Proposition 65 Maximum Allowable Dose Level (MADL) for Reproductive Toxicity for Methyl Bromide as a Structural Fumigant

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Reproductive and Cancer Hazard Assessment Section

Summary

The maximum allowable dose level (MADL) for methyl bromide as a structural fumigant is **810 micrograms/day (μg/day)** by the inhalation route of exposure, derived from a developmental toxicity study in rabbits reported by Breslin et al. (1990b).

Background

This report describes the derivation of a maximum allowable dose level (MADL) for methyl bromide (CAS No. 74-83-9) used as a structural fumigant.

"Methyl bromide as a structural fumigant" was listed under Proposition 65 (the Safe Drinking Water and Toxic Enforcement Act of 1986) as known to the State to cause reproductive toxicity (developmental toxicity), effective January 1, 1993. The Proposition 65 listing of methyl bromide as a structural fumigant was based on a formal requirement by agencies of the state (the California Department of Pesticide Regulation, CDPR) and federal government (the U.S Environmental Protection Agency, U.S. EPA) that methyl bromide as a structural fumigant be labeled or identified as causing developmental toxicity (Title 22 California Code of Regulations §12902). Although methyl bromide is also used as a fumigant on raw and processed agricultural commodities, in soil, and on ornamentals (CDPR, 2002), these uses are not subject to the requirements of Proposition 65.

Procedures for the development of Proposition 65 MADLs are provided in regulations (Title 22 Cal. Code of Regs. §12801 and 12803). Exposure at a level 1,000 times greater than the MADL is expected to have no observable effect. As defined in regulations, a MADL is derived from a No Observable Effect Level (NOEL) based on the most sensitive study deemed to be of sufficient quality (Title 22 Cal. Code of Regs. § 12803(a)(4)).

Study Selection

Relevant studies on the developmental toxicity of methyl bromide have been identified through literature searches. They include studies published in the open public literature and unpublished studies previously reviewed and summarized by CDPR (2002) or by the Office of Environmental Health Hazard Assessment (OEHHA, 2000). All the studies that provided relevant information on the developmental toxicity of methyl bromide were reviewed and considered by OEHHA for the establishment of the MADL. A brief summary of major findings from several studies that provide a Lowest Observable Effect Level (LOEL) or NOEL is presented in Table 1.

Table 1. Developmental Toxicity of Methyl Bromide observed in Several Studies

Study Reference	**Animals**	**Treatment**	**Maternal Toxicity**	**Developmental Effects & LOEL**	**NOEL**
Breslin et al., 1990b	New Zealand White rabbits, 26 per group	**Inhalation**, 0, 20, 40, 80 ppm, 6 h/d, GD 7-19	Decreased body weight gains and clinical symptoms of toxicity at 80 ppm.	Decreased fetal weights; increased incidence of gall bladder agenesis & fused sternebrae. LOEL = 80 ppm (28.29 mg/kg-day)	40 ppm (14.04 mg/kg-day)
Sikov et al., 1981	Wistar rats, 38-40 per group	**Inhalation**, 0, 20, 70 ppm, 7 h/d, GD 1-GD 19. Half exposed for 3 wks before mating.	Decreased maternal body weights on GD 14 at 70 ppm.	No obvious effects on fetal weights. Increased incidence of delayed skeletal ossification. LOEL = 70 ppm (53.90 mg/kg-day)	20 ppm (15.11 mg/kg-day)
American Biogenics Corp, 1986	Sprague-Dawley rats, 25 per group. Two-generation reproductive study	**Inhalation**, 0, 3, 30, 90 ppm; 6 hr/d, 5 d/wk; no treatment from GD 21 to PND 4.	No effect on maternal body weights. Decreased brain weights in F0 males & F1 adults at 90 ppm.	Decreased pup weights in F2a on PND 0 at 90 ppm. Other findings from this study are discussed in the text below. LOEL = 90 ppm (39.89 mg/kg-day)	30 ppm (12.97 mg/kg-day)
Kaneda et al., 1998	New Zealand White rabbits, 15-18 per group	**Gavage**, 0, 1, 3, 10 mg/kg-day, GD6-18	Decreased body weight gains at high dose	No developmental effects. LOEL not found.	10 mg/kg-day (the highest dose used)
Kaneda et al., 1998	Sprague-Dawley rats, 23-24 per group	**Gavage**, 0, 3, 10, 30 mg/kg-day, GD6-15	Decreased body weight gains and erosive lesions in the stomach	No developmental effects observed. LOEL not found.	30 mg/kg-day (the highest dose used)
Kaneda et al., 1993	SD rats, 24 per group. Two-generation reproductive study	**Fumigated diets**, 0, 80, 200, 500 ppm of bromine; premating to weaning.	Decreased food consumption in F1 females in the 500-ppm group during lactation. No other obvious maternal effects.	Decreased birth weights and pup body weights in F2 females during lactation at 500 ppm. LOEL= 500 ppm (41.6 mg/kg-day)	200 ppm (17.2-22.3 mg/kg-day of bromine as reported by the authors)

Notes: 1. Abbreviations: GD: gestational day; PND: Post-natal Day; F0: parent animals; F1: First generation (offspring of parent animals); F2a: first litters from F1 parents.
2. Conversion of inhalation NOEL units to mg/kg-day: see "MADL Calculation" and Table 2 below for calculations.

The findings from the studies listed in Table 1 indicate that methyl bromide causes developmental effects in rats and rabbits following inhalation or oral exposures (Sikov et al., 1981; ABC, 1986; Breslin et al., 1990a; 1990b). It should be pointed out that significantly decreased body weights in pups from the F2 litters during the postnatal periods (PND 4-28) were observed in rats exposed to 30 ppm methyl bromide in the two-generation study conducted by the American Biogenics Corp (ABC, 1986). However, developmental endpoints resulting from post-natal exposure are not covered by Proposition 65. The decreased pup weights in the F2 litters at 30 ppm as observed in the study by ABC (1986) are excluded from consideration because of potential postnatal exposure. Therefore, the LOEL for developmental effects in the rat study by the American Biogenics Corp is based on birth weights (body weights on PND 0) of F2a litters.

For purposes of MADL development under Proposition 65, the NOEL is based on the most sensitive study deemed to be of sufficient quality and should be the highest dose level which results in no observable reproductive effect, expressed in milligrams of chemical per kilogram of bodyweight per day (Title 22 Cal. Code of Regs. §12803(a)). Therefore, the LOELs and NOELs presented as air concentrations in these studies were converted to mg/kg-day as described in the following:

Table 2. Dose Conversion from ppm Air Concentrations to mg/kg-day

	Breslin et al., 1990		**ABC, 1986**		**Sikov et al., 1981**	
	LOEL	**NOEL**	**LOEL**	**NOEL**	**LOEL**	**NOEL**
Animal species	Rabbits	Rabbits	Rats	Rats	Rats	Rats
Air concentration (ppm)	80	40	90	30	70	20
Exposure period	GD 7-19	GD 7-19	Premating-GD20	Premating-GD20	GD 1- 19	GD 1- 19
Exposure frequency	6 hrs/d, daily	6 hrs/d, daily	6 hrs/d, 5d/wk	6 hrs/d, 5d/wk	7 hr/d, daily	7 hr/d, daily
Maternal body weights[a]	4.01 kg	4.19 kg	0.349 kg	0.376 kg	0.290	0.307
Inhalation rate (m^3/day)[b]	1.458	1.512	0.223	0.234	0.197	0.204
Dose (mg/kg)[c]	28.29	14.04	55.85	18.16	53.90	15.11
Dose (mg/kg-day)	28.29	14.04	39.89[d]	12.97[d]	53.90	15.11

Notes: a. The body weights of dams on GD 20 in rabbits in the Breslin et al. study; the body weights of dams for F2a litters on GD 20 in the ABC study, and the body weights of dams on GD 14 in the Silkov et al. study.
b. Inhalation rate (IR) for rabbits was derived from body weights (W): IR (m^3/day) = $0.46*W^{0.8307}$(U. S. EPA, 1988). For rats, the method by Anderson et al. was used: IR (m^3/day) = $0.105\ [W/0.113]^{2/3}$ (Anderson et al., 1983).
c. Dose (mg/kg) = air concentration (ppm) × conversion factor (3.89) × inhalation rate (m^3/hr) × exposure hours/day ÷ weights (kg).
d. For the ABC study, administered dose multiplied by 5/7 to account for 5 days/week dosing.

The developmental study in rabbits by Breslin et al. (1990b) is "the most sensitive study deemed to be of sufficient quality" for the purposes of Proposition 65 (developmental effect from prenatal exposure). Thus, the NOEL (40 ppm, equivalent to 14.04 mg/kg-day) is used as the basis for establishing the MADL for "methyl bromide as a structural fumigant."

In the study by Breslin et al. (1990b), pregnant New Zealand white rabbits, 15-21 animals per group, were exposed to methyl bromide (99.6% pure) at concentrations of 0, 20, 40, or 80 ppm for six hours per day by inhalation from days 7 to 19 of gestation. One pregnant rabbit from the 80 ppm exposure group died during the exposure period. All survival animals were sacrificed on GD 28 for necropsy. In the 80-ppm group, fetal body weights were significantly reduced; the incidences of gall bladder agenesis and fused sternebrae were significantly higher than those of the controls. The authors reported no apparent fetal malformations or variations in the 20 ppm or 40 ppm exposure groups. Thus, 40 ppm, equivalent to 14.04 mg/kg-day, is considered to be the NOEL and is used for calculation of the MADL.

MADL Calculation

The NOEL is the highest dose level which results in no observable reproductive effect, expressed in milligrams of chemical per kilogram of bodyweight per day (Title 22, Cal. Code of Regs., §12803(a)(1)). The NOEL is converted to a milligram per day dose level by multiplying the assumed human body weight by the NOEL (Title 22, Cal. Code of Regs., §12803(b)). For developmental toxicity, the assumed body weight of a pregnant woman is 58 kg.

For inhalation exposure, the MADL for "methyl bromide as a structural fumigant" was calculated as follows, based on a NOEL of 40 ppm found in the study in rabbits by Breslin et al. (1992b):

Conversion of air concentration in ppm to mg/m^3 using a conversion factor of 3.89 (CDPR, 2002):

$$40 \text{ ppm} \times 3.89 = 155.60 \text{ mg/m}^3$$

Conversion of air concentration (mg/m^3) from 6 hours per day to the equivalent concentration for 24 hours per day:

$$155.60 \text{ mg/m}^3 \times 6 \text{ hr} \div 24 \text{ hr} = 38.90 \text{ mg/m}^3$$

Calculation of the NOEL expressed as mg/kg-day, based on the reported body weight of 4.19 kg of pregnant rabbits in the group exposed to 40 ppm methyl bromide on GD 20, using the inhalation rate of 1.512 m^3/day (see footnote b to Table 2 above)):

$$(38.90 \text{ mg/m}^3 \times 1.512 \text{ m}^3\text{/day}) \div (4.19 \text{ kg}) = 14.04 \text{ mg/kg-day}$$

Calculation of the NOEL in mg/day for a 58 kg pregnant woman:

$$14.04 \text{ mg/kg-day} \times 58 \text{ kg} = 814.30 \text{ mg/day}$$

The MADL is derived by dividing the NOEL by one thousand (Title 22, Cal. Code of Regs., §12801(b)(1)). Thus, the adjusted NOEL was divided by 1,000 to obtain the MADL.

$\mathbf{MADL_{inhalation}}$ = 814.30 mg/day ÷ 1000 = 814.30 µg/day, or **810 µg/day** after rounding.

This MADL represents intake by the inhalation route of exposure. Approximately 50% of administered doses of methyl bromide were absorbed following inhalation administration (OEHHA, 2000; CDPR, 2002). Thus the MADL for the inhalation route of exposure (810 µg/day) is equivalent to an absorbed dose of approximately 400 µg/day.

The MADL of 810 µg/day is applicable to exposure via inhalation only. If a source or product results in exposures by non-inhalation or multiple routes, the total exposure to the chemical from the source or product must be considered. The absorbed dose resulting from any one or multiple routes of exposure should be calculated. If the total absorbed dose resulting from any one or multiple routes is less than or equal to 400 µg/day, the MADL has not been exceeded.

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