Methanol; CASRN 67-56-1

Human health assessment information on a chemical substance is included in IRIS only after a comprehensive review of toxicity data by U.S. EPA health scientists from several program offices, regional offices, and the Office of Research and Development. Sections I (Health Hazard Assessments for Noncarcinogenic Effects) and II (Carcinogenicity Assessment for Lifetime Exposure) present the positions that were reached during the review process. Supporting information and explanations of the methods used to derive the values given in IRIS are provided in the guidance documents located on the IRIS website at www.epa.gov/iris/backgrd.html.

STATUS OF DATA FOR Methanol

File First On-Line 09/07/1988

Category (section)	Assessment Available?	Last Revised
Oral RfD (I.A.)	yes	09/30/2013
Inhalation RfC (I.B.)	yes	09/30/2013
Carcinogenicity Assessment (II.)	not evaluated	

I. Chronic Health Hazard Assessments for Noncarcinogenic Effects

I.A. Reference Dose for Chronic Oral Exposure (RfD)

Substance Name — Methanol
CASRN — 67-56-1
Last Revised — 09/30/2013

The RfD is an estimate (with uncertainty spanning perhaps an order of magnitude) of a daily oral exposure to the human population (including sensitive subgroups) that is likely to be without an appreciable risk of deleterious effects during a lifetime. Diet can contribute to background levels of methanol, principally from the ordinary ingestion of fruits and vegetables. Thus, in the case of methanol, the RfD is further defined as an exogenous exposure (exposure from a source outside the body) that adds to background levels of methanol derived from a diet that includes fruits and vegetables (see further discussion in Section I.A.4). The RfD is intended for use in risk assessments for health effects known or assumed to be produced through a nonlinear (presumed threshold) mode of action. It is expressed in units of mg/kg day. Please refer to the guidance

documents at http://www.epa.gov/iris/backgrd.html for an elaboration of these concepts. Because RfD values can be derived for the noncarcinogenic health effects of substances that are also carcinogens, it is essential to refer to other sources of information concerning the carcinogenicity of this chemical substance. If the U.S. EPA has evaluated this substance for potential human carcinogenicity, a summary of that evaluation will be contained in Section II of this file.

The RfD of 2 mg/kg-day replaces the previous RfD of 0.5 mg/kg-day entered on the IRIS database on 09/07/1988. The previous RfD was based on a no-observed-adverse-effect level (NOAEL) of 500 mg/kg-day for liver enzyme changes and brain weight reduction in a subchronic oral gavage study of Sprague-Dawley rats (TRL, 1986), and a composite uncertainty factor (UF) of 1,000 (10 for extrapolation from rats to humans, 10 for human variation, and 10 for subchronic to chronic extrapolation).

I.A.1. Oral RfD Summary

Critical Effect	Point of Departure (POD)	UF	POD/UF	Chronic RfD[a]
Extra cervical ribs **CD-1 mice** **Inhalation developmental toxicity study; exposure during gestation days GD7-GD17** **Rogers et al. (1993b)**	$POD_{Internal}$ = 43.1 mg/L	100	0.43 mg/L	2 mg/kg/day

[a]The RfD is the oral dose predicted to yield a methanol blood concentration equal to the $RfD_{internal}$ ($POD_{Internal}$/UF) of 0.43 mg/L, using the human PBPK model described in Appendix B of the *Toxicological Review of Methanol (Noncancer)* (U.S. EPA, 2013); the final RfD is rounded to one significant figure.

I.A.2. Principal and Supporting Studies (Oral RfD)

EPA has derived an RfD by using exposure-response data from candidate principal inhalation studies of mice (Rogers et al., 1993b) and rats (NEDO, 1987) and route-to-route extrapolation with the aid of the EPA physiologically based pharmacokinetic (PBPK) model [see Section 3.4 of the *Toxicological Review of Methanol (Noncancer)* (U.S. EPA, 2013)]. The decision to use inhalation rather than oral study data is due to limitations in the database of oral studies, including the limited reporting of noncancer findings in the subchronic (TRL, 1986) and chronic

oral studies (Soffritti et al., 2002) of rats, the determination that developmental effects are the most sensitive effects of methanol exposure [see Section 5.1.1 of the *Toxicological Review of Methanol (Noncancer)* (U.S. EPA, 2013)], and the high-dose levels used in the rodent oral developmental studies [see Section 5.2 of the *Toxicological Review of Methanol (Noncancer)* (U.S. EPA, 2013)]. The candidate principal studies for the derivation of an RfC and RfD are summarized below and are described in more detail in Sections 4.3.2 and 4.4.2 of the *Toxicological Review of Methanol (Noncancer)* (U.S. EPA, 2013).

Rogers et al. (1993b) evaluated development toxicity in pregnant female CD-1 mice exposed to air or 1,000, 2,000, 5,000, 7,500, 10,000, or 15,000 ppm (0, 1,310, 2,620, 6,552, 9,894, 13,104, and 19,656 mg/m^3) methanol vapors (≥99.9% purity) in a chamber for 7 hours/day on GD6 GD15. There were no methanol-related reductions in maternal body weight gain or overt signs of toxicity. Dams were sacrificed on GD17 for a comparison of developmental toxicity in methanol-treated groups versus the chamber air-exposed control group. Fetuses in all exposure groups were weighed, assessed for viability, and examined for external malformations. Fetuses in the control, 1,000, 2,000, 5,000, and 15,000 ppm groups were also examined for skeletal and visceral defects. Reproductive and fetal effects included an increase in the number of resorbed litters, a reduction in the number of live pups, and increased incidences of exencephaly, cleft palate, and the number of cervical ribs. The incidences of these effects are listed in Table 4-4 of the *Toxicological Review of Methanol (Noncancer)* (U.S. EPA, 2013). As described below, the increase in cervical ribs/litter reported in this study was evaluated for possible use in the derivation of RfD and RfC values.

NEDO (1987) evaluated the effects of pre- and postnatal methanol (reagent grade) exposure (20 hours/day) on reproductive and other organ systems of Sprague-Dawley rats. In a two-generation study, F_0 generation rats (30 males and 30 females per exposure group) were exposed to 0, 10, 100, and 1,000 ppm (0, 13.1, 131, and 1,310 mg/m^3) from 8 weeks old to the end of mating (males) or to the end of lactation period (females). The F_1 generation was exposed to the same concentrations from birth to the end of mating (males) or to weaning of F_2 pups 21 days after delivery (females). Males and females of the F_2 generation were exposed from birth to 21 days old (one animal/sex/litter was exposed to 8 weeks of age). NEDO (1987) noted reduced brain, pituitary, and thymus weights, and early testicular descent in the offspring of F_0 and F_1 rats exposed to 1,000 ppm methanol. To confirm the possible compound-related effect of methanol on the brain, NEDO (1987) performed an additional study in which Sprague-Dawley rats were exposed to 0, 500, 1,000, and 2,000 ppm (0, 655, 1,310, and 2,620 mg/m^3) methanol from the first day of gestation through the F_1 generation. The number of F_0 parental animals included per group in this supplemental experiment was not reported. However, the number of pups per dose group per "period after birth" was reported as 11-14/sex/dose/postnatal period, and it is reasonable to assume that, consistent with the standard culling protocol used for both the F_1 and F_2 generations of the two-generation study (NEDO, 1987 pages 185 and 189), the pups for each gender, dose and exposure time combination came from a different litter (to avoid problems associated with litter correlation). Dose-related decreases in brain weights were observed in the male and female offspring at 3, 6, and 8 weeks of age. As described below, brain weight changes observed in these NEDO (1987) studies were evaluated for possible use in the derivation of RfD and RfC values.

Developmental Effects in Principal Studies

Skeletal effects have been observed in developmental studies of rats (Weiss et al., 1996; NEDO, 1987; Nelson et al., 1985) and mice (Bolon et al., 1993; Rogers et al., 1993b). The findings of Bolon et al. (1993) and Rogers and Mole (1997) indicate that methanol is toxic to mouse embryos in the early stages of organogenesis, on or around GD7. Rogers et al. (1993b) reported a NOAEL for the incidence of extra cervical ribs at 1,000 ppm (1,310 mg/m^3, 33.6% per litter) and a LOAEL of 2,000 ppm (2,620 mg/m^3, 49.6% per litter) when compared to controls (28.0% per litter). Increased incidence of cervical ribs was also observed in the rat organogenesis study (NEDO, 1987) in the 5,000 ppm dose group (65.2% per litter versus 0% in the control group), indicating that the endpoint is significant across species. There is evidence that incidence of supernumerary ribs (including cervical ribs) is related to a general alteration in the development and architecture of the axial skeleton as a whole. In CD-1 mice exposed during gestation to various types of stress, food and water deprivation, and the herbicide dinoseb, supernumerary ribs were consistently associated with increases in length of the 13th rib (Branch et al., 1996). This relationship was present in all fetal ages examined in the study. These findings are consistent with supernumerary ribs being one manifestation of a basic alteration in the differentiation of the thoraco-lumbar border of the axial skeleton. The biological significance of this endpoint is further strengthened by the association of supernumerary ribs with adverse health effects in humans. The most common effect produced by the presence of cervical ribs is thoracic outlet disease (Nguyen et al., 1997; Fernandez Noda et al., 1996; Henderson, 1914). Thoracic outlet disease is characterized by numbness and/or pain in the shoulder, arm, or hands. Vascular effects associated with this syndrome include cerebral and distal embolism (Bearn et al., 1993; Connell et al., 1980; Short, 1975), while neurological symptoms include extreme pain, migraine, and symptoms similar to Parkinson's (Evans, 1999; Saxton et al., 1999; Fernandez Noda et al., 1996). Schumacher et al. (1992) observed 242 rib anomalies in 218 children with tumors (21.8%) and 11 (5.5%) in children without malignancy, a statistically significant ($p < 0.001$) difference that indicates a strong association between the presence of cervical ribs and childhood cancers.

Brain weight changes observed in the NEDO (1987) developmental studies are also deemed biologically significant and relevant to humans. Decreases in brain weight have been associated with simultaneous deficits in neurobehavioral and cognitive parameters in animals exposed during gestation to various solvents, including toluene and ethanol (Gibson et al., 2000; Coleman et al., 1999; Hass et al., 1995). Further, a change in absolute brain weight alone is considered to be a biologically significant effect (U.S. EPA, 1998a). This is true regardless of changes in body weight because brain weight is generally protected during malnutrition or weight loss, unlike many other organs or tissues (U.S. EPA, 1998a). While brain weight reduction has not been reported in other developmental bioassays, it has been observed in adult rats exposed to methanol (TRL, 1986), and there are indications of possible developmental neurobehavioral effects associated with methanol inhalation exposure to monkeys (2004a; 2004b; Burbacher et al., 1999a; 1999b).

The NEDO (1987) developmental studies indicate that both gestational and postnatal exposure to methanol contribute to the brain weight decreases observed in Sprague-Dawley rat pups. This

finding is not unexpected, given that the rat brain undergoes tremendous growth beginning early in gestation and continuing in the postnatal period. Rats are considered altricial (i.e., born at relatively underdeveloped stages), and many of their neurogenic events occur postnatally (Clancy et al., 2007). However, brain effects from postnatal exposure are also relevant to humans given that, in humans, gross measures of brain growth increase for at least 2-3 years after birth, with the growth rate peaking approximately 4 months after birth (Rice and Barone, 2000).

Selection of Critical Effects

Taking into account the advantages and limitations of the studies available for quantification purposes and the relative sensitivities for the effects observed (see Section 5.1.1 of the *Toxicological Review of Methanol (Noncancer)* (U.S. EPA, 2013) for details), two developmental effect endpoints were chosen as co-critical effects for the purposes of this dose-response assessment, cervical rib anomalies in fetal CD-1 mice (Rogers et al., 1993b) and decreased brain weight in male Sprague-Dawley rats exposed throughout gestation and lactation (NEDO, 1987). These endpoints can be reliably quantified and represent adverse effects in two separate sensitive organ systems at key periods of their development. RfC derivations for these endpoints using various derivation options are summarized below and in Appendix D of the *Toxicological Review of Methanol (Noncancer)* (U.S. EPA, 2013). As discussed in Sections I.A.3 and I.A.4 below and in Sections 5.1.3.1 and 5.3.1 of the *Toxicological Review of Methanol (Noncancer)* (U.S. EPA, 2013), the monkey studies of Burbacher et al. (2004a; 2004b; 1999a; 1999b) and NEDO (1987) are qualitatively supportive, but are inadequate or inferior to the candidate principal rodent studies for quantitative dose-response analysis.

Methods of Analysis. Inhalation studies considered for derivation of the RfC are used to supplement the oral database using route-to-route extrapolation, as previously described. Benchmark dose (BMD) approaches were applied to the existing inhalation database, and the EPA PBPK model was used for species-to-species extrapolations. For the BMD analyses of the rat brain weight endpoint following gestational and lactational exposure, PBPK model estimates of AUC (mg-hr/L) methanol in blood for the dams of each dose group were used as the dose metric due to evidence that fetal and neonatal brain weight is susceptible to both the level and duration of methanol exposure. For the BMD analyses of the mouse cervical rib endpoint, internal C_{max} (mg/L) methanol blood concentrations reported by Rogers et al.(1993b) for mouse dams at day 6 of gestation were used as the modeled dose metric because the small gestational window of susceptibility for this endpoint (Rogers and Mole, 1997; Bolon et al., 1993) suggest that the level of exposure is more important than the duration of exposure.

Appendix D of the *Toxicological Review of Methanol (Noncancer)* (U.S. EPA, 2013) contains a description of the developmental endpoints and BMD modeling approaches used to estimate an internal dose BMD lower confidence limit (BMDL) point of departure ($POD_{internal}$) for each candidate endpoint. Appendix B of the *Toxicological Review of Methanol (Noncancer)* (U.S. EPA, 2013) describes PBPK models used to estimate a candidate RfD value for each $POD_{internal}$. As described in Section 5.2.2 of the *Toxicological Review of Methanol (Noncancer)* (U.S. EPA, 2013), slightly lower candidate RfDs were derived for extra cervical ribs in mice exposed during gestation days 6-16 (Rogers et al., 1993b) than for decreased male brain weight in rats exposed

throughout gestation and the F_1 generation (NEDO, 1987). Consequently, a $BMDL_{05}$ for the cervical rib endpoint of 43 mg/L methanol in blood serves as the $POD_{Internal}$ for the RfD derivation.

Because the same data set, endpoints, BMD methods and PBPK models used to derive the RfC were also used to calculate the candidate RfD values, the RfD derivation uses the same uncertainty factors as are described for the RfC derivation (Section 5.1.3.2 of the *Toxicological Review of Methanol (Noncancer)* (U.S. EPA, 2013). Consistent with the RfC derivation, in order to avoid the uncertainty associated with applying the human PBPK model to exposure levels that are above the levels for which the model was calibrated and to account for possible non-linearities in the external versus internal dose relationships at high doses, EPA applied the total 100-fold UF to the internal BMDL ($POD_{internal}$) prior to HED derivation to obtain an $RfD_{internal}$ [see Table 5-6 of the *Toxicological Review of Methanol (Noncancer)* (U.S. EPA, 2013)].

RfD = 43.1 mg/L ÷ 100 = 0.43 mg/L ==>PBPK==> 2 mg/kg-day
(rounded to 1 significant figure)

I.A.3. Uncertainty and Modifying Factors (Oral RfD)

UF = 100
= 10 (UF_H) × 3(UF_A) × 3(UF_D).

Interindividual variation UF_H. An uncertainty factor of 10 was applied to account for variation in sensitivity within the human population (UF_H). The UF_H of 10 is commonly considered to be appropriate in the absence of convincing data to the contrary. The data from which to determine the potential extent of variation in how humans respond to chronic exposure to methanol are limited, given the complex nature of the developmental endpoint employed and uncertainties surrounding the importance of metabolism to the observed teratogenic effects. Susceptibility to methanol is likely to involve intrinsic and extrinsic factors. Some factors may include alteration of the body burden of methanol or its metabolites, sensitization of an individual to methanol effects, or augmentation of underlying conditions or changes in processes that share common features with methanol effects. Additionally, inherent differences in an individual's genetic make-up, diet, gender, age, or disease state may affect the pharmacokinetics and pharmacodynamics of methanol, influencing susceptibility intrinsically. Co-exposure to a pollutant that alters metabolism or other clearance processes, or that adds to background levels of metabolites may also affect the pharmacokinetics and pharmacodynamics of methanol, influencing susceptibility extrinsically [see Section 4.9 of the *Toxicological Review of Methanol (Noncancer)* (U.S. EPA, 2013)]. The determination of the UF for human variation is supported by several types of information, including information concerning background levels of methanol in humans, variation in pharmacokinetics revealed through human studies and from PBPK modeling, variation of methanol metabolism in human tissues, and information on physiologic factors (including gender and age), or acquired factors (including diet and environment) that may affect methanol exposure and toxicity (see Section 5.1.3.2.1 of the *Toxicological Review of Methanol (Noncancer)* (U.S. EPA, 2013) for further details).

Animal-to-human extrapolation UF_A. A factor of 3 was applied to account for uncertainties in extrapolating from rodents to humans (UF_A). Application of a full UF of 10 would depend on two areas of uncertainty: toxicokinetic and toxicodynamic. The rodent-to-human toxicodynamic uncertainty is addressed by a factor of 3, as is the practice for deriving RfCs (U.S. EPA, 1994b). In this assessment, the toxicokinetic component of uncertainty is addressed by the determination of a HEC through the use of PBPK modeling. Use of PBPK-estimated maternal blood methanol levels for the estimation of HECs allows for the use of data-derived extrapolations rather than standard methods for extrapolations from external exposure levels. Although uncertainties exist, the PBPK modeling approach employed is considered to be sufficient to allow for reduction of the toxicokinetic uncertainty to a value of 1 for both of the candidate principal studies [see Section 5.1.3.2.2 of the *Toxicological Review of Methanol (Noncancer)* (U.S. EPA, 2013) for further details].

Database UF_D. A database UF of 3 was applied to account for deficiencies in the toxicity database (UF_D). While the database for methanol toxicity is extensive in terms of the laboratory species and study design coverage, consisting of chronic and developmental toxicity studies in rats, mice, and monkeys, a two-generation reproductive toxicity study in rats, and neurotoxicity and immunotoxicity studies, there still remains considerable uncertainty with respect to the potency, importance and relevance of reproductive, developmental and chronic effects observed in monkeys. As discussed in Section 5.1.1.1 of the *Toxicological Review of Methanol (Noncancer)* (U.S. EPA, 2013), chronic and developmental studies in monkeys, the species most likely to best represent the potential for developmental effects in humans, were considered inadequate or inferior to the candidate principal rodent studies for the purposes of RfC/D derivation. The lack of a quantifiable monkey study is an important data gap given the potential relevance to humans and the uncertainties raised by existing monkey studies regarding this species sensitivity to reproductive effects [e.g., shortened pregnancies discussed in Section 4.3.2 of the *Toxicological Review of Methanol (Noncancer)* (U.S. EPA, 2013)], CNS degeneration [e.g., stellate cell fibrosis described in Section 4.4.2 of the *Toxicological Review of Methanol (Noncancer)* (U.S. EPA, 2013)] and delayed neurobehavioral development [e.g., VDR response described in Section 4.4.2 of the *Toxicological Review of Methanol (Noncancer)* (U.S. EPA, 2013)] from methanol exposure. Due to the substantial uncertainty associated with the reproductive and chronic effects in monkeys, those data are not as informative or reliable for the purpose of establishing the appropriate UF_D level. However, the developmental neurotoxicity data are comparable across the two species and, of the uncertain effects observed in monkeys, the results of the visually directed reaching (VDR) test are likely to be the most reliable, discernible and relevant. A comparison of the lowest methanol blood LOAELs (excluding background) observed in rodent and monkey developmental neurotoxicity studies indicates that the rodent LOAEL blood level is 12-fold higher than the monkey LOEL blood level. Some of this 12-fold difference may be due to differences in species sensitivity, for which the UF_A of 3-fold is intended to account, but some of the difference may be due to other factors, including whether appropriate and comparable endpoints were examined and whether appropriate study designs and quality control measures were used. To account for these additional factors, a 3-fold UF_D is applied. [see Section 5.1.3.2.3 of the *Toxicological Review of Methanol (Noncancer)* (U.S. EPA, 2013) for further details].

Extrapolation from subchronic to chronic UF_S. A UF of 1 was applied for extrapolation from less than chronic results because developmental toxicity (cervical rib and decreased brain weight) was used as the critical effect. The developmental period is recognized as a susceptible lifestage where exposure during certain time windows is more relevant to the induction of developmental effects than lifetime exposure (U.S. EPA, 1991).

LOAEL-to-NOAEL extrapolation UF_L. A UF of 1 was applied for LOAEL to NOAEL (UF_L) because the current approach is to address this extrapolation as one of the considerations in selecting a benchmark response (BMR) for BMD modeling. In this case, the endpoint and benchmark response level employed for the RfD/C derivation is appropriate for use in deriving the RfD under the assumption that it represents a minimal biologically significant change.

I.A.4. Additional Studies/Comments (Oral RfD)

As discussed above and in greater detail in Sections 5.1.1 and 5.2.1 of the *Toxicological Review of Methanol (Noncancer)* (U.S. EPA, 2013), developmental effects observed in two candidate principal rodent studies were considered relevant and quantifiable for the purposes of RfC/D derivation. Uncertainties associated with choice of study/endpoint, BMD modeling, route-to-route extrapolation, choice of species/gender and the relationship of the RfC and RfD to endogenous methanol blood levels are discussed in detail in Section 5.3 of the *Toxicological Review of Methanol (Noncancer)* (U.S. EPA, 2013).

While insufficient for use as a quantitative basis for the RfC and RfD, monkey reproductive and neurotoxicity developmental studies (2004a; 2004b; Burbacher et al., 1999a; 1999b) and monkey chronic studies (NEDO, 1987) provide important supportive information for the RfD and RfC derivation. Burbacher et al. (2004a; 2004b; 1999a; 1999b) exposed *M. fascicularis* monkeys to air concentrations of 0, 200, 600, or 1,800 ppm (0, 262, 786, and 2,359 mg/m^3) methanol, 2.5 hours/day, 7 days/week during premating/mating and throughout gestation (approximately 168 days). They observed a statistically significant delay in visually directed reaching (VDR) in the 600m ppm (786 mg/m^3) group for males and the 1,800 ppm (2,359 mg/m^3) group for both sexes. However, a dose-response trend for this endpoint was only exhibited for females. Another test, the Fagan test of infant intelligence, indicated small but not significant deficits of performance (time spent looking at novel faces versus familiar faces) in treated monkeys. As discussed in Section 4.6.1.2 of the *Toxicological Review of Methanol (Noncancer)* (U.S. EPA, 2013), the monkey data are not conclusive, and there is insufficient evidence to determine if the primate fetus is more or less sensitive than rodents to methanol teratogenesis. Taken together, however, the NEDO (1987) rat study and the Burbacher et al. (2004a; 2004b; 1999a; 1999b) monkey study suggest that prenatal exposure to methanol can result in adverse effects on developmental neurology pathology and function, which can be exacerbated by continued postnatal exposure. Burbacher et al. (2004a; 2004b; 1999a; 1999b) also reported a shorter period of gestation in all exposure groups that did not appear to be dose related. As discussed in Section 4.6.1.2 of the *Toxicological Review of Methanol (Noncancer)* (U.S. EPA, 2013), though statistically significant, the shortened gestation finding may be of limited biological significance given questions concerning its relation to the methanol exposure.

In the NEDO (1987) chronic inhalation monkey studies, 8 animals (sex unspecified) were exposed to 10, 100, or 1,000 ppm (13, 131, and 1,310 mg/m^3) methanol, 21 hours/day, for 7 months (2 animals), 19 months, (3 animals), or 29 months (3 animals). There was no indication in the NEDO (1987) report that this chronic study employed a concurrent control group. As described in Section 4.4.2 of the *Toxicological Review of Methanol (Noncancer)* (U.S. EPA, 2013), NEDO (1987) reported histopathologic changes to the peripheral and central nervous system (CNS) of exposed monkeys that increased with increasing exposure. The most pervasive effect noted across the exposure concentrations and durations was "fibrosis of responsive stellate cells," characterized as "neurological disease" in the NEDO (1987) summary report. These "stellate cells" are likely to be astrocytes, star-shaped glial cells in the brain that are among the most numerous cells in all regions of the CNS. The limited information available from the NEDO (1987) summary report suggests that 100 ppm (131 mg/m^3) may be an effect level following continuous, chronic exposure to methanol. However, as noted in Section 4.2.2.1 of the *Toxicological Review of Methanol (Noncancer)* (U.S. EPA, 2013), the NEDO (1987) studies in nonhuman primates, have multiple reporting deficiencies and data gaps that make them difficult to interpret. In addition, confidence in the dose-response data from this study is weakened by the apparent lack of a concurrent control group and the small number of animals at each exposure level for each serial sacrifice (2-3 monkeys/time point/exposure level). In general, peer reviewers of this study felt that it provides descriptive, rather than quantitative, support for the evaluation of the inhalation toxicity of methanol (ERG, 2009).

A number of studies described in Section 4.3.2 and summarized in Section 4.6.1.2 of the *Toxicological Review of Methanol (Noncancer)* (U.S. EPA, 2013) have examined the potential toxicity of methanol to the male reproductive system (Lee et al., 1991; Cameron et al., 1985; Cameron et al., 1984). Some of the observed effects, including a transient decrease in testosterone levels, could be the result of chemically related strain on the rat system as it attempts to maintain hormone homeostasis. However, the data are insufficient to definitively characterize methanol as a toxicant to the male reproductive system.

Relationship of RfD/C to background methanol blood levels and monkey blood levels associated with effects of uncertain adversity

In Section 5.3.6 of the *Toxicological Review of Methanol (Noncancer)* (U.S. EPA, 2013), PBPK model predictions for the expected increase in methanol levels in blood resulting from exposure to methanol at the level of the RfC or RfD are compared to background blood levels of methanol estimated from (1) daily endogenous production and dietary exposure estimates from the U.K. report (COT, 2011) and (2) a sample background distribution derived from relevant study groups. Both the EPA and the U.K. data are consistent with approximately 2.5 mg/L representing a high end of the range of background methanol blood levels associated with a diet that includes fruits and vegetables. EPA estimates that the shift in EPA's sample background methanol blood level distribution that would be associated with daily exposures of the entire population to methanol at the RfC or the RfD would increase the number of individuals with peak methanol blood levels at or above 2.5 mg/L. from ~7% to ~14%.. EPA's PBPK model predicts that a continuous daily methanol exposure at the RfD or RfC would raise the peak methanol blood level of an individual with an background methanol blood level of 2.5 mg/L to just under 3

mg/L. As discussed in Section 5.3.7 of the *Toxicological Review of Methanol (Noncancer)* (U.S. EPA, 2013), this 3 mg/L methanol blood level is at the low end of the range of methanol blood levels that have been reported in monkey chronic and gestational exposure studies to be associated with CNS and reproductive/developmental effects of uncertain, but potential adversity.

I.A.5. Confidence in the Oral RfD

Study — Medium to High
Database — Medium
RfD — Medium to High

The confidence in the RfD is medium to high. Confidence in the Rogers et al. (1993b) study is high and confidence in the NEDO (1987) developmental studies is medium. The Rogers et al. (1993b) study was well designed, including large sample sizes, well documented, peer reviewed and published. While there are issues with the lack of detail regarding methods and results in the NEDO (1987) report, the observed effect (brain weight reduction) is a relevant endpoint that has been reproduced in an oral study of adult rats (TRL, 1986), and the exposure regimen involving pre- and postnatal exposures addresses a potentially sensitive human subpopulation. Thus, the overall confidence in the two critical studies is medium to high. Confidence in the database is medium. Despite the fact that skeletal and brain effects have been demonstrated and corroborated in multiple animal studies in rats, mice, and monkeys, some study results were not quantifiable, there is uncertainty regarding which is the most relevant test species, and there is limited data regarding reproductive or developmental toxicity of methanol in humans. There is also uncertainty regarding the potential active agent—the parent compound, methanol, formaldehyde, formic acid or some other (e.g., reactive oxygen) species. There are deficiencies in the knowledge of the metabolic pathways of methanol in the human fetus during early organogenesis, when the critical effects can be induced in animals. Thus, the medium-to-high confidence in the principal studies and the medium confidence in the database together warrant an overall confidence descriptor of medium to high. Confidence in the RfD is slightly lower than for the RfC due to the lack of adequate oral studies for the RfD derivation, necessitating a route-to-route extrapolation.

I.A.6. EPA Documentation and Review of the Oral RfD

Source Document - *Toxicological Review of Methanol (Noncancer)* (U.S. EPA, 2013)

This document has been provided for review to EPA scientists, interagency reviewers from other federal agencies and the Executive Office of the President, and the public, and peer reviewed by independent scientists external to EPA. A summary and EPA's disposition of the comments received from the independent external peer reviewers and from the public is included in Appendix A of the *Toxicological Review of Methanol (Noncancer)* (U.S. EPA, 2013).

Agency Completion Date - 09/30/2013

I.A.7. EPA Contacts (Oral RfD)

Please contact the IRIS Hotline for all questions concerning this assessment or IRIS, in general, at (202)566-1676 (phone), (202)566-1749 (FAX) or hotline.iris@epa.gov (internet address).

I.B. Reference Concentration for Chronic Inhalation Exposure (RfC)

Substance Name — Methanol
CASRN — 67-56-1
Section I.B. Last Revised — 09/30/2013

The RfC is an estimate (with uncertainty spanning perhaps an order of magnitude) of a continuous inhalation exposure to the human population (including sensitive subgroups) that is likely to be without an appreciable risk of deleterious effects during a lifetime. Diet can contribute to background levels of methanol, principally from the ordinary ingestion of fruits and vegetables. Thus, in the case of methanol, the RfC is further defined as an exogenous exposure (exposure from a source outside the body) that adds to background levels of methanol derived from a diet that includes fruits and vegetables (see further discussion in Section I.B.4). The RfC considers toxic effects for both the respiratory system (portal of entry) and for effects peripheral to the respiratory system (extrarespiratory effects). The inhalation RfC (generally expressed in units of mg/m^3) is analogous to the oral RfD and is similarly intended for use in risk assessments for health effects known or assumed to be produced through a nonlinear (presumed threshold) mode of action.

Inhalation RfCs are derived according to *Methods for Derivation of Inhalation Reference Concentrations and Application of Inhalation Dosimetry* (U.S. EPA, 1994b). Because RfCs can also be derived for the noncarcinogenic health effects of substances that are carcinogens, it is essential to refer to other sources of information concerning the carcinogenicity of this chemical substance. If the U.S. EPA has evaluated this substance for potential human carcinogenicity, a summary of that evaluation will be contained in Section II of this file.

An inhalation assessment for methanol was not previously available on IRIS.

I.B.1. Chronic Inhalation RfC Summary

Critical Effect	Point of Departure (POD)	UF	POD/UF	Chronic RfC[a]
Reduced brain weight in rat pups at 6 weeks of age	$POD_{Internal}$ = 858 mg-hr/L	100	8.58 mg-hr/L	2×10^{1} mg/m^3

Critical Effect	Point of Departure (POD)	UF	POD/UF	Chronic RfC[a]
Male Sprague-Dawley rats Developmental inhalation exposure through gestation and 3, 6 or 8 weeks postnatal **NEDO (1987)**				

[a]The RfC is the inhalation concentration predicted to yield a methanol blood concentration equal to the $RfC_{internal}$ ($POD_{Internal}$/UF) of 8.58 mg-hr/L, using the human PBPK model described in Appendix B of the *Toxicological Review of Methanol (Noncancer)* (U.S. EPA, 2013); the final RfC is rounded to one significant figure.

I.B.2. Principal and Supporting Studies (Inhalation RfC)

EPA has derived an RfC by using response data from candidate principal inhalation studies of mice (Rogers et al., 1993b) and rats (NEDO, 1987) [see Section 5.1 of the *Toxicological Review of Methanol (Noncancer)* (U.S. EPA, 2013)]. These candidate principal studies and associated developmental skeletal (extra cervical ribs) and neurological (reduced brain weight) effects identified for the derivation of an RfC and RfD are summarized in Section I.A.2 above.

Methods of Analysis. As described in Section I.A.2 above, PBPK model estimates of daily AUC (mg-hr/L) methanol in the blood of the NEDO (1987) rat dams was used as the dose metric for the BMD analyses of the rat brain weight endpoint, and internal C_{max} (mg/L) methanol blood concentrations reported by Rogers et al.(1993b) for mouse dams at day 6 of gestation was used as the dose metric for the BMD analyses of the mouse cervical rib endpoint. Appendix D of the *Toxicological Review of Methanol (Noncancer)* (U.S. EPA, 2013) contains a description of the developmental endpoints and BMD modeling approaches used to estimate an internal dose BMDL point of departure ($POD_{internal}$) for each candidate endpoint. Appendix B of the *Toxicological Review of Methanol (Noncancer)* (U.S. EPA, 2013) describes PBPK models used to estimate a candidate RfC value for each $POD_{internal}$. As described in Section 5.1.3 of the *Toxicological Review of Methanol (Noncancer)* (U.S. EPA, 2013), slightly lower candidate RfCs were derived for decreased male brain weight in rats exposed throughout gestation and the F_1 generation (NEDO, 1987) than for extra cervical ribs in mice exposed during gestation days 6-16 (Rogers et al., 1993b). Consequently, the BMDL for a one standard deviation reduction in brain weight ($BMDL_{1SD}$) in male rats of 858 mg-hr/L methanol in blood serves as the $POD_{Internal}$ for the RfC derivation.

As described in Section 5.1.3.2 of the *Toxicological Review of Methanol (Noncancer)* (U.S. EPA, 2013), in order to avoid the uncertainty associated with applying the human PBPK model to exposure levels that are above the levels for which the model was calibrated and to account for possible non-linearities in the external versus internal dose relationships at high doses, EPA applied the total 100-fold UF to the internal BMDL ($POD_{internal}$) to obtain an $RfC_{internal}$. The $RfC_{internal}$ is then converted to an RfC using the human PBPK model described in Appendix B [see Table 5-4 of the *Toxicological Review of Methanol (Noncancer)* (U.S. EPA, 2013)].

$$RfC = 858 \text{ mg-hr/L} \div 100 = 8.58 \text{ mg-hr/L} \Longrightarrow PBPK \Longrightarrow 2\times10^{1} \text{ mg/m}^3$$
(rounded to 1 significant figure)

I.B.3. Uncertainty Factors

UF = 100
= 10 (UF_H) × 3(UF_A) × 3(UF_D).

See Section I.A.3 for a complete description.

I.B.4. Additional Studies/Comments

See Section I.A.4.

I.B.5. Confidence in the Oral RfD

Study — Medium to High
Database — Medium
RfC — Medium to High

See Section I.A.5 for a complete description.

I.B.6. EPA Documentation and Review of the Inhalation RfC

Source Document – *Toxicological Review of Methanol (Noncancer)* (U.S. EPA, 2013)

This document has been provided for review to EPA scientists, interagency reviewers from other federal agencies and the Executive Office of the President, and the public, and peer reviewed by independent scientists external to EPA. A summary and EPA's disposition of the comments received from the independent external peer reviewers and from the public is included in Appendix A of the *Toxicological Review of Methanol (Noncancer)*(U.S. EPA, 2013).

Agency Completion Date - 09/30/2013

I.B.7. EPA Contacts

Please contact the IRIS Hotline for all questions concerning this assessment or IRIS, in general, at (202) 566-1676 (phone), (202) 566-1749 (fax), or hotline.iris@epa.gov (email address).

II. Carcinogenicity Assessment for Lifetime Exposure

Substance Name — Methanol
CASRN — 67-56-1

MESSAGE: A carcinogenicity assessment is not available at this time.

III. [reserved]
IV. [reserved]
V. [reserved]

VI. Bibliography

Substance Name — Methanol
CASRN — 67-56-1

VI.A. Oral RfD References

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VI.B. Inhalation RfC References

See Section VI.A.

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VI.C. Carcinogenicity Assessment References

None.

VII. Revision History

Substance Name — Methanol
CASRN — 67-56-1

Date	Section	Description

Date	Section	Description
09/07/1988	I.A.	Oral RfD summary on-line
09/30/2013	I., VI., VIII.	RfD assessment updated. RfC added.

VIII. Synonyms

Substance Name — Methanol
CASRN — 67-56-1
Last Revised — 09/30/2013

- 67-56-1
- Carbinol
- Methanol
- methyl alcohol
- Methyl hydroxide
- Monohydroxymethane
- Pyroxylic spirit
- wood alcohol
- Wood naptha
- wood-spirit