Report of the *ITER* Review Meeting on Literature Risk Values for Manganese Oxide

June 29, 2011

***ITER* Review Organized by**
Toxicology Excellence for Risk Assessment
(http://www.tera.org/peer)

NOTE

This report was prepared by scientists of TERA and reviewed by the panel members. The members of the panel served as individuals on this panel, representing their own personal scientific opinions. They did not represent their companies, agencies, funding organizations, or other entities with which they are associated. Their opinions should not be construed to represent the opinions of their employers or those with whom they are affiliated.

Table of Contents

Subject Publications and Participants

Manganese Oxide (CASRN 1344-43-0)

The paper by Lisa A. Bailey, Julie E. Goodman, and Barbara D. Beck entitled, "Proposal for a revised Reference Concentration (RfC) for manganese based" describes the derivation of an inhalation reference dose (RfC) for manganese oxide. One of the authors (Barbara D. Beck) has been named as an expert in litigation involving air exposures to manganese, among other constituents. Some of the underlying work for this manuscript was conducted in the context of an assignment from an industrial client. Preparation of the manuscript was not supported by any client and the opinions are solely those of the authors.

The review panel members included Dr. Michael Dourson, Toxicology Excellence for Risk Assessment (TERA); Dr. Lisa Sweeney, SAIC; and Dr. Susan Felter, Proctor and Gamble.

Background

The purpose of the International Toxicity Estimates for Risk *(ITER)* database is to provide risk assessors and managers with the latest human health risk values from organizations around the world. *ITER* includes chronic human health risk data from the Agency for Toxic Substances and Disease Registry (ATSDR), Health Canada, International Agency for Research on Cancer (IARC) (in progress), National Institute of Public Health and the Environment (RIVM) - The Netherlands, U.S. Environmental Protection Agency (U.S. EPA), and independent parties whose risk values have undergone peer review. However, the peer reviewed literature contains many more risk values that may be of value to risk practitioners. Therefore, TERA developed a process to include these peer-reviewed, "literature-based," values on the *ITER* database. In order to be considered for inclusion on *ITER*, "literature-based" values must meet the following criteria:

- Manuscript that includes derivation of a risk assessment value has been published in a peer-reviewed journal;
- Assessment follows an identified, commonly used methodology (e.g., U.S. EPA, IPCS, Health Canada); and
- The manuscript's acknowledgment clearly states the source of funding for the work, or the authors provide this source of funding at the review meeting for full disclosure to the panel and on *ITER*.

Authors of peer reviewed publications that meet these criteria submit their publications for an additional quality evaluation by a panel of risk experts. TERA staff screens each publication to determine: (a) if each value was developed using a commonly accepted methodology, and (b) if the resulting risk value is consistent with the types of information *ITER* is designed to include (e.g., chronic human health risk values). The review panel then meets to discuss issues and address the charge questions. The values that the panel deems to be scientifically sound are then loaded on the *ITER* database to make these values more widely available.

Panel Selection and Conflict of Interest Evaluation

TERA has developed an extensive list of expert scientists interested in serving on our peer review panels. For each *ITER* Review meeting, TERA sends an invitation to all potential panelists asking for volunteers willing to participate in the review meeting process on a pro-bono basis. TERA screens the panel volunteers to ensure that the resulting panel includes the necessary expertise to evaluate the risk values under review and to ensure there are no conflict of interest issues. In the instance that there are more volunteers than needed, TERA adjusts the panel membership and insures a proper balance of expertise. When a TERA value is being reviewed, an outside independent party reviews the panel membership and conflict of interest.

An essential part of an independent expert review is the identification of conflicts of interest and biases that would disqualify a candidate, as well as identification and disclosure of situations which may appear to be a conflict or bias. The purpose for evaluating conflict of interest is to ensure that the public and others can have confidence that the peer reviewers do not have financial or other interests that would interfere with their ability to carry out their duties objectively. TERA follows the U.S. National Academy of Sciences (NAS) guidance on selection of panel members to create panels that have a balance of scientific viewpoints on the issues to be discussed. As a result, the expert panels have a broad and diverse range of knowledge, experience, and perspective, including diversity of scientific expertise and affiliation. Panel members serve as *individuals*, representing their own personal scientific opinions. They do not serve as representatives of their companies, agencies, funding organizations, or other entities with which they are associated. Their opinions should not be construed to represent the opinions of their employers or those with whom they are affiliated.

For the June 29, 2011 meeting, three experts volunteered to serve on the panel:

- Dr. Michael Dourson, Toxicology Excellence for Risk Assessment (TERA);
- Dr. Lisa Sweeney, SAIC;
- Dr. Susan Felter, Proctor and Gamble.

TERA asked each candidate to report on his or her financial and other relationships with the authors and sponsors of the risk value by completing a questionnaire. The completed questionnaires were reviewed by TERA staff and discussed further with panel candidates as needed. (See www.tera.org/peer/COI.html for TERA conflict of interest and bias policy and procedures for panelist selection.) TERA determined that the selected panel members have no conflicts of interest and are able to objectively participate in this review. None of the panel members has a financial or other interest that would interfere with his or her abilities to objectively participate on the panel. None of the panel members is employed by the organizations that authored or sponsored the risk values. None of the panel members was involved in the preparation of the risk values.

Meeting Procedure

For the *ITER* Review meetings, the authors provide TERA with a documentation package, including supporting data and analyses, to support their risk value. TERA staff screens each package to ensure completeness. TERA has prepared a standard list of charge questions, which outlines the issues and

questions to guide these reviews (see Appendix A). TERA forwards these charge questions to the panel, and panel members have the opportunity to add to the charge if additional questions are needed. TERA distributes the review materials and charge to panel members prior to the meeting. Panel members are given the opportunity to request additional literature as needed and to submit written pre-meeting comments as necessary.

For the June 29, 2011 meeting, TERA distributed the review materials and charge to panel members on June 6, 2011.

At the meeting, the author briefly presents the assessment, and then the panel members are given the opportunity to ask clarifying questions. The panel then conducts a thorough, systematic discussion of the key data and decisions using the charge questions. The panel members are asked to indicate whether or not each risk value should be included on *ITER*. Panel members are also asked to note any substantive points or issues to include in the *ITER* file that they think would be helpful for the *ITER* user to be aware of when considering these values.

Panel comments and conclusions for the June 29, 2011 meeting are described for the manuscript in Appendix B.

Meeting Report

After the meeting, the panel (assisted by a TERA scientist) compiles its recommendations and summarizes them for inclusion on *ITER*. Appendix B provides the summaries of the panel's review and its comment on each of the subject publications that were reviewed on June 29, 2011. This meeting report serves as a record of the review; it has been reviewed by the panel members for accuracy before it is finalized. These comments are also available in the chemical entry in *ITER* in the quantitative estimate section.

Appendix A

Charge Questions for *ITER* Reviews

1) METHODOLOGY

- Was an appropriate risk assessment methodology used and applied correctly? Was the methodology applied correctly, and are the conclusions solid based on the work done? Other comments?

2) ASSESSMENT QUALITY

- Was a literature search done and fully explained/evaluated? Do the authors discuss alternative modes of action, viewpoints, or existing assessments? Other comments?

3) CONCLUSIONS

- Are the publication's conclusions scientifically sound and supported by the data? Do the authors fully explain and support the choice of critical effect, point of departure, and dose-response? Other comments?

4) VALUE

- Is this publication of sufficient value to include on *ITER*? Who are the intended users of the derived value, and how do they benefit from this information on *ITER*? Other comments?

5) OTHER

- Are there additional issues or comments relevant to the publication's risk value and its conclusions?

Appendix B

Manganese Oxides

CAS NO: 1344-43-0

ITER PR-June 2011

Source Document

Bailey L.A, Goodman J.E., and Beck B.D. (2009). Proposal for a revised Reference Concentration (RfC) for manganese based on recent epidemiological studies. *Regul. Toxicol. Pharmacol.* **55:** 330-339. Available at: http://www.ncbi.nlm.nih.gov/pubmed/19686793.

Data Summary

Organization	ITER PR
Chemical Name	Respirable Manganese Oxides
Risk Value Name	RfC
Risk Value	2-7 µg/m^3 (respirable Mn)
Year	2009
Basis (EXP)	*NOAEL 60 µg/m^3 (measured as respirable Mn) **BMDL$_{10}$ 200 µg/m^3
Basis (ADJ)	NOAEL$_{[HEC]}$ 21 µg/m^3 BMDL$_{[HEC]}$ 71 µg/m^3
Uncertainty Factor (UF)	UF$_A$ – 1 UF$_H$ – 10 UF$_S$ – 1 UF$_L$ – 1 UF$_D$ – 1*** Total UF – 10
Critical Organ or Effect	Subclinical neurobehavioral effects
Species	Human
Study	*Gibbs *et al*. (1999), Deschamps *et al*. (2001), Young *et al*. (2005); Clewell *et al*. (2003)
Methodology	US EPA methodology (US EPA, 1994, 2002)

Notes:

RfC = reference concentration.

NOAEL = no observed adverse effect level.

**Gibbs et al. (1999) (NOAEL = 66 µg/m^3), Deschamps et al. (2001) (NOAEL = 57 µg/m^3), and Young et al. (2005) (NOAEL = 58 µg/m^3).*

***Clewell et al. (2003).*

****Adjustment of the RfC to account for more soluble, more bioavailable, and potentially more toxic forms of Mn, such as Mn sulfates, should be considered on an exposure-specific basis. See text for more discussion.*

Determination of critical effect:

Manganese (Mn) is an essential nutrient and known neurotoxin following excessive exposures. Bailey et al. (2009) conducted a literature search in PubMed using the following search terms: "manganese AND (neuro* OR neurotox* OR neurology OR neurologic*)." They limited the human studies to those that:

- Were published after 1992;
- Examined and reported Mn dust, and not welding fumes.[1] as the exposure of concern from personal air monitoring data;
- Evaluated both an exposed and an unexposed population; and
- Evaluated neurological effects in relation to ongoing exposures to Mn in air.

Although human studies that measured total Mn in air were located, Bailey et al. (2009) focused on studies that measured the respirable fraction of Mn for the RfC calculation. Respirable particles are capable of penetrating the lung tissue, while larger particles are trapped in the nasal and pharyngeal passages, and do not penetrate the lung tissue where they could enter the circulation (Klaassen, 2001). Although some absorption may occur from mucociliary clearance and ingestion from deposition in the upper respiratory tract, the percent uptake from the gastrointestinal tract is very low (1-5%) (Dorman *et al*. 2006) and would contribute minimally compared to absorption *via* the inhalation route. Therefore, the majority of direct absorption into the circulation is from inhalation of small respirable particles into deep lung tissue.

In addition, to supplement the PubMed results, Bailey et al. (2009) conducted a search of the Developmental and Reproductive Toxicology (DART) database using the following search terms: "manganese OR colloidal manganese." Studies were limited to developmental studies conducted after 1992.

Sub-clinical neurological effects

Bailey et al. (2009) found eight cohorts (12 studies total) that met our criteria for subclinical neurological effects. All are occupational studies with chronic exposure durations (*i.e.*, greater than seven years, based on 10% of a 70-year lifetime exposure). Observed effects, to the extent there were any, were subclinical neurobehavioral effects, predominantly on motor function (typically visual reaction time, hand-eye coordination, and hand steadiness). Of the eight cohorts, three studies of respirable Mn were identified from which no observed adverse effect levels (NOAELs) were derived (Gibbs *et al*., 1999; Deschamps *et al*., 2001; Young *et al*., 2005). The rationale for choosing these studies is discussed below.

Gibbs *et al*. (1999) conducted a study of 75 Mn-exposed workers at an alkaline battery plant in northern Mississippi and 75 nearby plant workers with no known history of occupational

[1] Studies of welding fume exposures were excluded because, from a toxicological perspective, welding fumes are very different from particulate Mn. Welding fumes are complex mixtures, including other metals and carbon monoxide.

exposure to Mn (73 were employed at a pigment-grade titanium dioxide plant and two were employed at the alkaline battery plant working in sodium chlorate production). The mean Mn air concentration in respirable dust in exposed workers was measured by personal air monitors to be 66 $\mu g/m^3$ (ranging from 5-230 $\mu g/m^3$). The mean exposure duration was 12.7 (SD ± 10.11) years. Subjects were matched on sex, race, age, and salary and were administered multiple neuropsychological tests, including hand-eye coordination, hand steadiness, complex reaction time, and rapidity of motion. No significant effects of Mn exposure were found on any neurobehavioral test, resulting in a NOAEL of 66 $\mu g/m^3$.

Deschamps *et al*. (2001) conducted neurobehavioral examinations in 138 enamels-production workers exposed to Mn for an average of 19.9 (SD ± 9) years and 137 matched technicians from public service employers or local municipal operations laborers. Subjects were matched on age, education, and ethnic group. Based on personal monitor measurements, the mean respirable exposure concentration was 57 $\mu g/m^3$ (ranging from 10-293 $\mu g/m^3$). No differences were found between mean concentrations of Mn in blood of exposed and unexposed workers. Tests conducted included sensory and motor exam of cranial nerves; fine-touch, motor, and sensory exam of power of all main muscle groups; reflex tests; cerebellar abnormalities; and tests of domains of speech regulation and initiation, attention, concentration and memory, cognitive flexibility, and affect; and a questionnaire for neuropsychological status. In addition, the visual gestalt test score was higher in workers exposed to Mn for 11-15 years, but the authors attribute this to the higher technical skills of this group of six workers. This is supported by a lack of dose-response relationship, as no statistically significant effects were noted in the four people exposed 16-19 years or the 69 people exposed for 20+ years. There was a higher prevalence of self-reported asthenia (lack of energy and strength), sleep disturbance, and headache in exposed workers. It is unclear whether these non-specific symptoms were exposure related because they are self-reported and less reliable, and the other neurobehavioral endpoints that are more reliable show no relationship with Mn exposure. Based on these results, the authors concluded that "long exposure to low levels of Mn…showed no significant disturbance of neurological performance." Results of this study indicated a NOAEL of 57 $\mu g/m^3$.

Young *et al*. (2005) conducted a study of 509 South African Mn smelter workers exposed for 18.2 (SD ± 7.6) years and 67 unexposed electrical assembly plant workers. Respirable Mn exposures ranged from 3-510 $\mu g/m^3$, with a median of 58 $\mu g/m^3$. Exposure indices for individuals were attributed or interpolated from 98 personal samplers. The study authors assessed several neurobehavioral endpoints, including items from the Swedish nervous system questionnaire (Q16), the World Health Organization neurobehavioral core test battery (WHO NCTB), the Swedish performance evaluation system (SPES), the Luria-Nebraska (LN), the Danish Product Development (DPD) test batteries, and a brief clinical examination. The study found "few respirable Mn effects showing a clear continuity of response with increasing exposure." They observed dose-response associations primarily with exposures less than 100 $\mu g/m^3$, above which the relationship was flat. The authors concluded that the study was essentially negative and that "the small number of convincing effects, especially motor function effects, and the character of the exposure-response relationships where effects were observed in this study suggests that these are due to chance." Although these data are less reliable than those reported in the Gibbs *et al*. (1999) and Deschamps *et al*. (2001) studies, a NOAEL of 58 $\mu g/m^3$ is assumed based on the likelihood of positive findings being due to chance.

Five studies evaluated total rather than respirable Mn (Chia *et al*., 1993; Lucchini *et al*., 1995, 1999; Crump and Rousseau, 1999; Myers *et al*., 2003), and thus were not considered further for calculating a Mn RfC. Lowest observed adverse effect levels (LOAELs) in these studies ranged from 96-1,590 $\mu g/m^3$. Two additional occupational cohorts (four studies) evaluated subclinical nervous system effects from exposure to respirable Mn: 1) Mergler *et al*. (1994); Bouchard *et al*. (2007a,b); and 2) Bast-Pettersen *et al*. (2004). As discussed below, these studies were not considered robust enough for derivation of a NOAEL.

Mergler *et al*. (1994) evaluated neurological effects of 74 Mn alloy workers and 74 matched controls exposed for an average of 16.7 years to a wide range of respirable Mn air concentrations (ranging from 1-1,273 $\mu g/m^3$), with an arithmetic mean of 122 $\mu g/m^3$. The authors found that the exposed workers performed more poorly on tests of motor function. However, because of the wide range of exposure concentrations in this group, we concluded that this study would not provide a reliable basis, as compared to the selected studies, for development of an RfC. Follow-up studies (14 years after exposure) by Bouchard *et al*. (2007a,b) did divide the exposed group into a high, medium, and low tertile, based on total Mn, and found no observed adverse effects in the lowest tertile group (average total Mn of 390 $\mu g/m^3$).

Bast-Pettersen *et al*. (2004) conducted a large number of neuropsychological tests on 100 Mn alloy plant workers and 100 silicon and microsilica plant and titanium dioxide slag and pig iron plant workers, including tests for cognitive functions; motor tests; tests of motor speed, grip strength, coordination, and reaction time; and a questionnaire to evaluate self-reported neuropsychiatric symptoms. Average exposures were 64 $\mu g/m^3$ (range: 3-356 $\mu g/m^3$). Of the tests, three of eight motor tests (tremor tests) showed significant effects in the exposed *vs.* the control group. For the following reasons, we conclude that weight of epidemiological evidence does not sufficiently support applying the 64 $\mu g/m^3$ from this study as a LOAEL point of departure for deriving an RfC:

- There is moderate inconsistency among the neurological test results in this study compared to other studies (such as Roels *et al*., 1992) where results were more consistent across tests;
- There are three epidemiological studies suggesting a NOAEL at 60 $\mu g/m^3$ (Gibbs *et al*., 1999, Deschamps *et al*. 2001, Young *et al*., 2005);
- We did not have the raw data to calculate a BMDL, which would be more robust than applying a LOAEL as the point of departure;
- Clewell et al. (2003) derived a more reliable $BMDL_{10}$ (200 $\mu g/m^3$) from the Roels *et al*. (1992) and Gibbs *et al*. (1999) studies that is higher than the LOAEL from this study.

Developmental effects

From the developmental studies Bailey et al. (2009) reviewed (Dorman *et al*., 2005a,b; Erikson *et al*., 2005; HaMai *et al*., 2006; Rindernecht *et al*., 2005), the lowest concentration where a developmental effect (decreased liver weight in rat offspring) was observed was 500 $\mu g/m^3$ (Dorman *et al*., 2005a). If the 500 $\mu g/m^3$ LOAEL is divided by an uncertainty factor of 10-fold,

it translates to an estimated NOAEL of 50 μg/m^3 in rats and a NOAEL$_{[HEC]}$[2] of 32 μg/m^3, which is slightly higher than the neurological NOAEL$_{[HEC]}$ of 20 μg/m^3 (see below). Therefore, subclinical neurological effects are more sensitive than developmental endpoints.

In addition, pharmacokinetic studies have addressed potential neurodevelopmental effects. That is, Dorman and coworkers (Dorman *et al*., 2006; Yoon *et al*., 2009a,b) observed that Mn exposure concentrations of 0.05, 0.5, and 1 mg/m^3 for 6 hr/day for 7 days/week, for rat neonates (through PND 19) and dams (from 28 days prebreeding through PND 18), resulted in fetal and neonatal Mn brain concentrations (in rats) that were not very different from adult brain concentrations. Recently, as discussed below, a non-human primate PBPK model for Mn (Nong *et al*., 2009) has been scaled to humans (Schroeter *et al*., 2011), and has been used to model fetal and neonatal Mn exposure in humans during gestation and lactation (Yoon *et al*., 2011). Yoon *et al*. (2011) predict similar Mn tissue concentrations (from Mn inhalation exposure concentrations ranging from 1-10 μg/m^3) in the target brain region in the human fetus, nursing infant, and children compared to those in the mother and other adults.

Therefore, there is sufficient evidence to suggest that developmental and potentially neurodevelopmental effects from inhalation of Mn are not more sensitive than neurological effects in adults at typical human exposure concentrations.

Quantitative Estimates

Bailey et al. (2009) derived two Mn RfCs, following standard US EPA methodology (US EPA, 1994, 2002): one based on a NOAEL, and one based on the 95% lower confidence limit on a benchmark dose associated with 10% extra risk (BMDL$_{10}$) derived by Clewell *et al*. (2003).

The NOAEL is derived from the three key studies discussed above: Gibbs *et al*. (1999) (NOAEL = arithmetic mean of 66 μg/m^3), Deschamps *et al*. (2001) (NOAEL = arithmetic mean of 57 μg/m^3), and Young *et al*. (2005) (NOAEL = median of 58 μg/m^3). Because these NOAELs are all very close to 60 μg/m^3, Bailey et al. (2009) chose 60 μg/m^3 as the point of departure. Bailey et al. (2009) used arithmetic means of the concentrations for derivation of our NOAELs based on studies suggesting that the arithmetic mean provides a better summarization of group exposure with regard to a dose-response relationship, and is therefore more appropriate for use in risk assessment (Clewell *et al*., 2003; Crump, 1998). A median value was chosen from the Young *et al*. (2005) study since an arithmetic mean was not available.

The BMDL point of departure is from Clewell *et al*. (2003). The authors of this study calculated BMDLs based on data from Gibbs *et al*. (1999) and Roels *et al*. (1992). Clewell *et al*. (2003) derived BMDs on the order of 300 μg/m^3, and BMDL$_{10}$ values ranging from 90-270 μg/m^3, with a mean of approximately 200 μg/m^3 from each study. The 200 μg/m^3 BMDL$_{10}$ was applied as a second point of departure.

[2] The NOAEL$_{[HEC]}$ was calculated using the following assumptions: the geometric mean diameter was 1.03 μm, with a geometric standard deviation of 1.52. Sprague-Dawley rats were assumed to weigh 204 mg. The experiment exposures were adjusted to reflect 24-hour, 7 days/week exposure (US EPA, 1994).

The Mn RfC was calculated by first converting the NOAEL and $BMDL_{10}$ points of departure to a $NOAEL_{[HEC]}$ and $BMDL_{[HEC]}$ by converting the human occupational exposure to a continuous exposure for the general population (US EPA, 2009). That is:

$$NOAEL_{[HEC]} = NOAEL \times 5/7 \text{ days} \times 10/20 \text{ m}^3/\text{day}$$
$$21 \text{ μg/m}^3 = 60 \text{ μg/m}^3 \times 5/7 \text{ days} \times 10/20 \text{ m}^3/\text{day}$$

$$BMDL_{[HEC]} = BMDL_{10} \times 5/7 \text{ days} \times 10/20 \text{ m}^3/\text{day}$$
$$71 \text{ μg/m}^3 = 200 \text{ μg/m}^3 \times 5/7 \text{ days} \times 10/20 \text{ m}^3/\text{day}$$

The RfC is then calculated in the following manner, applying an uncertainty factor (UF) of 10 for intraspecies variability (see discussion below):

$$RfC = NOAEL_{[HEC]} \text{ or } BMDL_{[HEC]} / UF$$

RfC = 2 μg/m^3 = 21 μg/m^3 / 10 (intraspecies UF)
RfC = 7 μg/m^3 = 71 μg/m^3 / 10 (intraspecies UF)

UFs for use of a LOAEL or a subchronic study

Use of a NOAEL and BMDL as a point of departure eliminated the need for a LOAEL UF. In addition, the studies that are the basis of the points of departure are chronic (except for Roels *et al*., 1992, used to derive one BMDL), eliminating the need for a subchronic UF. Clewell *et al*. (2003) found that the duration of occupational exposure in the Roels *et al*. (1992) data was not significantly correlated to any measure of psychomotor response. In addition, as noted by Clewell *et al*., although the average duration of Mn exposure was three times greater in the Gibbs *et al*. (1999) study than in the Roels *et al*. (1992) study (14.1 years *vs*. 5.7 years), the BMDLs based on current exposure concentration calculated from these studies were comparable. Thus, Clewell *et al*. (2003) concluded that because the effects measured in these studies do not appear to depend on exposure duration, an adjustment downward for potentially longer exposure durations is not necessary for the BMDL based on the Roels *et al*. (1992) study.

UF for developmental effects

As discussed above, the neurological endpoint is more sensitive than the developmental endpoint, and there are minimal pharmacokinetic differences with regard to Mn brain concentrations between fetal, neonatal, and adult rats following exposures to 0.05, 0.5, and 1 mg/m^3 Mn for 6 h/day 7 days/week (rat neonates through PND 19, and dams from 28 days prebreeding through PND 18). Further, a human PBPK model for Mn exposure (Schroeter *et al*., 2011; Yoon *et al*., 2011) suggests that there are no differences in human neonatal, fetal, and adult brain concentrations following exposures < 10 μg/m^3. Therefore, there is sufficient evidence to confirm that neurotoxicity in adults is the critical effect and that similar toxicities would be expected in younger individuals since relevant concentrations are similar. This eliminates the need for an additional uncertainty factor for database deficiencies, and specifically for developmental toxicity.

UF for Mn species differences in toxicity

The more soluble forms of Mn lead to higher tissue concentrations (Dorman *et al*., 2006b), and are generally considered more toxic, with solubility of Mn species generally following soluble

sulfates > less soluble phosphates > less soluble oxides. Since the exposures in the epidemiological studies used to derive our proposed RfCs were to the least soluble Mn oxides [alkaline battery production (Roels *et al*., 1992); electrolytic Mn (Gibbs *et al*., 1999); ferromanganese smelting (Young *et al*., 2005)][3], our proposed RfCs should apply only to exposures to Mn oxides.

Adjustment of the RfC to account for potentially more soluble, more bioavailable, and potentially more toxic forms of Mn, such as Mn sulfates, should be considered on an exposure-specific basis. Such consideration might entail the use of an uncertainty factor for database deficiencies. It is noteworthy that the PBPK models described below are based on studies where the more soluble, and potentially more toxic, Mn sulfates were applied.

Intraspecies UF

Mn accumulation in the human striatum and globus pallidus is associated with neurotoxicity following inhalation exposure to high concentrations of Mn. There have been recent advances in the understanding of the pharmacokinetics of inhaled Mn in potentially sensitive individuals. This has been extensively studied (Dorman *et al*., 2004, 2005a,b) and reviewed by Dorman *et al*. (2006), where the authors compared (or reviewed studies of comparisons of) Mn brain concentrations of healthy young adult male rats to rats that were considered to reflect potentially susceptible subpopulations (aged; abnormal hepatobiliary function; sub-optimal iron or Mn intake; and fetuses, neonates, and children). The authors concluded that inhaled Mn particles result in "qualitatively similar end-of-exposure brain Mn concentrations" in the potentially susceptible subpopulations as compared to healthy young adult male rats.

More recently, physiologically-based pharmacokinetic (PBPK) models for inhaled Mn have been developed which provide a thorough quantitative analysis of Mn tissue concentrations in rats (Teeguarden *et al*., 2007a,b,c; Nong *et al*., 2008), including placental transfer to fetuses (Yoon *et al*., 2009a), lactational transfer to pups (Yoon *et al*., 2009b), and in non-human primates (Nong *et al*., 2009). Andersen *et al*. (2010) summarized these PBPK models, describing how the models consider ingestion and inhalation kinetics of Mn along with homeostatic control of Mn. Schroeter *et al*. (2011) describe scaling of the non-human primate PBPK model to a human PBPK model. The authors conclude that the human PBPK model indicates that globus pallidus Mn concentrations in the human brain would be unaffected by Mn air concentration < 10 $\mu g/m^3$. Schroeter *et al*. (2011) observed that Mn brain concentrations begin to increase when exposure concentrations approach 100 $\mu g/m^3$.

In addition, Yoon *et al*. (2011) describe application of the human PBPK model to predict human fetal and neonatal tissue concentrations. Yoon *et al*. (2011) predict similar Mn tissue concentrations (from Mn inhalation exposure concentrations ranging from 1-10 $\mu g/m^3$) in the target brain region in the human fetus, nursing infant, and children compared to those in the mother and other adults. The human PBPK studies suggest that concentrations below 10 $\mu g/m^3$ Mn in air would not lead to accumulation of Mn in the brains of human fetuses, children, and adults. An accumulation threshold for Mn is biologically plausible because Mn is an essential

[3] The form of Mn in the Deschamps *et al*. (2001) study is unclear.

nutrient, and homeostatic control mechanisms limit accumulation of essential nutrients at doses less than an accumulation threshold[4] (Santamaria, 2008).

These PBPK models also suggest that a database UF of 10 is not necessary for developmental toxicity in fetuses, neonates, or children. A UF for other potential toxicities, such as abnormal hepatobiliary function or sub-optimal iron or Mn intake) was considered. Dorman *et al*. (2006) described rat studies that suggest that old age or sub-optimal Mn intake did not affect Mn brain concentrations, suggesting that our currently proposed toxicokinetic UF = 3, or perhaps even 1, is appropriate for these subpopulations. Dorman *et al*. (2006) also described studies that suggested a three-fold increase in Mn brain concentrations in liver-diseased rats (suggesting that our currently proposed toxicokinetic UF = 3 is appropriate) and a two-fold increase in Mn brain concentrations in rats with suboptimal iron intake (suggesting perhaps a toxicokinetic UF = 2). These UFs are consistent with, or slightly higher than, chemical-specific adjustment factors (CSAF) proposed recently by Taylor *et al*. (2010) using PBPK modeling for susceptible human subpopulations. Therefore, our currently proposed toxicokinetic UF of three, as discussed in Dorman *et al*. (2006), is likely sufficient for individuals with sub-optimal iron intake or abnormal hepatobiliary function. We also apply a UF of three for uncertainties in toxicodynamics for these sensitive subpopulations, resulting in a total UF of 10 for intraspecies variability.

Applyng a UF of 10 for intraspecies variability to our BMDL and NOAEL points of departure results in a range of potential Mn RfCs of 2-7 $\mu g/m^3$. These RfCs fall below the proposed Mn accumulation threshold of 10 $\mu g/m^3$.

Overall assessment:

Based on the reading and analysis of the information provided, the panel identified their overall recommendation for the proposed *ITER* materials they reviewed as:

☑ Acceptable with comments (as indicated)

[4] Although there is some direct olfactory transport to the brain, which is not under homeostatic regulation, the amount is very small (Andersen *et al*., 2010; Leavens *et al*., 2007) and is accounted for in the PBPK model. The amount is small enough at 10 $\mu g/m^3$ inhalation that the resulting increased Mn brain concentration from the olfactory route is insignificant compared to normal Mn levels in the brain.

Appendix C

Panel Conclusions and Recommendations

On June 29, 2011, the panel determined that the risk value derived in "Proposal for a revised Reference Concentration (RfC) for manganese based on recent epidemiological studies" by Bailey et al. (2009) should be included on the *ITER* database.

Generally, the panel concluded that the value was derived using an appropriate risk assessment methodology that had been applied correctly. Although it is somewhat unconventional to mean BMDLs for different neurobehavioral responses from different studies as the point of departure (POD) to derive an RfC, after discussion the panel was less concerned with this issue because the critical effect was judged to be subclinical and the epidemiology studies did not have well defined exposures. The panel agreed that since these exposures were variable using mean BMDLs reflected this uncertainty. Therefore, the panel felt comfortable using the mean as a POD.

The panel also discussed the issues of homeostatic control for oral exposure and the anticipated lack of homeostatic control for inhalation exposure. In particular, the panel expressed some concern over direct exposure via the olfactory canal vs. systemic blood exposure from inhalation exposure. After discussion regarding the PBPK model, one panel member with PBPK expertise explained that the model includes oral and inhalation parameters, and in particular, accounts for homeostasis and whole body burden. The model shows that once manganese gets into the circulation, there is no difference in effect between oral and inhalation exposure. This panel member noted that manganese does get transported to the brain directly via the olfactory canal, but this pathway was judged to be minimal as compared to the major pathway associated with blood – which is the basis for the PBPK model. Upon review of the sensitivity analysis for the PBPK model (in Schroeter et al., 2011), the PBPK expert panel member confirmed that the PBPK model accounted for whole body burdens and the potential for olfactory transfer. The sensitivity analysis also supported a threshold value of 10 µg/m^3 (see for example, Figure 14 of Schroeter et al., 2011). And, since this RfC is for manganese oxide only, and the PBPK model is based on a more soluble and toxic form, an RfC below 10 µg/m^3 could be considered to be as protective as one based on soluble forms of the metal, although the panel felt that more work should be done to confirm this.

Choice of Uncertainty Factors and Discussion

Neonate susceptibility and interhuman variability:

The panel judged that the default uncertainty factor for this area was appropriate. This was because while the PBPK model implies a possible reduction in the toxicokinetic portion of within human variability (i.e. the data suggest a value less than the logarithmic midpoint of ~3), other susceptible subpopulations, including those with liver problems, suboptimal iron or higher level of manganese intake might indicate a toxicodynamic factor greater than ~3-fold. The panel

suggested rewriting the explanation paragraph to explicitly state why the default uncertainty factor was used, and for the authors to be more definitive in their discussion of data-derived alternatives.

The panel also discussed whether the apparent PBPK 10 µg/m^3 threshold in adults was protective for neonates. Neonates are known to exert less homeostatic control over manganese blood levels, as well as having reduced blood-brain barrier function when compared with adults. In contrast, there was some discussion as to whether neonates need higher levels of manganese for brain development, but there were no data brought forth to support that neonates in fact do require higher levels of Mn. There was significant debate here, as it was mentioned that most risk assessments for Mn call out neonates as a potentially high risk populations and the fact that human milk is naturally very low in Mn. These facts suggest that neonates do not, in fact, need higher levels of Mn. The panel noted the existence of data showing that oral exposure to manganese in neonatal rats (25 mg/kg), that resulted in higher Mn brain concentrations than those from inhalation exposure to 10 µg/m^3 Mn, resulted in minimal neurologic effects (Dorman et al. 2000). It was also brought forth that the issue of neonate susceptibility would be associated more with the derivation of an acute value since one is not chronically an infant. Thus, there was no further justification for an additional child factor, other than the default of 10 for within human variability.

Subchronic to chronic:

The panel judged that the 1-fold uncertainty factor for this area was appropriate. One panel member mentioned that at low levels of atmospheric Mn in terms of accumulation, one will accumulate all that they are able after a few months. Therefore, any variation in body burden will be the result of dietary or other non-inhalation exposures. This panel member noted that these conclusions regarding variation in body burden would not be applicable at high levels of occupational exposure. The panel noted that no additional accumulation occurs due to increasing age. Furthermore, the toxicokinetics and PBPK models support the judgment of no difference between subchronic and chronic exposure at low levels of Mn.

A panel member mentioned that the longest exposure from the epidemiological studies is 15-19 years. Yet Clewell et al. (2003) derived two comparable BMDLs using two very different exposure durations. The basis for one of the BMDLs was Gibbs et al. (1999) that had a high standard deviation (+/- 10 years), meaning some of the exposed population may have had lower exposures near 2 years. This could bias the BMD to be higher, as these people may not show effects for some time. However, since the basis for exposure duration is an average, there are people on the other side of the average who may have had up to 30 years exposure.

Overall, the panel felt confident that an additional UF for exposure duration was not necessary – the discussion in Clewell et al. (2003) regarding lifetime exposure vs. more recent exposure (within 5 yrs) concludes that “duration of occupational exposure was not a significant correlate

of any measure of psychomotor response in Roels et al. (1987)". And, according to the PBPK expert panel member, the PBPK model supports no difference between subchronic and chronic exposure.

Database Uncertainty factor

The panel judged that the 1-fold uncertainty factor for this area was appropriate because all panel members agreed that neurotoxicity is the most sensitive endpoint, and therefore the correct choice of critical effect. The issue of whether reproductive toxicity might be the critical effect was discussed by the panel. Both PBPK modeling and a limited database that was reviewed were helpful to the panel in deciding that it was not the critical effect. For example, NOELs and LOELs for developmental effects were above those for subclinical neurobehavioral effects by a substantial amount.

This proposed range of values is a fairly different from values already loaded on *ITER*, but it uses the most recent epidemiology studies and PBPK models. The proposed range is likely to be valuable to the risk assessment community as well.

Bibliography

Andersen, ME; Dorman, DC; Clewell, HJ III; Taylor, MD; Nong, A. 2010. Multi-dose-route, multi-species pharmacokinetic models for manganese and their use in risk assessment. J. Toxicol. Environ. Health A 73(2), 217-234.

Bast-Pettersen, R., Ellingsen, D.G., Hetland, S.M., Thomassen, Y., 2004. Neuropsychological function in manganese alloy plant workers. Int. Arch. Occup. Environ. Health 77(4), 277-287.

Bouchard, M., Mergler, D., Baldwin, M., Panisset, M., Roels, H.A., 2007a. Neuropsychiatric symptoms and past manganese exposure in a ferro-alloy plant. Neurotoxicology 28(2), 290–297.

Bouchard, M., Mergler, D., Baldwin, M., Panisset, M., Bowler, R., Roels, H.A., 2007b. Neurobehavioral functioning after cessation of manganese exposure: A follow-up after 14 years. Am. J. Ind. Med. 50(11), 831–840.

Chia, S.E., Foo, S.C., Gan, S.L., Jeyaratnam, J., Tian, C.S., 1993. Neurobehavioral functions among workers exposed to manganese ore. Scand. J. Work Environ. Health 19(4), 264-270.

Clewell, H.J., Lawrence, G.A., Calne, D.B., Crump, K.S., 2003. Determination of an occupational exposure guideline for manganese using the benchmark method. Risk Anal. 23(5), 1031-1046.

Crump, K.S., 1998. On summarizing group exposures in risk assessment: Is an arithmetic mean or a geometric mean more appropriate? Risk Anal. 18(3), 293-297.

Crump, K.S., Rousseau, P., 1999. Results from eleven years of neurological health surveillance at a manganese oxide and salt producing plant. Neurotoxicol. 20(2-3), 273-286.

Deschamps, F.J., Guillaumot, M., Raux, S., 2001. Neurological effects in workers exposed to manganese. J. Occup. Environ. Med. 43(2), 127-132.

Dorman, D.C., McManus, B.E., Marshall, M.W., James, R.A., Struve, M.F., 2004. Old age and gender influence the pharmacokinetics of inhaled manganese sulfate and manganese phosphate in rats. Toxicol. Appl. Pharmacol. 197 (2), 113–124.

Dorman, D.C., McElveen, A.M., Marshall, M.W., Parkinson, C.U., James, R.A., Struve, M.F., Wong, B.A., 2005a. Tissue manganese concentrations in lactating rats and their offspring following combined in utero and lactation exposure to inhaled manganese sulfate. Toxicol. Sci. 84(1), 12-21.

Dorman, D.C., McElveen, A.M., Marshall, M.W., Parkinson, C.U., James, R.A., Struve, M.F., Wong, B.A., 2005b. Maternal-fetal distribution of manganese in the rat following inhalation exposure to manganese sulfate. Neurotoxicol. 26(4), 625-632.

Dorman, D.C., Struve, M.F., Clewell, H.J., Andersen, M.E., 2006b. Application of pharmacokinetic data to the risk assessment of inhaled manganese. Neurotoxicol. 27(5), 752-764.

Erikson, K.M., Dorman, D.C., Lash, L.H., Aschner, M., 2005. Persistent alterations in biomarkers of oxidative stress resulting from combined in utero and neonatal manganese inhalation. Biol. Trace Elem. Res. 104(2), 151-163.

Gibbs, J.P., Crump, K.S., Houck, D.P., Warren, P.A., Mosley, W.S., 1999. Focused medical surveillance: A search for subclinical movement disorders in a cohort of U.S. workers exposed to low levels of manganese dust. Neurotoxicology 20(2-3), 299-313.

HaMai, D., Rinderknecht, A.L., Guo-Sharman, K., Kleinman, M.T., Bondy, S.C., 2006. Decreased expression of inflammation-related genes following inhalation exposure to manganese. Neurotoxicol. 27(3), 395-401.

Klaassen, CD (Ed.) 2001. Casarett and Doull's Toxicology: The Basic Science of Poisons (Sixth Edition). 1280p.

Leavens, TL; Rao, D; Andersen, ME; Dorman, DC. 2007. Evaluating transport of manganese from olfactory mucosa to striatum by pharmacokinetic modeling. Toxicol. Sci. 97(2), 265-278.

Lucchini, R., Apostoli, P., Perrone, C., Placidi, D., Albini, E., Migliorati, P., Mergler, D., Sassine, M.P., Palmi, S., Alessio, L., 1999. Long-term exposure to 'low levels' of manganese oxides and neurofunctional changes in ferroalloy workers. Neurotoxicol. 20(2-3), 287-297.

Lucchini, R., Selis, L., Folli, D., Apostoli, P., Mutti, A., Vanoni, O., Iregren, A., Alessio, L., 1995. Neurobehavioral effects of manganese in workers from a ferroalloy plant after temporary cessation of exposure. Scand. J. Work Environ. Health 21(2), 143-149.

Mergler, D., Huel, G., Bowler, R., Iregren, A., Belanger, S., Baldwin, M., Tardif, R., Smargiassi, A., Martin, L., 1994. Nervous system dysfunction among workers with long-term exposure to manganese. Environ. Res. 64, 151-180.

Myers, J.E., Thompson, M.L., Ramushu, S., Young, T., Jeebhay, M.F., London, L., Esswein, E., Renton, K., Spies, A., Boulle, A., Naik, I., Iregren, A., Rees, D.J., 2003. The nervous system effects of occupational exposure on workers in a South African manganese smelter. Neurotoxicology 24(6), 885-894.

Nong, A., Teeguarden, J.G., Clewell, H.J., Dorman, D.C., Andersen, M.E., 2008. Pharmacokinetic modeling of manganese in the rat IV: Assessing factors that contribute to brain accumulation during inhalation exposure. J. Toxicol. Environ. Health A 71(7), 413-426.

Nong, A., Taylor, M.D., Clewell, H.J., Dorman, D.C., Andersen, M.E., 2009. Manganese tissue dosimetry in rats and monkeys: accounting for dietary and inhaled Mn with physiologically based pharmacokinetic modeling. Toxicol. Sci. 108(1), 22-34.

Rindernecht, A., McGregor, J., Rouse-Ho, A., Kleinman, M., 2005. Environmental air pollution and in utero brain damage: Maternal manganese (Mn) inhalation alters brain development and susceptibility to postnatal brain injury. Am. J. Obstet. Gynecol. 193(6 (Suppl.)), S36.

Roels, H.A., Ghyselen, P., Buchet, J.P., Ceulemans, E., Lauwerys, R.R., 1992. Assessment of the permissible exposure level to manganese in workers exposed to manganese dioxide dust. Br. J. Ind. Med. 49, 25-34.

Santamaria, A.B., 2008. Manganese exposure, essentiality & toxicity. Indian J. Med. Res. 128(4), 484-500.

Schroeter, JD; Nong, A; Yoon, M; Taylor, MD; Dorman, DC; Andersen, ME; Clewell, HJ III. 2011. Analysis of manganese tracer kinetics and target tissue dosimetry in monkeys and humans with multi-route physiologically based pharmacokinetic models. Toxicol. Sci. 120 (2), 481-498.

Taylor, MD; Yoon, M; Schroeter, JD; Dorman, DC; Andersen, ME; Clewell, HJ (III) 2010. "Target-tissue dosimetry modeling to support the risk assessment of manganese." Toxicologist 114 (1) : 189. Abstract No. 885. In Society of Toxicology 49th Annual Meeting, Salt Lake City, Utah.

Teeguarden, J.G., Dorman, D.C., Covington, T.R., Clewell, H.J., Andersen, M.E., 2007a. Pharmacokinetic modeling of manganese. I. Dose dependencies of uptake and elimination. J. Toxicol. Environ. Health A 77(18), 1493-1504.

Teeguarden, J.G., Dorman, D.C., Nong, A., Covington, T.R., Clewell, H.J., Andersen, M.E., 2007b. Pharmacokinetic modeling of manganese. II. Hepatic processing after ingestion and inhalation. J. Toxicol. Environ. Health A 77(18), 1505-1514.

Teeguarden, J.G., Gearhart, J., Clewell, H.J., Covington, T.R., Nong, A., Andersen, M.E., 2007c. Pharmacokinetic modeling of manganese. III. Physiological approaches accounting for background and tracer kinetics. J. Toxicol. Environ. Health A 70(18), 1515-1526.

US EPA, 1994. Methods for derivation of inhalation reference concentrations and application of inhalation dosimetry. Environmental Criteria and Assessment Office (Research Triangle Park, NC) October. EPA-600/8-90/066F. Accessed on September 30, 2008 at http://cfpub.epa.gov/ncea/cfm/recordisplay.cfm?deid=71993.

US EPA, 2002. December 2002 Final Report A Review of the Reference Dose and Reference Concentration Processes. EPA/630/P-02/002F.

US EPA, 2009. EPA-540-R-070-002 OSWER 9285.7-82 January 2009 Risk Assessment Guidance for Superfund Volume I: Human Health Evaluation Manual (Part F, Supplemental Guidance for Inhalation Risk Assessment), Final.

World Health Organization (WHO), 2001. Chapter 6.8: Manganese. WHO Regional Publications, European Series No. 91. In Air Quality Guidelines for Europe, Second Edition. (World Health Organization, Regional Office for Europe), Accessed on June 01, 2009 at http://www.euro.who.int/document/aiq/6_8manganese.pdf 13p.

Yoon, M; Nong, A; Clewell, HJ; Taylor, MD; Dorman, DC; Andersen, ME. 2009a. Lactational transfer of manganese in rats: Predicting manganese tissue concentration in the dam and pups from inhalation exposure with a pharmacokinetic model. Toxicol. Sci. 112 (1), 23-43.

Yoon, M; Nong, A; Clewell, HJ; Taylor, MD; Dorman, DC; Andersen, ME. 2009b. Evaluating placental transfer and tissue concentrations of manganese in the pregnant rat and fetuses after inhalation exposures with a PBPK model. Toxicol. Sci. 112 (1), 44-58.

Yoon, M; Schroeter, JD; Nong, A; Taylor, MD; Dorman, DC; Andersen, ME; Clewell HJ. 2011. Physiologically Based Pharmacokinetic Modeling of Fetal and Neonatal Manganese Exposure in Humans: Describing Manganese Homeostasis during Development. Toxicol. Sci. May 27 [Epub ahead of print], doi : 10.1093/toxsci/kfr141.

Young, T., Myers, J.E., Thompson, M.L., 2005. The nervous system effects of occupational exposure to manganese – measured as respirable dust – in a South African manganese smelter. Neurotoxicol. 26(6), 993-1000.

Further Information:

Bailey L.A, Goodman J.E., and Beck B.D. (2009). Proposal for a revised Reference Concentration (RfC) for manganese based on recent epidemiological studies. Regul. Toxicol. Pharmacol. 55, 330-339. Available at: http://www.ncbi.nlm.nih.gov/pubmed/19686793.