Review of the Draft Public Health Goal for Nitrate and Nitrite in Drinking Water by

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Overall, I found the OEHHA *Draft Public Health Goal for Nitrate and Nitrite in Drinking Wa*ter to be very well written and a useful summary of the scientific evidence concerning the health hazards associated with nitrate and nitrite in drinking water. However, I did find that the review of the human studies fell short in certain respects, and I question the basis for the proposed Public Health Goal (PHG) of 45 ppm for reasons discussed below.

I am particularly concerned that the proposed PHG of 45 ppm (10 mg/L of nitrogen) of nitrate and 3 mg/L of nitrite is not sufficiently protective for adverse effects in children for several reasons. The PHG is based on the assumption that there is a no adverse effect level (NOAEL) of 45 ppm for nitrate, and that there is an uncertainty factor (UF) of one (i.e. no uncertainty). The justification for the UF of 1 presented in the documents is that it was based on "human data" data in the most sensitive population (i.e. infants and children). I believe this justification is inadequate. The U.S. EPA guidelines for assessing health risk of environmental exposures to children (U.S. EPA 2006) recommends that an intraspecies uncertainty factor (UF_H) of 10 be used to account for "variation in susceptibility among the members of the human population (i.e., interindividual or intraspecies variability)" (U.S. EPA 2006). The OEHHA guidelines (Appendix II) appear to allow for a UF_H factor of 1 when human study includes sensitive subpopulations (e.g. infants and children). However, even within children there is certainly variability in susceptibility. For example, the draft assessment states that "inherited metabolic disorders, such as a deficiency of NADH diaphorase (Kumar et al., 1989) and genetically controlled deficiencies of the enzymes glucose-6-phosphate dehydrogenase or metHb reductase have also been described to cause methemoglobinemia". It seems likely that children with these conditions might be particularly susceptible to the effects of nitrate in drinking water. One cannot assume that the relatively small case reports that have been conducted have included children with these or other conditions that may make them more susceptible.

A NOAEL of 45 mg/L also seems to be highly uncertain. The draft proposal indicates that the NOEAL was based on two small and very old case reports (Bosch 1950 and Walton 1951) and one more recent cross-sectional study (Sadeq 2008). However, several other case reports cited in the review appear to have observed cases at levels below 45 mg/L. The study by Gupta (1999) reported metHB levels of 12.7% in infants ($\leq$1 year) exposed to 26 mg/L. The review dismisses these findings largely because there was no control group in this study. However, these levels appear to be remarkably high given background levels of metHB are generally < 2%. In the report by Knobeloch

and Proctor (2001) one of the cases appears to have had exposure of < 25 mg/L. Given the limitations in the studies for methemoglobinemia, one could also argue that another uncertainty factor could be applied for uncertainty in the database. All in all, it seems that some margin of safety should be applied in developing the proposed (and current) PHG. Common sense suggests that setting a PHG (45 mg/L) so close to the level at which there are known effects (50 mg/L) is not wise. What UF to use is largely a risk assessment policy decision but assuming a UF of 1 is clearly inappropriate.

I am also concerned that the review rejects all of the other endpoints (e.g. thyroid, reproductive and developmental effects and carcinogenicity) for consideration in developing the PHG. These endpoints were rejected due to "inconsistency among the studies, limitation of study designs, and the presence of confounding factors". In general, there are almost always inconsistencies and limitations in the epidemiologic literature of environmental hazards. A careful review tries to determine if these inconsistencies can be explained by the study characteristics. In this respect, I believe the report failed in some areas of its review of the human studies. Of particular concern, is its review on developmental and reproductive toxicity. I believe that there is now very strong evidence of an association between nitrate in drinking water and the risk of central nervous system (CNS) birth defects and particularly for neural tube defects (NTD). An increased risk of CNS birth defects has been reported in four of the studies summarized in Table 9 (Arbuckle et al. 1988, Brender et al. 2004a, Croen et al. 2001 and Dorsch et al. 1984). One of the studies listed in Table 9 was negative for NTDs (Ericson et al. 1988). However, the EPA review does not discuss that there are aspects of its study design that might explain their null findings. As noted in Table 9, this study was based on the earliest known maternal address and thus may have been subject to misclassification of exposures since many women move during their pregnancies. Furthermore, this study was relatively small (only 145 cases) and only presented crude findings that were not adjusted for any other potential risk factors. The review also does not include the most recent and informative study performed to date by Brender et al. (2013). This population based case-control study reported a significant association and a linear trend between maternal ingestion of nitrate in drinking water and the risk of NTDs, limb deficiencies and oral cleft defects. This study is notable because it is the first to include information on water consumption habits (e.g. use of bottled water) at work and at home. The study is also unique in that it included data on dietary sources of nitrate and nitrite, the use of nitrosatable drugs, and all major risk factors for birth defects. The biologic plausibility of the observed increased risk of CNS BD is supported by several experimental animal studies in which an increased risk of CNS birth defects with exposure to N-nitroso compounds has been observed (Givelber and DiPaolo 1969, Koyama et al. 1970 and Pfaffenroth et al. 1974). These experimental studies are relevant and should have been discussed in the document.

The findings from the most recent study by Brender et al. (2013) along with the previous epidemiologic and toxicological studies provide strong evidence of an increased risk of CNS birth defects from maternal consumption of water contaminated with nitrate. It is always difficult to infer causality from observational studies. However, I have participated in many IARC evaluations for carcinogenicity and I believe that the evidence for CNS birth defects from in utero exposure to nitrate meets their criteria for sufficient evidence in human studies, which is that a positive association has been

observed “in which chance, bias and confounding could be ruled out with reasonable confidence”. I would strongly urge the Cal EPA to reconsider the evidence for this endpoint and whether this endpoint might be used as an alternative basis for their assessment of risk.

I am also aware of a few relevant publications that were not discussed in the section on reproductive and developmental effects. Two of these are very recent publications one of which I authored (Stayner et al. 2017) and the other that is a doctoral dissertation by one of my students (Almberg et al. 2017). In both studies, we found evidence of an association between nitrate in drinking water and the risk of very low birth weight. The third study is a publication by Migeot et al. (2103) which reported an association between nitrate in drinking water and the risk of being small for gestational age. While it is understandable how the review could have missed the most recent papers from my research group it is more difficult to understand how it missed the paper by Migeot et al. (2013), and the other papers on birth defects cited above (Brender et al. 2013, Givelber and DiPaolo 1969, Koyama et al. 1970 and Pfaffenroth et al. 1974). This raises questions about the thoroughness of the literature search and the approach used for this systematic review. The missing papers that I have identified are from the areas of research on this topic that I am the most familiar with, and it is quite possible that papers are missing from other areas. Judging from the reference list it appears that the review only included papers published before 2015. The document should at least mention the literature search strategy and criteria used for selecting papers for review.

Finally, I am aware of a few recent epidemiologic studies on bladder cancer (Jones et al. 2016 and 2017) and colon cancer (Espejo-Herrera et al. 2016) that showed a positive association with nitrate in drinking water that were not included in this review. I am also aware of another study on colon cancer and nitrate in drinking water that should be published in the next few months.

Additional references not cited in the OEHHA review

Almberg, Kirsten S. *Adverse Birth Outcomes and Contamination of Drinking Water by Arsenic, Atrazine, and Nitrate-Nitrite. 2016.* (Doctoral Dissertation). (Thesis). University of Illinois – Chicago. Retrieved from http://hdl.handle.net/10027/21275

Brender JD, Weyer PJ, Romitti PA, Mohanty BP, Shinde MU, Vuong AM, Sharkey JR,Dwivedi D, Horel SA, Kantamneni J, Huber JC Jr, Zheng Q, Werler MM, Kelley KE,Griesenbeck JS, Zhan FB, Langlois PH, Suarez L, Canfield MA; National Birth Defects Prevention Study. Prenatal nitrate intake from drinking water and selected birth defects in offspring of participants in the national birth defects prevention study. Environ Health Perspect. 2013 Sep;121(9):1083-9.

Espejo-Herrera N, Gràcia-Lavedan E, Boldo E, Aragonés N, Pérez-Gómez B, Pollán M, Molina AJ, Fernández T, Martín V, La Vecchia C, Bosetti C, Tavani A, Polesel J, Serraino D, Gómez Acebo I, Altzibar JM, Ardanaz E, Burgui R, Pisa F,Fernández-Tardón G, Tardón A, Peiró R, Navarro C, Castaño-Vinyals G, Moreno V,Righi E, Aggazzotti G, Basagaña X, Nieuwenhuijsen M, Kogevinas M, Villanueva CM. Colorectal cancer risk and nitrate exposure through drinking water and diet. Int J Cancer. 2016 Jul 15;139(2):334-46.

Givelber HM, DiPaolo JA. Teratogenic effects of N-ethyl-N-nitrosourea in the Syrian hamster. Cancer Res. 1969;29(5):1151-5.

Jones RR, Weyer PJ, Dellavalle CT, Robien K, Cantor KP, Krasner S, Beane Freeman LE, Ward MH. Ingested nitrate, disinfection by-products, and kidney cancer risk in older women. Epidemiology. 2017 Mar 1. doi: 10.1097/EDE.0000000000000647. [Epub ahead of print.

Jones RR, Weyer PJ, DellaValle CT, Inoue-Choi M, Anderson KE, Cantor KP, Krasner S, Robien K, Freeman LE, Silverman DT, Ward MH. Nitrate from Drinking Water and Diet and Bladder Cancer Among Postmenopausal Women in Iowa. Environ Health Perspect. 2016 Nov;124(11):1751-1758.

Koyama T, Handa J, Handa H, Matsumoto S. Methylnitrosourea-induced malformations of brain in SD-JCL rat. Arch Neurol. 1970;22(4):342-7.

Migeot V, Albouy-Llaty M, Carles C, Limousi F, Strezlec S, Dupuis A, Rabouan S. Drinking-water exposure to a mixture of nitrate and low-dose atrazine metabolites and small-for-gestational age (SGA) babies: a historic cohort study. Environ Res. 2013;122:58-64.

Pfaffenroth MJ, Das GD, McAllister JP. Teratologic effects of ethylnitrosourea on brain development in rats. Teratology. 1974;9(3):305-15.

Stayner LT, Almberg K, Jones R, Graber J, Banda E, Pedersen M and Turyk M. Atrazine and Nitrates-Nitrites in Drinking Water and the Risk of Preterm Delivery and Low Birth Weight in Four States in the Midwestern United States. Environmental Research. 2017:152;294-303.

U.S. Environmental Protection Agency (EPA). (2006) A framework for assessing health risks of environmental exposures to children. National Center for Environmental Assessment, Washington, DC; EPA/600/R-05/093F. Available from the National Technical Information Service, Springfield, VA, and online at http://www.epa.gov/ncea.