (EPA, 2011). While considering the respiratory tract effects as a possible effect for deriving the RfC, the US EPA's final conclusion was to base the RfC on the neurological effects, due to the consistent observation of these effects across experiments in both rats and dogs (EPA, 2011).

Using the NOAEL of 465 mg/m^3 identified above as the POD, EPA then adjusted this concentration to continuous exposure as follows:

$$NOAEL[_{ADJ}] = (465\ mg/m^3) \times (6/24\ hours) \times (5/7\ days) = 83.0\ mg/m^3$$

The US EPA identified hexachloroethane as a Category 3 gas, in which the dosimetric adjustment factor for determining the human equivalent concentration (HEC) is based on the regional gas dose ratio (RGDR), where the RGDR is the ratio of the animal to human blood:gas partition coefficients. When these coefficients are unknown, as in the case of hexachloroethane, a RGDR of 1 is used. Therefore, the $NOAEL_{(HEC)}$ is determined as follows:

$$NOAEL_{(HEC)} = NOAEL[_{ADJ}] \times RGDR$$

$$NOAEL_{(HEC)} = 83.0\ mg/m^3 \times 1 = 83.0\ mg/m^3$$

The $NOAEL_{(HEC)}$ of 83.0 mg/m^3 was then divided by a total uncertainty factor (UF) of 3000 to arrive at a RfC of 30 $\mu g/m^3$. The uncertainty factor of 3000 was composed of the following:

UF_A (interspecies UF) = 3
UF_H (intraspecies UF) = 10
UF_S (subchronic to chronic UF) = 10
UF_D (database UF) = 10

The US EPA (2012a) justified the use of the database UF as follows:

> The toxicity data for inhalation exposure to HCE is limited and largely restricted to one subchronic (6-week) inhalation study (Weeks et al, 1979) in rats, male dogs, male guinea pigs, and quail. The same investigators performed a developmental/teratogenic study and an acute study (single 6 or 8 hour inhalation exposures) in rats. Although maternal toxicity was reported in the developmental/teratogenic study, fetuses of HCE-exposed dams did not exhibit any significant skeletal or soft tissue anomalies. The toxic effects observed in the dams in the developmental/teratogenic study (11-day exposure) were similar to those observed in the rats exposed for 6 weeks, although additional effects were observed in the rats exposed for the longer duration. The database lacks a long-term study and a multigeneration reproductive toxicity study. In addition, the database lacks studies of neurotoxicity and developmental neurotoxicity, endpoints of concern based on the available inhalation data demonstrating neurotoxicity in rats and dogs.

RfCs, which include an UF_D, are examined on a case-by-case basis to determine the appropriateness of including this uncertainty factor in derivation of the ITSL. In the case of hexachloroethane, use of such an uncertainty factor is justified considering the limitations of the existing database. First of all, only one inhalation toxicity study was available for hexachloroethane and the duration of exposure was limited to six weeks. Typically, the minimum duration study used to derive a RfC or RfD is 90 days, especially with the use of a subchronic to chronic uncertainty factor of 10. Other limitations of this study include the minimal reporting of effects, especially the lack of quantitative data. Secondly, while the available data identifies neurotoxicity as an endpoint of concern, the mechanism of action for this endpoint is unknown (EPA, 2011), and the existing data have not adequately characterized the spectrum of this effect. In addition to the neurotoxic effects observed in the inhalation study by Weeks et al (1979), EPA (2011) also cites two studies showing neurotoxic effects in sheep via oral exposure. The concern for potential neurotoxic effects is increased, considering that tetrachloroethylene has been identified as a metabolite of hexachloroethane (EPA, 2011). The RfC for tetrachloroethylene is based upon neurotoxic effects observed in humans, the most sensitive endpoint identified for this chemical (EPA, 2012b). The US EPA's review of the epidemiologic data for tetrachloroethylene indicates support for a "broad range of cognitive, motor, behavioral, and visual functional deficits" following exposure to this chemical (EPA, 2012b).

Considering all of the above information, the inhalation RfC as listed in the IRIS database (EPA, 2012a) is appropriate to use in deriving the ITSL. Therefore, pursuant to Rule 232(1)(a) of the Michigan Air Pollution Rules:

ITSL = RfC

ITSL = 30 $\mu g/m^3$

Rule 232(2)(b) specifies that the averaging time for an ITSL based on an inhalation RfC is 24 hours. Rule 229(2)(b), however, allows for the use of alternative methods for deriving an ITSL from those specified in Rule 232, provided those methods are more appropriate based on toxicological grounds and supported by the scientific data. Available data indicate that hexachloroethane causes neurological effects by both acute and longer term exposure. A single ITSL of 30 µg/m^3 based on a 24-hour averaging time should be protective of both acute and chronic neurological effects of hexachloroethane; however, it is likely over-conservative, and a more appropriate approach would be to set separate acute and chronic based ITSLs if adequate data are available. An evaluation of the data has been done and an acute-based ITSL derived as discussed in the section below. Therefore, pursuant to Rule 229(2)(b), the ITSL for hexachloroethane is 30 µg/m^3 based on an annual averaging time. This ITSL should provide adequate protection from chronic exposures to hexachloroethane.

Acute ITSL

The Agency for Toxic Substances and Disease Registry (ATSDR) has established a minimal risk level (MRL) of 6 ppm for acute inhalation exposure to hexachloroethane (ATSDR, 1997). No other acute health based benchmark value established by a federal or a state agency was identified to evaluate for purposes of establishing an acute based ITSL, although the American Council of Governmental Industrial Hygienists (ACGIH) has established a threshold limit value (TLV) of 1 ppm for hexachloroethane (ACGIH, 2008). The National Institute for Occupational Safety and Health (NIOSH) recommended exposure level, and the Occupational Safety and Health Administration (OSHA) standard for hexachloroethane are also both 1 ppm. Both the ATSDR MRL of 6 ppm and the ACGIH TLV of 1 ppm were evaluated for consideration of establishing an acute ITSL for hexachloroethane.

The ATSDR acute inhalation MRL was derived from the study by Weeks et al (1979). The ATSDR identified a LOAEL of 260 ppm and a NOAEL of 48 ppm, based on neurological effects, from the teratology study by this author. The ATSDR (1997) converted the NOAEL of 48 ppm in rats to a human equivalent concentration (HEC) as follows:

HEC = 48 ppm x [(0.22 m^3/day/0.204 kg) / (20 m^3/70 kg)] = 181 ppm

The HEC of 181 ppm was then divided by an uncertainty factor of 3 for extrapolating from animals to humans, and an uncertainty factor of 10 for human variability, resulting in an acute MRL of 6 ppm.

The ACGIH TLV of 1 ppm was derived to be protective of kidney and liver effects, and is based upon a study by Gorzinski et al (1985) in which rats were administered hexachloroethane in the diet for 16 weeks (ACGIH, 2008). The ACGIH identified a LOAEL of 15 mg/kg/day from this study, and a NOAEL of 1 mg/kg/day. The ACGIH stated that the NOAEL is equivalent to an inhaled concentration of 7 mg/m^3 (0.7 ppm) for a worker during a normal work day. While not stated, it is assumed the ACGIH utilized a body weight of 70 kg for a worker, and an inhalation rate of 10 m^3 for a workday, resulting in the following concentration:

1 mg/kg/day x 70 kg/10 m^3 = 7 mg/m^3

Again, while not specifically stated, it is assumed that the value of 0.7 ppm was rounded up to get a TLV of 1 ppm for hexachloroethane.

The study by Weeks et al (1979) was considered a better basis for establishing an acute ITSL for hexachloroethane than the ACGIH TLV, as the TLV was based on an oral study of repeated exposures for 16 weeks. Advantages of the Weeks et al (1979) study include the relevant route of exposure (inhalation) and the identification of adverse effects which occurred after a single exposure. While the ATSDR used the data in rats for deriving for an acute MRL, the data from the dogs is considered more appropriate for derivation of an acute ITSL. Neurological effects in rats exposed to 260 ppm in the rat teratology study used by ATSDR did not occur until day 12 of gestation (7th day of exposure), whereas these effects were observed on the first day of exposure in the dog study. The NOAEL for neurological effects from the dog study by Weeks et al is 48 ppm (465 mg/m^3), and is supported by the data in rats from this same study. Additionally, the US EPA uncertainty factors and methodology for deriving a human equivalent concentration were considered more appropriate for derivation of the acute ITSL than those of the ATSDR. Therefore, the acute ITSL is derived as follows:

$$\text{Acute ITSL} = \frac{Acute\ NOAEL_{(HEC)}}{UF_A\ x\ UF_H\ x\ UF_D}$$

Where UF_A, UF_H, and UF_D are the same uncertainty factors as used in the derivation of the RfC, and since the RGDR = 1, the $NOAEL_{(HEC)}$ is equivalent to the animal NOAEL. Therefore:

$$\text{Acute ITSL} = \frac{465\ mg/m^3}{3\ x\ 10\ x\ 10}$$

Acute ITSL = 1.6 mg/m^3 = 1600 µg/m^3

In the study by Weeks et al (1979), the dogs experienced neurological effects in a single 6-hour exposure to hexachloroethane. The acute ITSL of 1,600 µg/m^3 is based on an 8-hour averaging time. The rationale for selection of an 8-hour averaging time for the acute ITSL is: 1) typical short-term averaging times used in implementation of the Michigan Air Toxics Rules are either 8-hour or 1-hour; and 2) the combined uncertainty factor of 300 utilized in the derivation of the acute ITSL, especially considering the database UF of 10, should adequately account for the difference between a 6-hour and 8-hour averaging time.

Evaluation of the Unit Risk Factor and Risk Screening Levels

No new cancer bioassays have been done since the original unit risk values were derived by the US EPA and listed in IRIS in 1991. At that time, two oral studies were available, one in Osborne-Mendel rats and B6C3F1 mice (NCI, 1978), and the other in F344/N rats (NTP, 1989). In the NCI (1978) study, the incidence of hepatocellular carcinomas was significantly increased in male and female B6C3F1 mice, whereas no evidence of carcinogenicity was observed in either sex of Osborne-Mendel rats. In the NTP (1989) study, clear evidence of carcinogenicity was found in male rats, based on the increased incidence of kidney tumors. The NTP (1989) also found that the incidence of pheochromocytomas of the adrenal glands was significantly increased in low dose male rats compared to vehicle controls, and increased in both dose groups when compared to historical controls. No evidence of carcinogenicity was found in female F344/N rats (NTP, 1989).

The previous oral slope factor of 1.4 x 10^{-2} (mg/kg/day)$^{-1}$ listed in the IRIS database, was derived from the tumor incidence data for hepatocellular carcinoma in male and female mice

from the NCI (1978) study. The inhalation unit risk value of 4.0 x 10^{-6} $(\mu g/m^3)^{-1}$ was transformed from the oral slope factor, assuming a 70 kg person with an inhalation rate of 20 m^3/day.

The updated oral slope factor listed in the IRIS database is 4 x 10^{-2} $(mg/kg/day)^{-1}$, and is based upon the incidence of renal adenomas or carcinomas in male F344 rats from the NTP (1989) study. This slope factor was derived by fitting the multistage model to this tumor incidence data. Using the modeled results, a BMR of 0.1 was selected and divided by the $BMDL_{10}$ of 2.45 (mg/kg/day) to give an oral slope factor of 4 x 10^{-2} $(mg/kg/day)^{-1}$. Other tumor response data were also modeled but not selected for derivation of the final slope factor, either due to lack of adequate model fit or because the resulting slope factor was lower than that obtained using the male kidney tumor data. Other differences between the previous cancer risk assessment and the current one, besides the use of different tumor incidence data and modeling methodology, is the use of different animal-to-human scaling procedures. The current methodology bases dose equivalencies on the ¾ power of body weight instead of the previous ⅔ power of body weight.

The updated oral slope factor of 4 x 10^{-2} $(mg/kg/day)^{-1}$ listed in the current IRIS database was derived using the methodology provided in the US EPA's *Guidelines for Carcinogen Risk Assessment* (EPA, 2005). These guidelines represent the current state of the art methods for cancer risk assessment, and have gone through an extensive scientific review process. Therefore, the above slope factor represents the best science for assessing the risks from exposure to hexachloroethane.

No inhalation unit risk value was provided in the updated IRIS database; however, one can be derived using the oral slope factor, and assuming a 70 kg person inhales 20 m^3 of air per day as follows:

Inhalation unit risk value = 4 x 10^{-2} $(mg/kg/day)^{-1}$ x $\frac{20\ m^3/day}{70\ kg}$

Inhalation unit risk value = 1 x 10^{-2} $(mg/m^3)^{-1}$ = 1 x 10^{-5} $(\mu g/m^3)^{-1}$

Based on the above unit risk value, the resulting IRSL is 0.1 $\mu g/m^3$ and the SRSL is 1 $\mu g/m^3$. Both the IRSL and SRSL are based on an annual averaging time.

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