The EPA and the authors of this review of the non-cancer effects of methanol are to be commended for this latest version. The overall document is much more concise and direct in presenting the key features of the risk assessment that has been conducted. This version includes a number of edits that responded well to the comments of previous reviewers as well as the public. Several key items have contributed to this improvement include the utilization of background methanol levels in the PBPK model, the discussion of the relevance of the blood levels resulting from the RfC/RfD numbers in comparison to endogenous methanol levels, as well as a better explanation of various parameters in the PBPK models (such as using only the Sprague-Dawley rat data, not the F344 rat, and adding more human data to the validation).

Remaining issues of concern

1. There is insufficient rationale for the use of the UFd (data) of 3. The existing database on the developmental toxicity of methanol is sufficiently robust as to set the UFd at 1. In addition, the argument cited in the text for claiming a less than robust database (i.e. for justifying the UFd of 3) is not logical and can be disavowed. On the one hand, it is stated that the Burbacher and NEDO monkey studies are necessary to demonstrate the biological correlates (i.e., neurobehavioral toxicity) for the decreased brain weights in the NEDO rat study. In fact these monkey studies are cited to validate the use of the NEDO rat study as the principal study for deriving the RfC. However in discussing the database uncertainty, it is argued that the monkey studies are insufficient. While they may be insufficient as a principal study for deriving an RfC, they do corroborate the principal study and hence they offer robust data that obviates the need for UFd. As such, it isn't logical to consider these studies as being necessary and simultaneously insufficient. The UFd should be set at 1.

2. A positive feature of the revised toxicological review is that the authors have addressed the issue of whether the blood levels that would result from an RfC/RfD exposure would be distinguishable from the endogenous background. However I disagree with the assessment. There are two issues that appear relevant. First is that the data in Table 3-1 were almost all derived from special human populations, with some restrictions on dietary consumption, which would thus not reflect the endogenous levels in the general population. The endogenous levels in the general population are likely higher than the EPA-derived 1.5 $\pm$ 0.7 mg/L. (1 SD) (from the special "meta-analaysis" of studies in Table 3-1). The EPA should search for and include data from studies without dietary restrictions.

Second, the review states that the RfC/RfD exposures would contribute roughly 0.4 mg/L individually and 0.86 mg/L together additional blood methanol above the assumed methanol background level of 1.5 $\pm$ 0.7 mg/L. (1 SD). By definition, 1 SD implies that 2/3 of the human population should have endogenous levels between 0.8 and 2.2 mg/L. As an example, if persons with an endogenous levels of 0.8 mg/L were exposed and therefore obtained an added 0.86, their blood level would now be at 1.66 mg/L, which is barely (and not distinguishably) above the mean level! Figures 5.3 and 5.4 clearly show how the added levels are NOT distinguishable from background – this is reinforced by the slide #6 of the presentation by Dr. Gift, where the two distributions of blood methanol levels (background and background + RfC/RfD derived) are substantially overlapping. Interestingly in slide #11 of the presentation, when the the UFd happened to be set at 1, the two distributions of blood methanol are readily distinguishable. This slide shows clearly how an added blood methanol can be distinguishable from the endogenous level – and further reinforces the argument to set the UFd at 1, not 3.

3. The EPA used a BMD analysis of the key NEDO data and suggested that it would obviate the need to re-analyze the NEDO data using ANOVA. While the NEDO data on brain weights do appear

"visually" to be statistically significant and dose-related, the methanol-exposed groups have not been shown by a proper statistical analysis to be statistically different from controls. This still presents a problem that could be solved readily – why would the EPA utilize data from a study for RfC determination where the data has not been shown to be not statistically significant by the original authors using a valid statistical analysis? A re-analysis using ANOVA would be easily done and should be done, just to show validity of the NEDO data prior to using it for the BMD analysis.

Minor issues

An underlying theme of the toxicological review is that methanol is a development neurotoxicant and that the effects seen in the developmental studies result from methanol (or formaldehyde generated locally from methanol delivered to the target site). Based on all the available data, this seems to be a justifiable assumption. However, there are a number of studies cited in Appendix C from acute overdoses where subjects suffered various degrees of CNS damage as sequelae, including Parkinson-like syndrome. However, under the circumstances of acute overdoses, such CNS damage has been correlated with the presence of the severe acidosis in these patients – and the acidosis results from an initial accumulation of formate, accompanied later by an increase in lactate. A short statement that the CNS damage seen in the acute overdose exposures most likely results from the acidosis and not from methanol per se should be added to the beginning of Appendix C. In relation to this, on pages 4-2 and 4-3 of the main document, the authors indirectly state that formaldehyde and not formate is the likely cause of the ocular toxicity of methanol. This is clearly not true and needs to be revised, as shown by Martin-Amat et al (Methanol poisoning: Ocular toxicity produced by formate. Toxicol. Appl. Pharmacol. 45: 201-208, 1978; also, McMartin et al, Lack of a role for formaldehyde in methanol poisoning in the monkey. Biochem. Pharmacol. 28: 645-649, 1979).