820-R-10-016

COMMENT-RESPONSE SUMMARY REPORT

for the

PEER REVIEW

of the

FLUORIDE:

DOSE-RESPONSE ANALYSIS FOR NON-CANCER EFFECTS

DOCUMENT

November 2010

Office of Water
Office of Science and Technology
Health and Ecological Criteria Division

U.S. Environmental Protection Agency
Washington, D.C. 20004

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ACKNOWLEDGMENTS

This document was prepared with the technical assistance of the Toxicology and Hazard Assessment Group of the Environmental Sciences Division of Oak Ridge National Laboratory, Oak Ridge, Tennessee, under EPA IA No. DW-89-92209701 and DOE IAG No. 1824-S881-A1. Principal EPA Scientists are Joyce Morrissey Donohue, PhD, and Tina Duke, MPH, Health and Ecological Criteria Division, Office of Science and Technology, Office of Water, U.S. Environmental Protection Agency, Washington, DC.

The Oak Ridge National Laboratory is managed and operated by UT-Battelle, LLC., for the U.S. Department of Energy under Contract No. DE-AC05-00OR22725.

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I. INTRODUCTION

The United States Environmental Protection Agency (EPA), Office of Water is charged with protecting public health and the environment from adverse exposure to chemicals and microbials in water media, such as ambient and drinking waters, waste water/sewage sludge and sediments. In support of this mission, the Office of Water/Office of Science and Technology (OST) develops health standards, health criteria, health advisories, and technical guidance documents for water and water-related media. Under this work assignment, documents prepared by OST are to undergo external peer review.

Peer review is an important component of the scientific process. It provides a focused, objective evaluation of a research proposal, publication, risk assessment, health advisory, guidance or other document submitted for review. The criticisms, suggestions and new ideas provided by the peer reviewers ensure objectivity, stimulate creative thought, strengthen the reviewed document and confer scientific credibility on the product. Comprehensive, objective peer review leads to good science and product acceptance within the scientific community.

The Peer Review for *Fluoride: Dose-Response Analysis for Non-cancer Effects* was conducted on March 11, 2008, in Washington, DC, to allow the external peer reviewers to discuss their evaluations of the EPA/OW document. The Peer Review was conducted under EPA Contract Number EP-C-07-021 with ToxServices, Washington DC (Work Assignment B-02 Task 5) and managed by ICF International, Fairfax, VA.

The list of external peer reviewers and their affiliations are shown below:

Jane A. Cauley, Dr.P.H, Professor, Department of Epidemiology, University of Pittsburgh

Pamela Den Besten, D.D.S., M.S., Professor and Chair, Division of Pediatric Dentistry, Department of Orofacial Sciences, University of California at San Francisco

Richard D. Jackson, D.M.D., Assistant Professor, Preventive and Community Dentistry, School of Dentistry, Indiana, University, Oral Health Research Institute

Gary M. Whitford, D.M.D., Ph.D., Regents' Professor, Department of Oral Biology and Maxillofacial Pathology, Medical College of Georgia

The Charge to the Peer Reviewers is presented in Section II of this report. The complete peer review process, including pre-peer review meeting comments, post-peer review meeting

comments and a summary of the meeting comments, as prepared by ICF, are presented in Section V. OW, with the assistance of ORNL, prepared a response to each of the general comments of the reviewers as summarized by ICF, as well as point by point responses to the specific comments of each of the peer reviewers; and these are presented Section III. One reviewer (Dr. Whitford) had major concerns about the analytical methodology that was used in the 1930's and 40's to measure fluoride in drinking water. As a result, ORNL provided a detailed evaluation of the analytical methodology, and this is presented in Section IV.

II. CHARGE TO THE PEER REVIEWERS

1. Was the document clear and transparent in its presentation of data and explanation of the analytical approaches used to characterize the concentration-response and dose-response relationships for severe dental fluorosis? If not, do you have any suggestions that will enable the authors to improve its clarity?

2. Are you aware of any significant publications related to severe dental fluorosis or the skeletal effects of fluoride that are not included in the noncancer assessment document?

3. Are the strengths and weaknesses of Dean (1942) study (selected as critical) fully characterized?

4. Are some teeth more susceptible to severe fluorosis than others? If so, should the OW make modifications to the age range identified as the period of concern?

5. Are the data on cavities as collected and graphically presented by the OW consistent with the hypothesis that there is an increased risk for cavities in susceptible individuals with severe dental fluorosis?

6. Are there recent data that would impact the Institute of Medicine (IOM, 1997) Adequate Intake Value of 0.05 mg/kg/day for fluoride that the OW should consider in its assessment?

7. Can you suggest an approach to transform the water concentration data from the Dean (1942) study to units of dose for the population susceptible to severe dental fluorosis other than that used by the OW?

8. Can you provide input concerning the strengths and weaknesses of the approach utilized by the OW to identify a lower bound dose for severe dental fluorosis?

9. Are you aware of dose estimates other than those from IOM (1997) and the World Health Organization (WHO; 2002) that are appropriate critical doses for skeletal effects and/or can you suggest a different approach that the OW might use to estimate the fluoride dose associated with skeletal fractures using available data?

10. Are you aware of any data that can be used to demonstrate that protection of the secondary teeth from severe dental fluorosis will also protect the primary teeth?

11. Are there factors other than those discussed in Section 3.1.4 that increase sensitivity to fluorosis of the teeth or bones?

12. Do you support the OW's conclusion that an RfD of 0.07 mg/kg/day will be protective for severe dental fluorosis in children and skeletal effects in adults while still providing for the beneficial effects of fluoride?

III. PEER REVIEW COMMENTS AND EPA RESPONSES

1. Was the document clear and transparent in its presentation of data and explanation of the analytical approaches used to characterize the concentration-response and dose-response relationships for severe dental fluorosis? If not, do you have any suggestions that will enable the authors to improve its clarity?

Reviewers' General Comments. In general, the reviewers thought that the document was clear and transparent. In addition, the reviewers commented that the document clearly described the available literature and presented the information in an understandable format. In particular, Dr. Jackson noted that the individual tables summarizing the studies were very helpful.

EPA Response: No response needed.

Specific Comments:

Dr. Cauley: Recommended reorganizing the document in the format of a manuscript. She also asked for clarification on OW and NRCs roles in the project.

EPA Response: EPA reports are not normally written in the format of a research manuscript; however, additional information on background and rationale and purpose of the report were provided in a preface and added to the document.

Dr. DenBesten: Page 13. State the purpose of the Secondary Maximum Contaminant Level.

EPA Response: The definition of a National Secondary Drinking Water Standard was added to the text.

Dr. Den Besten: Page 15. The statement with regards to fluoroapatite, at the end of paragraph 2.1.1 is not accurate. The mineral formed in tooth enamel exposed to higher fluoride levels is fluoride containing carbonated apatite. Fluoride levels in subsurface fluorotic enamel are about 200 ppm rather than the 10-100 ppm fluoride in normal enamel, whereas fluorapatite is about 30,000 ppm fluoride. Precipitation of fluoride mineral salts at the surface of enamel results in high surface level, though this also is not fluoroapatite. This fluoride-substituted apatite has some increased resistance to bacterial acids that cause tooth decay. However, the primary function of fluoride in drinking water in reducing tooth decay is topical, primarily by the enhancement of remineralization.

EPA Response: Reference to "fluoroapatite" has been removed form the text and Dr. Den Besten's clear explanation of the mechanism has been used in its place.

Dr. Den Besten: Page 19 section 2.2. The apatite formed in bone is also a fluoride-substituted hydroxyapatite rather than a fluoroapatite.

EPA Response: Reference to "fluoroapatite" removed from text.

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Dr. Den Besten: Page 21 section 3. Change "…are preferred for evaluating the potential effects of fluoride in drinking water", to the more accurate statement, "…are preferred for evaluating the potential effects of ingested fluoride".

EPA Response: Text modified accordingly.

Dr. Den Besten: Page 57 paragraph 1. It should be made clear that studies on water fluoridation conducted after 1980, are confounded by additional sources of fluoride, and changes in use of tap water. For example, decreasing fluorosis in more recent studies may be related to reduced consumption of tap water as use of bottled water increases. In general for all of section 3.2.2, when fluoride effects on dental caries are discussed, the data should be divided into studies before and after 1980 when fluoride became widely available in toothpaste, and perhaps before and after the late 1990s when bottled beverages became widely used.

EPA Response: Statements similar to the ones requested by Dr. Den Besten are presented in the first paragraph of Section 3.2.2 of the EPA report, although a specific time frame for the introduction of fluoridated toothpaste and the increased use of bottled water is not given. These statements were revised to include Dr. Den Besten's estimates.

Dr. DenBesten: Page 86; Table 3-52. Please indicate what does "complete" and "total" refer to?

EPA Response: Complete means complete fractures; total means total fractures (complete plus incomplete, the latter of which was defined by the study authors as stress fractures observed by roentgenography in participants reporting acute lower extremity pain syndrome.). Table 3-52 has been modified to include the incomplete fractures, and a footnote has been added defining incomplete fractures.

Dr. Den Besten: Page 94 section 4.4. Explain what the "NOAEL/LOAEL" approach is, or at least spell out the acronym.

EPA Response: Acronyms were spelled out, and added to the List of Acronyms.

Dr. Den Besten: Page 97 paragraph 1. The importance of fluoride as a nutrient may need to be reassessed, given that its primary function in caries prevention is topical. It would seem more appropriate to focus on the upper limits for ingestion of this caries preventive agent, and leave it to future panels to assess the relative importance of the IOM's recommended intake of fluoride and risk of severe fluorosis.

EPA Response: The issue of the importance of fluoride as a nutrient is not within the scope of the current document. The primary task of the OW was to assess the dose-response data for severe fluorosis as recommended by the NRC.

Dr. Den Besten: Page 98, paragraph 2, sentence 3. The statement as to the timing of secondary incisor tooth formation is incorrect. The secondary incisors and molars begin development in utero. Change "development" to "mineralization".

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EPA Response: The word "mineralization" was substituted for development; i.e., "The mineralization of the secondary teeth begins at about 6 ± 2 months with the incisors, whereas that for the primary teeth begins *in utero* (Massler and Schour, 1958)".

Dr. Den Besten: Page 101. What is the rationale for setting the BMDL at an incidence of 1% severe fluorosis? I recommend setting the BMDL at an incidence of 99% moderate fluorosis, which would show an intent to eliminate the adverse effect of severe fluorosis secondary to fluoride added to drinking water.

EPA Response: A BMR (Benchmark Response) of 1% is the standard level used by the EPA, as most data sets cannot statistically support calculating a lower level because of the potential variability in background levels. However, if the data set is large enough, a lower response level can be used. A statistical analysis was conducted on the Dean data and it was determined that it would support a lower BMR of 0.5% severe dental fluorosis, but not 0.01%. The BMD software was run for 99% moderate fluorosis; the log probit model predicted a BMD of 31.6 mg/L and a BMDL of 25.6 mg/L (a poor data fit due to the fact that the occurrence of moderate fluorosis for all study populations was less than 50%; therefore, the use of a BMR of 99% moderate dental fluorosis is not considered appropriate.

Dr. Den Besten: Page 103. As stated above, I question a recommended fluoride intake, and feel that this document should focus only on the dose response analysis.

EPA Response: See response above.

Dr. Den Besten: Page 104, paragraph 1. The statement that "…fluoroapatite crystals disrupt the hydroxyapatite crystal lattice…" is incorrect and should be deleted.

EPA Response: That statement was deleted from the text.

2. Are you aware of any significant publications related to severe dental fluorosis or the skeletal effects of fluoride that are not included in the noncancer assessment document?

Reviewers' General Comments: Three of the reviewers, Dr. Cauley, Dr. Jackson, and Dr. Whitford, thought that there were additional publications related to severe dental fluorosis and/or skeletal effects of fluoride that should be reviewed for possible inclusion.

EPA Response: The literature suggested by the reviewers was obtained and evaluated for inclusion in the document.

Specific Comments:

Dr. Cauley noted that while the dental fluorosis literature review in the report covered a wide range of fluoride levels, the skeletal/ fracture studies were limited to those with fluoride levels > 4 mg/L. She suggested that the Office of Water consider several specific publications that discuss the occurrence of fractures at lower levels of fluoride. Citations for the suggested references were provided.

EPA Response: A limited number of additional papers on the association of bone density and skeletal fractures with water fluoridation levels of less than 1 mg/L were added to the report and the reader was referred to several review papers for more detailed summaries. Because the NRC (2006) identified a water fluoride level of 4 mg/L as being the potential threshold for skeletal effects, the EPA report intentionally focuses on such studies rather than those that examined lower levels of fluoride in drinking water.

3. Are the strengths and weaknesses of Dean (1942) study (selected as critical) fully characterized?

Reviewers' General Comments: In general, the reviewers believe that the strengths of the Dean (1942) study were well characterized in the draft dose-response report.

EPA Response: No response needed.

Specific Comments:

Dr. Cauley noted that additional strengths of the study include: (1) the wide range of fluoride concentrations, although there were fewer subjects in the high fluoride categories; (2) the dose-response relationship illustrated by the data showing increasing risk of severe skeletal fluorosis with increasing fluoride; and (3) the consistency of the findings across several different communities.

EPA Response: These additional points were added to the report.

Dr. Jackson doubted that anyone would disagree that the Dean (1942) study has been and will continue to be a benchmark study in the dental literature, as well as the much broader literature related to public health and epidemiology. He noted that Dean's study was performed when confounding fluoride sources were not available and, thus, probably gives a very clear picture of the prevalence of the relationship of fluoride ingestion and the subsequent development of dental fluorosis.

EPA Response: No response needed.

Dr. Cauley and **Dr. Jackson** noted that the Dean (1942) study does not provide information on race/ethnicity, and given the probable characteristics of the region, the population of subjects examined was likely not diverse in terms of racial or cultural characteristics.

EPA Response: Dean (1942: page 31) did indicate that all his study populations were Caucasian. The dose-response report has been modified to communicate this more clearly.

Dr. Cauley noted that the data were collected the late 1930-40's. Although confounding by use of other fluoride products would be minimal, there are many other cohort differences between children exposed to fluoride in the late 1930-40's as compared to now. For example, dental hygiene, dietary intakes (e.g., less water and more carbonated beverages), and body weight are very different in today's children compared to those in the 1930s.

EPA Response: These factors were added to Section 3.1.1 in the listing of weaknesses of the study.

Dr. Cauley questioned whether puberty and/or hormonal changes may influence fluoride effects, which may be important because age of menarche has been decreasing. Another weakness of the studies

reviewed is that there appears to be no information on exposure duration (e.g., How long did these children live in each community? Did the inclusion criteria include a minimum time of residence?).

EPA Response: The concern about hormonal changes was added as a potential weakness of the study. Dean (1942) specifically included only children who were lifetime residents of the communities sampled.

Dr. Jackson further noted that it has been postulated that genetic factors may impact the expression of dental fluorosis at identical levels of ingestion. The fact that data were collected in what may have been an exclusively white population appears to limit its applicability for use as a benchmark.

EPA Response: Additional papers on genetic factors influencing susceptibility to fluoride were added to the report (new Section 3.1.4.4). Only one study was located which specifically looked at fluorosis in both Afro-Americans and Caucasian populations.

Dr. Whitford believes that the weaknesses of the study were not necessarily fully characterized. One example given in his discussion of how the weaknesses of the Dean (1942) study were not fully characterized was the lack of review of the publications presented in his response to Question #2 (Elvove, 1933). He also had the following specific comments:

A major weakness of the Dean (1942) report is the chemical method used for the determination of fluoride concentrations in water (Elvove, 1933). The zirconium-alizarin method is rarely, or probably never, used today because of its relative insensitivity, several interfering substances, and lack of specificity for fluoride. In their 1952 report that described improvements to the method, Megregian and Maier (1952) noted that Elvove's original method (1933) had several shortcomings including "non-conformity to the color laws, limited effective fluoride range, and little color change per increment of fluoride." It also appears that Elvove (1933) used the visual method to determine color changes in the zirconium-alizarin reagent (since he referred to "Nessler tubes") which requires subjective judgments and is less accurate that spectrophotometric methods.

EPA Response: For a complete response to Dr. Whitford's comment, see Section IV of this document. A shortened version of EPA's response was added to the EPA report (see Section 3.1.1). It is acknowledged that the method used by Elvove (1933) did not have the sensitivity or minimum level of detection as more modern methods; however, the results are internally consistent, appear to be supported by later studies on some of the same water sources, used mostly average values of 12 consecutive monthly samples, therefore compensating for potential individual analysis error or seasonal variation, and, based on water quality data from the same time period, were not likely to be compromised by high levels of interfering substances. In addition, according to Megregian and Maier (1952) the reagent used was sensitive to small increments of fluoride over a range of 0.0 to 3.0 ppm, the critical range for assessing the threshold for severe fluorosis, and within this range the response approximated Beer's law.

Dr. Whitford: The study population in the Dean (1942) may not have been continuously exposed to the community's communal water supply. Dean (1942, page 25) listed two major requisites for quantitative evaluation of the dental effects of ingesting water containing fluoride. One of these requisites was "a population continuously exposed throughout life to the variable under investigation (the communal water supply)." **Dr. Whitford** recommended that the original papers summarized in Dean (1942) be examined to determine the extent to which the children met the requisite cited above and that the information be

included in the Dose-Response Analysis document. If such information is not available, then the document should note this and discuss the implications in its conclusions.

EPA Response: By listing "continuous residence" in a community as a pre-requisite for inclusion in a drinking water study, the implication is that Dean would have used only children who were lifetime residents of those particular communities. In support of this supposition is the statement made by Dean (1942, pg. 29) that 289 children studied in Amarillo, TX, "used continuously throughout life the municipal water for drinking and cooking..." In an earlier 1936 paper (Amer. J Public Health 26:567-575), in which Dean evaluated fluorosis in children in 10 cities, it is specifically indicated (Table 11) that the children included in the survey were those who had "used municipal water continuously". Some of these were the same cities studied in 1942. Therefore, the conclusion is that Dean (1942) included in his study only children who were lifetime residents of their communities.

Dr. Whitford further commented that the appropriateness of the LOAEL (2.2 ppm) and the calculated reference dose (RfD) (0.07 mg F/kg bw/day) reported in the OW's draft dose-response report are based largely on the accuracy of the water fluoride concentrations shown in Dean (1942), as well as on several other variables that may have affected the outcomes of the epidemiological studies. He indicated that his preceding comments draw attention to several shortcomings of the chemical method used and other limiting aspects of the studies summarized by Dean (1942) and recommended that the uncertainties associated with these factors be discussed wherever appropriate and certainly in the "Uncertainty Factors" section.

EPA Response: The issues raised by Dr. Whitford's are discussed in Section IV of this report. The uncertainties associated with the analytical method used by Elove are mentioned in the EPA report.

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4. Are some teeth more susceptible to severe fluorosis than others? If so, should the OW make modifications to the age range identified as the period of concern?

Reviewers' Overall Comment: The reviewers had differing opinions on whether some (and which) teeth are more susceptible to severe fluorosis.

EPA Response: No response needed.

Specific Comments:

Dr. Cauley thought age may be a factor in the susceptibility to severe dental fluorosis and suggested that a more direct discussion of the age at risk is needed and that this topic should be highlighted in a separate section. She suggested that, in particular, a table summarizing the ages of the children in each study and the age range which appeared to be at highest risk would be helpful. She recommended that the writers add this to Table 3-16.

EPA Response: Average ages of the study populations were added to Table 3-16. No specific information was found in the available literature indicating that a particular age or tooth type is more susceptible to fluorosis than any other. Because different teeth undergo mineralization at different times, they will have different susceptibility periods; however, as noted below by Dr. Jackson, susceptibility may be a function of the length of the mineralization period.

Dr. Den Besten thought that the report was clear in showing that posterior teeth also are susceptible, and the case for including children up to age 14 was clear and compelling.

EPA Response: No response needed.

Neither **Dr. Jackson** nor **Dr. Whitford** has come across any literature that indicates that some teeth are more susceptible to the development of severe dental fluorosis.

EPA Response: No response needed.

Dr. Jackson believes that the susceptibility is the same assuming that the exposure is constant and taking into consideration how long the developing tooth is exposed to higher levels of fluoride. He stated that, for example, maxillary third molars may present greater evidence of severe fluorosis (pitting, staining, etc) because they take longer to develop (12-16 years-of-age) and erupt as opposed to a maxillary central incisor (5 years-of-age).

EPA Response: No response needed. Dr. Jackson's information is supportive of the age range EPA chose as that for greatest susceptibility to dental fluorosis

Dr. Whitford also hypothesized that the posterior teeth may be more susceptible since their development is more protracted than that of the anterior teeth.

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EPA Response: No response needed. Dr. Whitford's comment is also supportive of including the age for the development of the third molars in the dose-response assessments. Many earlier studies have only included the age to the development of the second molars in their assessment of fluorosis.

5. Are the data on cavities as collected and graphically presented by the OW consistent with the hypothesis that there is an increased risk for cavities in susceptible individuals with severe dental fluorosis?

Reviewers' General Comment: Dr. Cauley, Dr. Den Besten, and **Dr. Jackson** agreed that the data on cavities presented in the report are consistent with the stated hypothesis. **Dr. Whitford** disagreed with the other reviewers, noting that he felt the available data present an unclear relationship.

EPA Response: EPA has characterized the data on an increased risk for cavities in individuals with severe dental fluorosis as suggestive yet supportive of the NRC conclusion that the thinning and pitting of the enamel that occurs with severe dental fluorosis can lead to an increase in cavities as observed in some but not all studies. The data also indicate that at levels beyond very mild fluorosis the reduction in decay compared to the increase in fluorosis becomes less dramatic than that observed between no fluorosis and mild fluorosis.

In evaluating the NRC conclusion, it is important to compare across groups that fall in the same age range and have enough permanent dentition for a long enough period of time to support an evaluation of differences in decayed, missing and filled teeth or decayed, missing and filled surfaces. Many of the studies reviewed did not present the data in a manner that was suitable for evaluating the NRC determination that there is an increased cavity risk for individuals with severe dental fluorosis compared to those with less than severe fluorosis.

Specific Comments:

Dr. Cauley noted that the data summarized in Table 3-40 to 3-44 are convincing that dental caries are more common in subjects with severe fluorosis. The data suggest a U-shaped relationship with fluoride: a higher risk in those with very low intakes and intakes >2–3 mg/L.

Dr. Jackson noted that the OW may want to consider examiner bias in the methodology of these studies. Although it would be impossible to eliminate this bias, it is something that should at least be mentioned as a possible confounder. He also described the findings of the following study (see Appendix B for a description of the study findings):

> Jackson R, Kelly S, Katz B, Hull J, Stookey G. (1995). Dental fluorosis and caries prevalence in children residing in communities with different levels of fluoride in the water. *J Publ Health Dent.* 88:79-84.

EPA Response: Text was added to Section 3.2 to point out the possibility of examiner bias. The reference mentioned by Dr. Jackson was ordered and incorporated into the report.

Dr. Whitford disagreed with the other reviewers, noting that the available data present an unclear relationship. He discussed conflicting results as follows: Driscoll's work (1983, 1986) in Illinois and Iowa did not indicate a relationship between severe fluorosis and caries (pages 46- 47). Eklund's report

(1987) did not find a relationship when the entire dentition was considered. They found more caries in severely fluorosed anterior teeth and premolars but not in the molars. In their Chinese study, Chen et al (1989) found no difference in caries scores between the group without fluorosis and the group with severe fluorosis. Warnakulasuriya et al (1992) reached a similar conclusion in their Sri Lanka study but the validity of the conclusion was less clear because of the way they grouped the fluorosis categories. On the other hand, Mann et al (1987, 1990) and Olsson (1979) found that DMFS scores were directly related to the severity of fluorosis in Israel as did Wondwossen et al (2004) in Ethiopia. Ermis et al (2003) reported a slightly higher prevalence of caries in moderate-to-severely fluorosed teeth but the relationship was not statistically significant

Dr. Whitford noted that overall, and as summarized in Figure 3-7 on page 71, the relationship between the severity of dental fluorosis and the risk of caries is suggestive, but not convincing. He believes that this subject requires more study with control for variables that are known risk factors for caries before a reasonably firm conclusion can be drawn about the possibility of an association between severe dental fluorosis and an increased risk of caries.

EPA Response: EPA agrees that the results of the various studies are not entirely consistent (as is documented in Tables 3-16 and 3-40 in the EPA report), but notes that some of the comparisons cited by Dr. Whitford are between caries scores in populations without fluorosis and those with severe fluorosis. Groups without fluorosis are likely to include individuals not receiving the anticaries benefits of fluoride; therefore, the results would be biased towards higher levels of caries in the non-fluorosis group, which would obscure the significance of any increase in caries at the higher levels of fluorosis compared to those at the lower levels. When these individuals are excluded from the evaluation (see results of the Driscoll et al. 1986 study as shown in Table 3-41 in the EPA report), there is clear support for an increase in caries with severe fluorosis. Because the Driscoll et al. study was conducted in the U.S., its results are likely to be more relevant than studies conducted on non-U.S. populations. Nevertheless, Dr. Whitford's suggestion that additional studies are needed that control for variables that are known risk factors for caries is a valid one, and this suggestion has been added to the text of Section 3.2.4 of the EPA report.

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6. Are there recent data that would impact the Institute of Medicine (IOM, 1997) Adequate Intake Value of 0.05 mg/kg/day for fluoride that the OW should consider in its assessment?

Reviewers' General Comments: Dr. Cauley, Dr. Jackson, and Dr. Whitford were unaware of any recent data that could influence the adequate intake (AI) value.

EPA Response: No response needed.

Specific Comments:

Dr. Cauley suggested that the writers mention life stage and the upper limit of toxicity. She pointed out that the American Dental Association website may have updated their fluoride document in 2006. In her final comment for Question 6 (see Appendix B), she provided two tables: Criteria and Dietary Reference Intake Values for Fluoride by Life Stage Group and Tolerable Upper Intake Levels by Life Stage Group.

EPA Response: Life stage and the upper limit of toxicity (IOM Criteria) are discussed in the EPA report. Parts of the tables listing the IOM Criteria have been added to the EPA report (as new Table 5-1). The ADA Report basically supports the USPHS fluoride drinking water levels.

Dr. Whitford: Although **Dr. Whitford** did not know of any additional data, he commented on the interpretation of the IOM's values. He noted that the IOM's AI value represents the amount of intake of any substance "needed to maintain a defined nutritional state or criterion of adequacy in essentially all members of a specific healthy population." In the case of fluoride, the AI (see page 301, Dietary Reference Intakes, 1997) "is based on estimated intakes that have been shown to reduce the occurrence of dental caries maximally in a population without causing unwanted side effects including moderate dental fluorosis." He noted that this does not mean that intakes somewhat higher than 0.05 mg/kg/day increase risk of moderate dental fluorosis. In fact, the IOM's estimate for the threshold for that risk is 0.10 mg/kg/day.

EPA Response: The IOM's estimate of 0.10 mg/kg/day as the threshold for risk of moderate fluorosis is in the EPA Report (see Section 5.1 Nutritional Guidelines). The IOM (1997) based this estimate on the data of Dean (1942), concluding that there was less than a 5% prevalence of moderate fluorosis at 2 mg F/L. EPA ran the Benchmark Dose software using the Dean data for moderate fluorosis (excluding the three highest concentrations as they appeared to be outliers) and, from a log probit analysis, derived a BMD of 2.17 mg/L and a BMDL of 2.03 mg/L for 5% moderate fluorosis. Using the same log probit model in the BMD software, the BMD for 1% severe fluorosis is 2.31 mg/L and the BMDL is 2.13 mg/L. Therefore, IOM's "threshold" for moderate fluorosis is at a level close to the level associated with 1% severe fluorosis.

Dr. Den Besten disagreed with the presumption that the IOM's value is sufficient. The weight of evidence indicates that the primary mechanism by which fluoride protects against tooth decay is a topical

effect. Therefore, she noted that the IOM's recommendation of an adequate intake value, at least relating to tooth decay, should be reassessed.

EPA Response: If it becomes generally accepted that the beneficial anti-caries effect of fluoride is achieved only through topical application, the status of fluoride as a "nutrient" may be subject to re-evaluation. However, as long as an Adequate Intake for fluoride is recognized by authoritative groups such as the IOM, EPA is obliged to treat fluoride as it does other substances which have recommended AIs by taking those values into consideration when proposing an oral Reference Dose.

7. Can you suggest an approach to transform the water concentration data from the Dean (1942) study to units of dose for the population susceptible to severe dental fluorosis other than that used by the OW?

Reviewers' General Comments: The reviewers were generally accepting of the approach EPA used in estimating doses from drinking water using the Dean (1942) data on drinking water fluoride concentrations associated with severe dental fluorosis and estimates from Ershow and Cantor (1989) for water intake and body weight. Several were of the opinion that EPA should present their dose estimates without selecting a Reference Dose, leaving that decision to the users of the document.

EPA Response: Although the Office of Water might be able to use the drinking water concentration data as the basis for their evaluation of the MCLG and MCL for fluoride, an RfD is required by other programs within EPA. Accordingly, the post peer review version of the dose-response document still presents an RfD based on the drinking water data and an estimate for the contributions to total exposure from the diet at the time for the Dean (1942) study.

Specific Comments:

Dr. Cauley suggested that writers use more generalizable data from the National Health and Nutrition Examination Survey (NHANES) for body weight. She noted that if this approach were used, the writers could still utilize the water intake estimates from the Ershow and Cantor (1989) paper.

EPA Response: The objective was to use data that related most closely to the time period when the Dean study was conducted (1942). The Ershow and Cantor (1989) data were from the 1977/1978 Nationwide Food Consumption Survey. NHANES I included surveys for 1971-1975. Although the records indicate that 24-hour recall dietary information was collected for individuals ages 1 to 74 during NHANES I (http://www.cdc.gov/nchs/nhanes/nhanesi.htm), EPA found no indication that the data had been analyzed for age-specific water consumption as have the data from the Nationwide Food Consumption Survey.

Dr. Den Besten and Dr. Jackson noted several difficulties in developing an approach for transforming the water concentration data from the Dean study to units of dose for the population susceptible to severe dental fluorosis.

EPA Response: The Dose-Response report acknowledges the limitations of the approach that was applied.

Dr. Den Besten stated that serum fluoride levels would be the most useful measurement, but these levels are not available. She believes that using Dean's data as a starting point to quantify total ingestion is the best option. She recommended that future studies include random sampling of serum fluoride levels to strengthen future decision-making relative to fluoride intake.

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EPA Response: A workgroup representing several EPA program offices has submitted a list of chemical nominations for future biomonitoring efforts at CDC through NHANES. Fluoride was among the EPA nominations listed as a tier 1 high priority chemical for future monitoring cycles.

Dr. Jackson noted that the U.S. marketplace is constantly changing and ingestion of tap water continues to decline. U.S. EPA's (2004) report stated that bottled water accounted for only 13% of water consumption in the United States. More recent trade manufacturing data indicates that bottled water consumption in the United States exceeds this percentage by a wide margin, and bottled water consumption may surpass tap water consumption in the near future. Other published data suggest that among Hispanic individuals, tap water is commonly perceived as "unhealthy" and again bottled water is almost exclusively consumed. Another point that should be further explored is the possible "halo effect" of imported foods and beverages into the United States and the fluoride content of these consumables.

EPA Response: Increases in bottled water consumption will be taken into consideration by EPA during the preparation of the Relative Source document. Use of bottled water is assumed to have been minimal by the children in Dean's (1942) study populations.

8. Can you provide input concerning the strengths and weaknesses of the approach utilized by the OW to identify a lower bound dose for severe dental fluorosis?

Reviewers' General Comments: Dr. Cauley and **Dr. Den Besten** listed strengths and weaknesses to the approach utilized by OW to identify a lower bound dose for severe dental fluorosis.

Strengths:

Dr. Cauley said that the main strengths of the approach were that: (1) conservative estimates were carried out with and without the outlier community; (2) several different models were considered; and (3) sensitivity analysis showed that even eliminating three highest data points had little effect on the model goodness of fit or on the BMDL.

Dr. Den Besten added that: (1) the data used were limited to studies found in the Dean report where the only exposure to fluoride was drinking water; and (2) a careful assessment was done regarding tap water consumed and mean body weights.

Weaknesses:

Dr. Cauley said that the inherent weakness of the approach relates primarily to the weaknesses previously cited related to the parent Dean (1942) study. The writers may consider adding a formal test for trend in data.

EPA Response: The EPA appreciated the acknowledgement of the strengths of the approach applied. In response to Dr. Cauley's recommendation that EPA add a test for trend in the data, the Cochran-Armitage Test for Tend was applied to the Dean data and the results indicated that there was a positive trend for increasing fluorosis with increasing fluoride concentration in drinking water.

Specific Comments:

Dr. Den Besten added that the assumption that 0.05 mg/kg/day is a required amount of fluoride and that dose estimates must be above this level could be considered a weakness because the purpose of U.S. EPA's analysis was to determine risk, not to conduct risk benefit analyses. In addition, she noted that the assumption that the small number of children who displayed severe dental fluorosis were those who had excess exposure to fluoride could also be considered a weakness. This assumes that these children drank significantly more water since water was the only source of fluoride. It is more likely that genetic or other causes are responsible for this small outlier group.

EPA Response: EPA's recognition of 0.05 mg/kg/day as a required amount of fluoride is based on the IOM's conclusion that this dose represents an Adequate Intake (AI) which reduces the occurrence of caries without unwanted side effects. The original choice of the 90th percentile for water consumption in the derivation of the RfD was based on EPA policy that drinking water regulations be derived for the protection of consumers at that level of exposure. The possibility that some children may be inherently more susceptible to fluoride, at least above some minimal exposure level, is very plausible; however, at the moment, there is no way to quantify the increased risk or the size of such a subpopulation. Using the

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90th percentile for water consumption could, in a way, be considered a surrogate adjustment for increased susceptibility in a segment of the entire population. However, as recommended by the peer reviewers, the dose calculations for the mean, 75th, 90th, and 95th percentile are presented in the final document so that they can be considered from a risk management perspective.

Dr. Whitford expressed additional concerns about the approach utilized by the OW, particularly in reference to the determination of the LOAEL. He proposed that the variability among published studies regarding the relationship between estimated fluoride intakes and the risk of severe dental fluorosis may be due to the relative altitude of the cities that were studied. He provided a detailed discussion of this issue in his final comment for Question 8. He suggested that based on the variability among published studies regarding the relationship between estimated fluoride intakes and the risk of severe dental fluorosis, consideration should be given to establishing a lower bound dose other than 1 percent.

Dr. Whitford had the following to say about the Clovis, New Mexico, data in Table 3-1 (page 24). "The water fluoride concentration was listed as 2.2 ppm, the lowest concentration at which severe dental fluorosis was recorded. The prevalence was 0.7 percent, i.e., only one subject out of the 138 examined exhibited what was classified as having severe fluorosis. This concentration, 2.2 ppm, was selected as the LOAEL. Unfortunately, nothing is known about the individual with severe fluorosis including whether he/she was a permanent resident of Clovis or had lived in one or more other communities before moving to Clovis. The prevalence of moderate dental fluorosis (Dean score 3) in Clovis was 11.0 percent. These prevalence values for moderate and severe fluorosis are markedly higher than those for Elmhurst and Galesburg (about 1.1 percent for moderate and an absence of severe dental fluorosis) where the water fluoride concentrations were listed as 1.8 and 1.9 ppm, respectively, just slightly lower than the concentration in Clovis. In view of the small differences in the water fluoride concentrations between Clovis and the other two communities, the large differences in fluorosis prevalence values suggest that another factor may have influenced the appearance of the teeth in Clovis. Unlike Elmhurst and Galesburg, Clovis is located at a relatively high altitude (4300 feet). As summarized elsewhere in the EPA document under review (pages 39-41), there is evidence from laboratory animal studies and epidemiological studies that residence at high altitude affects amelogenesis in a way that resembles fluorosis and that its effects may be additive to the effects of fluoride exposure. This too adds uncertainty regarding the selection of 2.2 ppm fluoride (Clovis) as the LOAEL for severe dental fluorosis and the appropriateness of the RfD."

He stated that a similar (but weaker) argument could be made for Colorado Springs where the average water fluoride concentration is listed as 2.6 ppm (but with a wide range, see item 3 above). "This city is located at an altitude of 6,035 feet. In addition to these comments and based on the variability among published studies regarding the relationship between estimated fluoride intakes and the risk of severe dental fluorosis, I think consideration should be given to establishing a LBD other than a LBD-1%."

EPA Response: Altitude was evaluated as a contributing factor in the EPA report (Section 3.1.4.2). Compared to non-U.S. high altitude populations (all African populations) which showed an increased prevalence of "fluorosis", the U.S. high altitude populations included in the Dean study did not appear to show the same level of increased "fluorosis" at the higher altitudes represented within the US datasets. It is unknown to what extent inherent racial susceptibility to fluorosis or hypobaric hypoxia contributed to the higher levels of severe "fluorosis" seen in the non-U.S. populations.

As noted by Dr. Whitford, two of Dean's study populations which were high altitude communities (Clovis NM and Colorado Springs, CO) were key populations near the threshold for severe fluorosis. Therefore, to test the weight that these two communities had in determining the threshold, the BMD analysis was rerun allowing for the possibility that some of the cases of severe fluorosis seen in these two communities were due to the high altitude and perhaps not fluoride-related. The prevalence of severe fluorosis was down "adjusted" from 6/404 to 3/404 in Colorado Springs and from 1/138 to 0/138 in Clovis. The resulting log probit BMD for 1% severe fluorosis was 2.43 mg/L and the BMDL 2.24 mg/L. In comparison, the BMD for 1% severe fluorosis with the complete Dean data set was 2.31 mg/L and the BMDL was 2.13 mg/L. The "altitude adjusted" values differ by only 5% from the unadjusted values. Given the uncertainty as to whether altitude was actually a contributory factor, and given the predicted small impact on the BMDL if it were, EPA has chosen to follow its original and slightly more conservative approach to use the unadjusted Dean data set.

The post-peer review dose-response document followed the suggestion of Dr. Whitford and others that a response other than 1% be used for the BMD analysis if it could be justified statistically. The post peer review document uses a 0.5% response level.

9. Are you aware of dose estimates other than those from IOM (1997) and the World Health Organization (WHO; 2002) that are appropriate critical doses for skeletal effects and/or can you suggest a different approach that the OW might use to estimate the fluoride dose associated with skeletal fractures using available data?

Reviewers' Comments:

Dr. Cauley referred to her response to Question 2 when addressing this charge question. That response included references that would address this question. She expressed some concern about the inconsistencies across the age groups suggesting that the significance of the differences in results across ages could have occurred by chance. In addition, she noted that referring to the "radius" (one of the bones of the forearm) as one unit is problematic because the distal radius (the end of the radius closest to the wrist), is primarily trabecular bone whereas the mid radius is predominantly cortical bone. She added that considering the radius as one unit is very problematic if the effects of fluoride differ by bone type. Lastly, she indicated that studies that used sodium fluoride as the fluoride source may have had poor compliance with the prescribed dosing part of the protocol because it causes gastric irritation and is poorly tolerated. Participants in such studies may have had poor compliance with the prescribed treatment régime.

EPA Response: EPA has incorporated the relevant references in their revisions to the peer review draft. The distinction between trabecular bone of the distal radius and cortical bone of the mid-radius is duly noted, and where appropriate, the text of the EPA report has been modified to clarify which portion of the radius was being examined.

Both **Dr. Den Besten** and **Dr. Jackson** noted that they were not aware of additional dose estimates that would be appropriate for determining critical dosage levels and their possible skeletal effects. **Dr. Jackson** added that he could not think of an alternative to the approach taken to estimate fluoride intake associated with skeletal fractures.

EPA Response: No response needed.

Dr. Whitford noted that as in the case of dental fluorosis, the critical doses for skeletal effects will be difficult to establish with a reasonable degree of certainty. The IOM's estimate (page 307) of fluoride exposures that may result in clinical signs of the "milder forms" of skeletal fluorosis (preclinical and perhaps stage I) is 10 mg/day for 10 or more years. There are published exceptions suggesting that higher exposure levels and durations are required as noted on the same page of the IOM report. Further, recent case reports of "tea fluorosis" in the U.S. suggest that, at least for some individuals, a much higher chronic intake is tolerated without progression to stage II skeletal fluorosis (Whyte et al, Am J Med 118: 78-82, 2005; Whyte et al, J Bone Min Res, in press). In the former report the intake was estimated at 37-74 mg F/day from tea throughout the patient's adult life. The intake in the latter report was estimated at more than 40 mg F/day throughout the patient's adult life. Both patients showed marked osteosclerosis but without ligamentous calcifications which was consistent with stage I skeletal fluorosis and neither

patient experienced fractures. Hallanger-Johnson et al (Mayo Clin Proc 82: 719-724, 2007) reported four cases with axial osteosclerosis with elevated serum fluoride levels due to chronic consumption of large amounts of tea.

In addition to the several variables that can affect the quality and quantity of the skeleton cited in the present document, it is of interest that much of the data relating bone fluoride concentrations to the stages of skeletal fluorosis comes from studies of workers in aluminum processing factories. High, chronic exposures to aluminum lead to skeletal changes that share some features in common with skeletal fluorosis which makes it difficult to attribute the skeletal changes only to fluoride. This subject is worthy of further exploration.

EPA Response: The IOM's estimate of fluoride exposure (at least 10 mg/day for 10 or more years) that may result in clinical signs of the "milder forms" of skeletal fluorosis, was based on a review of the available epidemiological studies. As with other chemicals, response to fluoride exposure is likely to vary from individual to individual, and responses in one individual may not be typical for a population as a whole. As noted by Dr. Whitford, IOM does point out that studies in the U.S. have not revealed many cases of skeletal fluorosis even at fluoride drinking water levels up to 9 mg/L.

10. Are you aware of any data that can be used to demonstrate that protection of the secondary teeth from severe dental fluorosis will also protect the primary teeth?

Reviewers' Comments:

Dr. Cauley, Dr. Jackson, and Dr. Whitford said that they are not aware of any additional data that demonstrate that protection of the secondary teeth from severe dental fluorosis also will protect primary teeth. **Dr. Whitford** added that there are a few reports suggesting that dental fluorosis in the primary teeth may correlate with the condition in secondary teeth.

Dr. Den Besten stated that fluorosis in both primary and permanent teeth is caused by ingested fluoride, and there are no data to suggest that primary teeth are more susceptible to fluorosis than permanent teeth. Therefore, a measure that would protect permanent teeth would require limiting ingestion of fluoride. These same measures would protect primary teeth.

EPA Response: Based on the reviewers' comments, it does not appear that there is any evidence that primary teeth are more susceptible to fluoride than permanent teeth.

11. Are there factors other than those discussed in Section 3.1.4 that increase sensitivity to fluorosis of the teeth or bones?

Reviewers' General Comments: All four reviewers suggested additional factors that may have some effect on an individual's sensitivity to fluorosis of the teeth and/or bone including race/ethnicity, pubertal stage, genetics, and amoxicillin use in children during the period of tooth formation.

EPA Response: No response needed.

Specific Comments:

Dr. Whitford noted that some researchers have suggested that African-Americans are more susceptible to dental fluorosis but less susceptible to skeletal fluorosis.

EPA Response: EPA did not identify any data identifying a unique susceptibility of African-Americans for either severe dental fluorosis or skeletal fluorosis.

Dr. Den Besten said that there seem to be individuals who are uniquely sensitive to the effects of fluoride on enamel formation. These individuals may have other more "hidden" enamel defects that are exacerbated by the effects of fluoride. Little is currently known as to why some individuals seem to be more fluoride sensitive.

EPA Response: EPA has added information to the document on genetics as it related to dental fluorosis.

Dr. Jackson cited a study by Hong et al (2004) that appears to indicate that the use of amoxicillin could play a contributing role in the development of primary tooth fluorosis, especially for children exposed to lower levels of fluoride. The full citation was provided.

> Hong L, Levy S, Warren J, Bergus G, Dawson D, Wefel J, Broffitt B. (2004) Primary tooth fluorosis and amoxicillin use during infancy. *J Publ Health Dent*. 64: 38-44.

EPA Response: EPA has added a discussion on the Hong et al. (2004) publication to the dose-response document.

Dr. Jackson also indicated that there have been studies (i.e., Vieira et al., 2005; Yan et al., 2007) in mice that suggest there may be a varying genetic response to identical levels of fluoride ingestion, and these responses have been identified in both tooth and bone formation. He noted that Vieira et al. (2005) found that genetic influences have a direct bearing on the biomechanical properties of the teeth. Furthermore, Yan et al. (2007) found strain-specific effects, like increased osteoclastogenesis, when mice were exposed to physiological level of fluoride. While he was unable to find comparable human trials, he recommended that this area be further explored as the technological means become available. The full citations were provided:

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Vieira A, Hanocock R, Eggertsson H, Everett E, Grynpas M. (2005). Tooth quality in dental fluorosis: genetic and environmental factors. *Calcified Tissue International.* 76:17-25.

Yan D, Gurumurthy A, Wright M, Pfeiler T, Loboa E, Everett E. (2007). Genetic background influences fluoride's effects on osteoclastogenesis. *Bone.* 41:1036-1044.

EPA Response: EPA added data from the Viera et al. and Yan et al. studies to the dose-response document.

Dr. Whitford cited published data that indicate that the susceptibility to dental fluorosis (Everett et al., J Dent Res 81: 794-698, 2002) and the mechanical properties in the bone (Mousny et al,. Bone 39: 1283-1289, 2006) are different among strains of mice. He noted that the differences presumably are due to genetic differences among the strains.

EPA Response: EPA added data from Everett et al, as well as that from a later publication by Mousny on the role of genetics on bone formation and mineralization to the dose-response document.

12. Do you support the OW's conclusion that an RfD of 0.07 mg/kg/day will be protective for severe dental fluorosis in children and skeletal effects in adults while still providing for the beneficial effects of fluoride?
[NOTE: Since completion of the Peer Review, OW has evaluated the contribution of food to total fluoride intake, and has adjusted the oral RfD to 0.08 mg/kg/day to account for the additional fluoride exposure from dietary intake. The dietary contribution to total fluoride exposure at the time of the Dean (1942) study was peer reviewed as part of the exposure and relative source contribution document and included as an appendix in the noncancer dose-response document.

Reviewers' General Comments: In general, **Dr. Cauley, Dr. Jackson,** and **Dr. Whitford** support the use of the 0.07 mg/kg/day RfD. However, all three of these reviewers stated that there is a considerable degree of uncertainty regarding the estimate.

EPA Response: No response needed.

Specific Comments:

Dr. Jackson indicated that he does not think that a claim can be made that no single individual will be completely immune from the development of severe fluorosis even at this recommended RfD. As a result of an individual's potential genetic predisposition, he does not think that the possibility of developing severe fluorosis can be totally ruled out. Additionally, he noted that his concern is not as much about the amount of fluoride that is ingested through the public water supply, but the other well-known sources of fluoride (see his final comment for Question 12 in Appendix B for other sources) that have an additive effect to that derived from consuming fluoridated drinking water.

EPA Response: The Office of Water has developed a companion report to the dose-response document which looks at total fluoride exposures under current conditions. In addition, the results from the Iowa Fluoride study (Hong et al., 2006) that provide support for the conclusions reached by EPA have been added to the report.

Dr. Whitford added that, as estimated by the IOM (1997), the RfD may be closer to 0.10 mg/kg/day for moderate (not severe) dental fluorosis and substantially higher than that for clinically significant skeletal effects in the United States.

EPA Response: See response to Dr. Den Besten's comment below.

Dr. Den Besten did not agree with Dr. Whitford's recommendation because it is based on limiting severe fluorosis to 1% of the population and the IOM's recommended adequate intake level. She suggested that the level be lowered to eliminate severe fluorosis. She noted that one percent of the population represents a relatively large number of individuals, and these individuals are most likely uniquely sensitive to fluoride. She recommended that the data be analyzed without taking the IOM's recommended adequate

levels into account. Then, a secondary analysis could be conducted to include the IOM's recommendations.

EPA Response: The Dean data were evaluated statistically to determine whether they could be used to estimate a fluoride concentration in drinking water associated with a prevalence of severe dental fluorosis less than 1%. It was determined that the data could support an estimate for 0.5%, but not 0.1%; therefore, the 95% confidence limit on the lower bound for 0.5% severe dental fluorosis was used by EPA, instead of 1%. In the development of any RfD, a level of uncertainty ("perhaps spanning an order of magnitude") is acknowledged by EPA due to intrinsic variables which oftentimes can not be quantified. As indicated by the differing opinions expressed by the reviewers, this uncertainty can argue in favor of a higher or a lower RfD. Although genetic factors may make certain individuals more susceptible to fluorosis; there is currently no way to quantify such an increase in susceptibility. As noted by Dr. Jackson, other potential sources of fluoride will contribute to dental fluorosis. The assumption was made by EPA that drinking water was the only source of fluoride in the populations studied by Dean (1942); however, OW later evaluated the potential contribution of dietary items to total fluoride intake during the time of the Dean (1942) study. Using data from the studies of McClure (1943, 1949), an adjustment was made in the derivation of the RfD to account for this additional intake of fluoride through the diet, which was estimated to be 0.01 mg/kg/day. The resulting RfD is of 0.08 mg/kg/day.

The uncertainty associated with the RfD estimate is to some degree conservatively adjusted for by taking the lower bound of the 95% confidence interval for the occurrence of 0.5% severe dental fluorosis (equal to 1.87 mg/L), since it is equally likely that the value could be as high as 2.06 mg/L (upper bound on the 95% CI).

IV. RESPONSE TO DR. WHITFORD'S COMMENTS CONCERNING FLUORIDE ANALYTICAL METHODOLOGY

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16 May 2008

Dr. Whitford (1):
(1) A major weakness of the Dean (1942) report is the chemical method used for the determination of fluoride concentrations in water (Elvove E, 1933). The zirconiumalizarin method is rarely, or probably never, used today because of its relative insensitivity, several interfering substances and lack of specificity for fluoride. In their 1952 report that described improvements to the method, Megregian and Maier (1952) noted that Elvove's original method (1933) had several shortcomings including "non-conformity to the color laws, limited effective fluoride range, and little color change per increment of fluoride." It also appears that Elvove (1933) used the visual method to determine color changes in the zirconium-alizarin reagent (since he referred to "Nessler tubes") which requires subjective judgments and is less accurate than spectrophotometric methods.

EPA Response (1): Elvove employed a reagent containing zirconium oxychloride and alizarin sodium monosulfate. The most recent standard methods employ two reagents related to that discussed in Elvove. One employs an acidic reagent containing zirconyl chloride and the complexing agent SPADNS [sodium 2-(parasulfophenylazo)-1, 8-dihydroxy-3, 6-naphthalene disulfonate] (APHA, AWWA and WEF 2005a). The other is used only in an automated system to form a blue complex, and employs both alizarin and lanthanum nitrate (APHA, AWWA and WEF 2005b). *Standard Methods* clearly indicates the electrode and colorimetric methods are most satisfactory. Hence, while Elvove's reagent may not be used any more, related procedures not only are being used, but have found their way into *Standard Methods*.

The Reviewer implied a lack of specificity. It is very true that more current methods for fluoride ion, employing an ion-selective electrode (APHA, AWWA and WEF 2005c; Omega Engineering, Inc. 2008) and ion chromatography (Hautman et al 1997) will exhibit a range, sensitivity, and specificity that are superior to that described in Elvove. The Reviewer could also argue, correctly, that the range could be improved by replacing the Nessler tubes, which allow color comparison with known standards, with a standard spectrophotometer. However, when Elvove performed his measurements in 1933, the first commercial spectrophotometer, the Beckman DU, was eight years away (Simoni et al 2003). The fluoride-selective electrode was not available until the mid-1960's (Buck and Lindner 2001), while the first commercial ion chromatograph was first marketed in 1981 (Evans 2004).

The only method of comparison that Elvove had at his disposal was colorimetric with Nessler tubes, which permitted him to compare the color of his reacted unknown with that of a known standard. It could be argued that the colorimetric difference between samples containing 0.4 and 0.7 ppm fluoride might be difficult to distinguish; however, there should be no difficulty distinguishing 0.6 and 1.2 ppm, for example. Analytical chemists in those days were usually trained more carefully to distinguish subtle changes in color than chemists of today.

The Reviewer criticized the "non-conformity to the color laws", but did not state what the non-

conformity was. It is true that Elvove did not specifically state that his color reaction obeyed Beer's Law, in which the absorbance of a solution is directly proportional to the concentration of analyte (or derivatized analyte) present. However, such a specification is typically omitted in a professional paper unless the reaction produces a color change that does not obey Beer's Law. In related work, Megregian and Maier (1952) commented "This reagent [modified zirconium-alizarin reagent] is sensitive to small increments of fluoride over a range of 0.0 to 3.0 ppm and it approximates Beer's law over this range." The Reviewer is correct when he commented on the subjective analysis using Nessler tubes, but comparison with Nessler tubes was clearly the standard approach when Elvove published his work.

Dr. Whitford (2):
Megregian and Maier (1952) also reported the effects of interfering substances on the analytical results. Sulfate at 400 ppm in the water increased the fluoride result by 0.1 ppm as did 1.1 ppm hexametaphosphate. Chloride at 1800 ppm, bicarbonate at 400 ppm and iron at 5 ppm decreased the fluoride result by 0.1 ppm. When the water fluoride concentration was 1.0 ppm, 1.0 ppm aluminum reduced the result by 0.39 ppm and 3.0 ppm aluminum reduced the result by 0.63 ppm. When the fluoride concentration was 2.0 ppm, aluminum at 1.0, 2.0 and 3.0 ppm reduced the results by 0.47, 0.86 and 1.13 ppm, respectively.

The report by Elvove (1933) that described the chemical method used to produce the water fluoride concentrations shown in Table I of Dean's 1942 report made no mention of interfering substances. It can be reasonably assumed that most, if not all, of the substances listed in the preceding paragraph were present in all the water samples analyzed but at unreported or unknown concentrations. However, in his original publication Elvove (1933) shows the concentrations of several ions in water samples obtained from 20 different sources. With the exception of Amarillo, these sources were not those shown in the Dean (1942) report. Among the interfering ions listed in the preceding paragraph, sulfate concentrations were more than 400 ppm in two of the 20 water samples; bicarbonate concentrations were more than 400 ppm in five, and aluminum concentrations were more than 1.0 ppm in five. At these concentrations, each of these ions would have affected the apparent water fluoride results.

EPA Response (2): The Reviewer's concerns regarding the presence of several interfering substances and lack of specificity for fluoride are legitimate, and have been investigated extensively in the literature. Elvove's reagent appears to respond primarily to fluoride ion, although the presence of certain inorganic species will suppress fluoride response to some degree. Grutsch et al (1953) noted that modest quantities (< 5 ppm) of either aluminum or iron will suppress, but not eliminate, a response to fluoride ion, while the presence of sub-ppm concentrations of manganese will enhance such a response.

The critical region for community water supply characterization in the Dean (1942) study is that for supplies with fluoride concentrations in the range of 0.9 to 4.5 ppm. USEPA has prepared the following summary table (Table 1) compiling contemporaneous water quality values for chloride, sulfate, aluminum, iron and bicarbonate for those communities from Dean (1942) bounded by this critical fluoride range. In no case was chloride >1800 ppm (range of 0.5 to 689.0 ppm), sulfate or bicarbonate >400 ppm (sulfate range of 3.4 to 308.6 ppm; bicarbonate range of 23.2 to 389.2 ppm), or aluminum >1.0 ppm (range of 0 to 0.3 ppm), or iron >5 ppm (range of 0.01 to 0.1 ppm). Instead, the values for each of these water quality parameters falls below the concentration identified by the Reviewer as cause for concern regarding analytical interference. Chloride ion at these concentrations appears to have no affect upon the determination of fluoride ion.

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Table 1. Water Quality Analysis for Select Towns Used in the Dean (1942) Study						
Town	Fluoride (ppm)	Chloride (ppm)	Sulfate (ppm)	Aluminum (ppm)	Iron (ppm)	Bicarbonate (ppm)
Kewanee, IL[a]	0.9	689.0	308.6	0	0.01	300.1
Galesburg IL[a]	1.9	190.5	351.7	0	0.1	295.2
Clovis. NM[b]	2.2	16.5	24.2	0.2	0.02	234.9
Colorado Springs, CO[a]	2.6	0.5	4.9	0	0.04	23.2
Plainview, TX[c]	2.9	31.8	32.5	0.3	0.1	331.8
Amarillo, TX[c]	3.9[d]	13.5	52.0	0.3	0.08	389.2
Conway, SC[c]	4.0[d]	49.5	3.4	0.06	0.06	256.0
Lubbock, TX	4.4[d]	111.8	255.0	0.2	0.1	342.8

[a]Dean, 1942.
[b]Dean, 1937.
[c]Dean and Elvove, 1936.
[d]Fluoride value as reported in Dean, 1942.

Evaluation of the literature includes experimental tests by Grutsch et al (1953) with varying concentrations of Fe (III) in solution with 0.5 and 1.00 ppm fluoride. There was no suppression of fluoride at these concentrations when the Fe (III) concentration was 0.1 ppm. When the concentration of Fe (III) reached 0.5 ppm, the "found" values for 0.50 and 1.00 ppm fluoride are 0.48 and 0.96 ppm, respectively, or an approximate 4% suppression for the measured concentration of fluoride. Since the community water supplies of interest (Table 1) all exhibit Fe concentrations $\leq$ 0.1 ppm, no suppression of actual fluoride concentrations is expected from the concentrations of Fe present.

Grutsch et al (1953) also evaluated Al (III) in water with the same two target concentrations of fluoride and observed no suppression at either fluoride concentration when the Al (III) concentration was 0.1 ppm. When 0.3 ppm Al (III) was present, the "found" values for 0.50 and 1.00 ppm fluoride were 0.48 and 0.98 ppm, respectively, or a suppression of 4% at the low-concentration (0.5 ppm F) sample and 2% at the 1 ppm F sample. Four out of the 8 community water supplies summarized in Table 1 exhibit Al concentrations <0.1 ppm, while two exhibit Al concentrations of 0.2 ppm and two exhibit Al concentrations of 0.3 ppm. No suppression of actual fluoride concentrations is expected in water samples from communities with Al concentrations <0.1 ppm. The data of Grutsch et al (1953) indicate that the maximal suppression of fluoride expected at 0.3 ppm Al would be 4%.

Dr. Whitford (3):
Another indication of the problem with the accuracy of the Elvove method is found in the footnote to Table 3-1 on page 24 of the EPA Dose-Response Analysis where it is said (quoting Elvove who was the principal chemist) that "as little as 0.01 mg F/50cc, or 0.2 ppm F, could be differentiated from the control by application of this technique." This appears to mean that Elvove's method could differentiate between water without fluoride and water containing 0.2 ppm fluoride. The magnitude of the error at higher concentrations is not known to me. The scatter in the analytical results seen in the 1933/34 monthly results for water in Colorado Springs is of particular interest (see Table 4 in Dean and Elvove, 1935). The average of the 12 results was 2.5 ppm but the range was 1.8 to 3.0 ppm despite the fact that the water came from a single source. While some seasonal variation in water concentrations can expected, this

wide range (1.2 ppm) appears excessive. Further, the 12 monthly results from Monmouth ranged from 1.6 to 1.9 ppm, those from Galesburg ranged from 1.8 to 2.0 ppm, and those from Pueblo ranged from 0.3 to 0.7 ppm.

EPA Response (3): It has previously been noted by Megregian and Maier (1952) that the use of this reagent [modified zirconium-alizarin reagent] is sensitive to small increments of fluoride over a range of 0.0 to 3.0 ppm and approximates Beer's law over this range, which includes the range of results for Colorado Springs (fluoride content of 1.8 to 3.0 ppm from Dean and Elvove 1935). There are a number of alternate sources of variability for natural fluoride concentrations in community water supplies of Colorado Springs, such as snowfall, rainfall, reservoir maintenance and pumping schedules, etc., and it should thus not be assumed that the range of 1.8 to 3.0 ppm F noted by the Reviewer in Dean and Elvove (1935) was a consequence of some analytical difficulty. It is also noted that reported fluoride concentrations for Colorado Springs during the 14 months observed between Jan 1940 and Feb 1941 in Dean (1942) exhibited the narrow range of 2.4–2.8 ppm F (mean of 2.55 ppm F). It appears that the source of variability displayed in the Dean and Elvove (1935) data set for Colorado Springs had been resolved by the time of the Dean (1942) data collection and analysis.

The critical range for F determinations in the current assessment is <4.5 ppm F (see Table 1), a range over which the analytical method and reagents of Elvove appear to be appropriate, adequate and customary for the time.

REFERENCES CITED

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American Public Health Association (APHA), American Water Works Association (AWWA) and Water Environment Federation (WEF), 2005b. Method 4500 F- E. "Complexone Method." *Standard Methods for the Examination of Water & Wastewater, 21st Edition.* Pp. 4-87.

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V. EXTERNAL PEER REVIEW REPORT

External Peer Review of U.S. EPA's Draft Dose-Response Assessment for Severe Dental Fluorosis and the Risk for Increased Bone Fractures Related to Fluoride

Peer Review Report

Prepared for:
U.S. Environmental Protection Agency
Office of Water
Office of Science and Technology

Prepared by:
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EPA Contract No. PR-CI-06-10561
Work Assignment Number B-02

April 2009

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Introduction

On April 2, 1986, the United States Environmental Protection Agency (U.S. EPA) set a revised Maximum Contaminant Level (MCL) for fluoride at 4 mg/L to protect against crippling skeletal fluorosis, an adverse health effect. In addition, U.S. EPA set a non-enforceable Secondary Maximum Contaminant Level (SMCL) at 2 mg/L to protect against objectionable dental fluorosis. In 2003, U.S. EPA requested that the National Research Council (NRC) review the scientific and technical basis for U.S. EPA's drinking water MCL and SMCL for fluoride as a part of its review of all regulations promulgated prior to the 1996 reauthorization of the Safe Drinking Water Act. NRC was asked to:

- Review new toxicology, epidemiology, and clinical data;
- Examine exposure data on orally ingested fluoride from water and other sources;
- Advise U.S. EPA on the adequacy of its MCL and SMCL to protect children and others from adverse effects; and
- Identify data gaps and make research recommendations.

In 2006, the National Research Council (NRC) panel recommended that U.S. EPA conduct a new quantitative risk assessment for severe dental fluorosis and the risk for increased bone fractures related to fluoride based on the most recent dose-response data. The findings of the 2006 NRC panel diverged from those of the previous 1993 panel by categorizing severe dental fluorosis, which results in thinning and pitting of the tooth enamel, as an adverse health effect rather than a cosmetic effect. The 2006 panel also concluded that the present MCL of 4 mg/L was not protective for severe dental fluorosis and may not be protective for the skeletal fracture endpoint. The NRC committee did not evaluate health effects from lack of exposure to fluoride or fluoride's efficacy in preventing dental cavities.

In response to the NRC 2006 report, the Office of Water (OW) has completed a dose-response assessment based on data from studies of severe dental fluorosis and evaluated the available dose-response data on the skeletal effects of fluoride as recommended by NRC. The report provides a quantitative estimate of a lower bound confidence interval on the concentration in water associated with a 1 percent risk for severe dental fluorosis derived from data collected before fluoridation of drinking water and the introduction of fluoride into dental products. The concentration-response data were then converted to dose using water intake and body weight data for children during the period for formation of the secondary teeth. The relative source contribution for drinking water compared to total fluoride exposure will be the subject of a separate effort by the OW.

Prior to releasing the document for external peer review, it was evaluated within U.S. EPA by representatives from the OW, Office of Children's Health, Office of Pesticide Programs, and Office of Research and Development.

Peer Review Meeting

On March 11, 2008, U.S. EPA convened a panel of external and independent technical experts in Washington, D.C. to address charge questions regarding the quality, clarity, and transparency of the draft dose-response report. The names and affiliations of the four peer reviewers for this draft report can be found in Exhibit 1, and their areas of expertise are shown in Exhibit 2. The peer reviewers were asked to draft initial responses to the charge questions, attend a one-day meeting in Washington, D.C., and submit final responses to the charge questions and any additional comments on the dose-response document following the meeting.

Exhibit 1: Peer Reviewer Names and Affiliations

Reviewer Name	Reviewer Affiliation
Jane A. Cauley, Dr.P.H	Professor, Department of Epidemiology, University of Pittsburgh
Pamela DenBesten, D.D.S., M.S.	Professor and Chair, Division of Pediatric Dentistry, Department of Orofacial Sciences, University of California at San Francisco
Richard D. Jackson, D.M.D.	Assistant Professor, Preventive and Community Dentistry, School of Dentistry, Indiana University, Oral Health Research Institute
Gary M. Whitford, D.M.D., Ph.D.	Regents' Professor, Department of Oral Biology and Maxillofacial Pathology, Medical College of Georgia

Exhibit 2: Peer Reviewer Expertise Relative to the Selection Criteria

	Dr. Cauley	Dr. DenBesten	Dr. Jackson	Dr. Whitford
Dentist or researcher with experience relating to dental pitting as a result of fluoride and the associated potential for tooth decay		X	X	X
Doctor or researcher with knowledge of the link between fluoride and bone fractures	X			X
Epidemiologist with familiarity of the health risks from environmental fluoride exposure	X			X

The technical experts that comprised the peer reviewer panel were selected by ICF International based upon their independence and expertise in the subject matter. The peer reviewers' qualifications were assessed based on the following selection criteria specified by EPA: (1) dentists or researchers with experience relating to dental pitting as a result of fluoride and the associated potential for tooth decay; (2) doctors or researchers with knowledge of the link between fluoride and bone fractures; and/or (3) epidemiologists with familiarity of the health risks from environmental fluoride exposure.

The peer reviewers' draft of initial responses to the charge questions are provided in Appendix A, and their final responses to the charge questions and any additional comments on the dose-response document are provided in Appendix B. (Comments are organized alphabetically by reviewer in both appendices.) Additionally, final comments from the four peer reviewers are organized and summarized in the main body of this document according to the charge questions. In most cases, the comments are presented in alphabetical order according to the peer reviewer's last name. If there is agreement among reviewers, this is stated at the beginning of the summary for that question. Except for minor clarifications, the comment summary retains most of the reviewers' original wording. The use of quotation marks is reserved for situations in which the reviewer quoted a specific document or the reviewer's original wording and/or intent was unclear.

Comment Summary

The overall response from the reviewers was positive; however, all four reviewers suggested changes to improve the overall quality of the document. Their responses to each charge question are summarized in the remainder of this document.

Charge Question 1. Was the document clear and transparent in its presentation of data and explanation of the analytical approaches used to characterize the concentration-response and dose-response relationships for severe dental fluorosis? If not, do you have any suggestions that will enable the authors to improve its clarity?

In general, the reviewers thought that the document was clear and transparent. In addition, the reviewers commented that the document clearly described the available literature and presented the information in an understandable format. In particular, **Dr. Jackson** noted that the individual tables summarizing the studies were very helpful.

However, the reviewers provided the following suggestions to improve the clarity of the document:

- **Dr. Cauley** believes that the document could be strengthened by reorganizing it as a manuscript. When reorganizing the document as a manuscript, the following changes should be considered: the introduction of the document should provide background information and a scientific rationale for this analysis; the specific objectives should be listed; and the methods for identifying all the literature should be transparent and consistently applied to both the fluorosis and the fracture sections.

- **Dr. Cauley** also believes that it would also be helpful if the document included a discussion on U.S. EPA OW's and NRC's roles (e.g., How will each agency find this information helpful? What prompted this report?).
- **Dr. DenBesten** suggested several specific changes by page number (see Dr. DenBesten's final comments for Question 1 in Appendix B).
- **Dr. Jackson** noted that he would have preferred that the studies be arranged chronologically as opposed to alphabetically by author.

Charge Question 2. Are you aware of any significant publications related to severe dental fluorosis or the skeletal effects of fluoride that are not included in the noncancer assessment document?

Three of the reviewers, **Dr. Cauley, Dr. Jackson,** and **Dr. Whitford**, thought that there were additional publications related to severe dental fluorosis and/or skeletal effects of fluoride that should be reviewed for possible inclusion.

Dr. Cauley noted that while the dental fluorosis literature review in the report covered a wide range of fluoride levels, the skeletal/ fracture studies were limited to those with fluoride levels $\geq$ 4 mg/L. She suggested that the Office of Water consider the following publications that discuss the occurrence of fractures at lower levels of fluoride:

- Cauley JA, Murphy PA, Riley TJ, Buhari AM. (1995). Effects of fluoridated drinking water on bone mass and fractures: The study of osteoporotic fractures. *J Bone Miner Res.* 10(7):1076-1086.
- Phipps KR, Orwoll ES, Mason JD, Cauley JA. (2000). Community water fluoridation, bone mineral density, and fractures: prospective study of effects in older women. *BMJ* 321(7265):860-864.
- Phipps KR, Burt BA. (1990). Water-borne fluoride and cortical bone mass: A comparison of two communities. *J Dent ResT.* 69(6):1256-1260. [**Note**: this study compares bone mineral density in Deming, NM (0.7 mg/L) and Lordsburg, NM (3.5 mg/L)]

Dr. Cauley also expressed concern that the bone fractures section is less thorough than the dental fluorosis section. To address this issue, she suggested the following publications be reviewed for possible inclusion and noted that the studies by K. Phipps, C. Cooper, S. Jacobsen, and T. Hillier are particularly noteworthy and of high quality:

- Demos LL, Kazda H, Cicuttini FM, Sinclair MI, Fairley CK. (2000). Water fluoridation, osteoporosis, fractures--recent developments. *Aust Dent J.* 46(2):80-87.
- Fabiani L, Leoni V, Vitali M. (1999). Bone-fracture incidence rate in two Italian regions with different fluoride concentration levels in drinking water. *J Trace Elem Med Biol.* 13(4):232-237.
- Kleerekoper M. (1996). Fluoride and the skeleton. *Crit Rev Clin Lab Sci.* 33(2):139-161.
- Raheb J. (1995). Water fluoridation, bone density and hip fractures: a review of recent literature. *Community Dent Oral Epidemiol.* 23(5):309-316.
- Søgaard CH, Mosekilde L, Schwartz W, Leidig G, Minne HW, Ziegler R. (1995). Effects of fluoride on rat vertebral body biomechanical competence and bone mass. *Bone.* 16(1):163-169.

- Kröger H, Alhava E, Honkanen R, Tuppurainen M, Saarikoski S. (1994). The effect of fluoridated drinking water on axial bone mineral density—a population-based study. *Bone Miner.* 27(1):33-41.
- Ripa LW. (1993). A half-century of community water fluoridation in the United States: review and commentary. *J Public Health Dent.* 53(1):17-44.
- Gordon SL, Corbin SB. (1992). Summary of workshop on drinking water fluoride influence on hip fracture on bone health. (National Institutes of Health, 10 April, 1991) *Osteoporos Int.* 2(3):109-117.
- Colquhoun J. (1991). Water fluoride and fractures. *N Z Med J.* 104(917):343.
- McNeill KG, Coote GE, Hitchman AJ. (1991). Uptake of fluorine in cortical and trabecular bone. *J Bone Miner Res.* 6(8):859-864.
- Arnala I, Alhava EM, Kivivuori R, Kauranen P. (1986). Hip fracture incidence not affected by fluoridation. Osteofluorosis studied in Finland. *Acta Orthop Scand.* 57(4):344-348.
- Ericsson Y, Luoma H, Ekberg O. (1986). Effects of calcium, fluoride and magnesium supplementations on tissue mineralization in calcium- and magnesium-deficient rats. *J Nutr.* 116(6):1018-1027.
- Simonen O, Laitinen O. (1985). Does fluoridation of drinking-water prevent bone fragility and osteoporosis? *Lancet.* 2(8452):432-434.
- Madans J, Kleinman JC, Cornoni-Huntley J. (1983). The relationship between hip fracture and water fluoridation: An analysis of national data. *Am J Public Health.* 73(3):296-298.
- Schamschula RG, Barmes DE. (1981). Fluoride and health: dental caries, osteoporosis, and cardiovascular disease. *Annu Rev Nutr.* 1:427-435.
- Alhava EM, Olkkonen H, Kauranen P, Kari T. (1980). The effect of drinking water fluoridation on the fluoride content, strength and mineral density of human bone. *Acta Orthop Scand.* 51(3):413-420.
- Stein ID, Granik G. (1980). Human vertebral bone: relation of strength, porosity, and mineralization to fluoride content. *Calcif Tissue Int.* 32(3):189-194.
- Hegsted DM. (1967). Osteoporosis and fluoride deficiency. *Postgrad Med.* 41(1):A49-53.
- Bernstein DS, Sadowsky N, Hegsted DM, Guri CD, Stare FJ. (1966). Prevalence of osteoporosis in high- and low-fluoride areas in North Dakota. *JAMA.* 198(5):499-504.
- Hillier S, Cooper C, Kellingray S, Russell G, Hughes H, Coggon D. (2000). Fluoride in drinking water and risk of hip fracture in the UK: a case-control study. *Lancet.* 355(9200):265-269.
- Hillier S, Inskip H, Coggon D, Cooper C. (1996). Water fluoridation and osteoporotic fracture. *Community Dent Health.* 13 Suppl 2:63-68.
- Jacobsen SJ, Goldberg J, Cooper C, Lockwood SA. (1992). The association between water fluoridation and hip fracture among white women and men aged 65 years and older. A national ecologic study. *Ann Epidemiol.* 2(5):617-626.
- Cooper C, Wickham C, Lacey RF, Barker DJ. (1990). Water fluoride concentration and fracture of the proximal femur. *J Epidemiol Community Health.* 44(1):17-19.
- Phipps KR, Orwoll ES, Mason JD, Cauley JA. (2000). Community water fluoridation, bone mineral density, and fractures: prospective study of effects in older women. *BMJ.* 321(7265):860-864.

- Phipps KR, Orwoll ES, Bevan L. (1998). The association between water-borne fluoride and bone mineral density in older adults. *J Dent Res.* 77(9):1739-1748.
- Phipps K. (1995). Fluoride and bone health. *J Public Health Dent.* 55(1):53-56.
- Phipps KR, Burt BA. (1990). Water-borne fluoride and cortical bone mass: a comparison of two communities. *J Dent Res.* 69(6):1256-1260.

Dr. Jackson noted that the document should consider the findings from a study that examined 357 school-age children in 1994 from three Indiana communities with varying levels of fluoride in the water (0.2, 1.0, and 4.0 ppm). Severe fluorosis, as determined by the Tooth Surface Index of Fluorosis (TSIF) (score 5, 6, or 7), was observed in 9% of the children examined in the 4.0-ppm community. No scores of this magnitude were seen in the other two communities. A comparison using 1992 prevalence data for the same communities was conducted, and results showed an increase in the prevalence of fluorosis in each community, primarily for TSIF scores 1, 2, or 3. The following citation for this study was provided:

- Jackson R, Kelly S, Katz B, Brizendine E, Stookey G. (1999). Dental fluorosis in children residing in communities with different fluoride levels in the water: 33 month follow-up. *Pediatric Dentistry.* 21:248-254.

Dr. Whitford suggested that the committee review a copy of the book entitled "Fluoride Drinking Waters." He stated that this book is a compilation of many of the early papers that address several aspects of fluoride in water. The book was published in 1962, edited by Frank J. McClure, and is Public Health Service Publication No. 825. He highlighted the following three references from the book that contain information pertinent to the reliability of the water fluoride concentrations shown in the Dean (1942) study, the study selected as the "critical study for severe dental fluorosis:"

- Elvove E. (1933). Estimation of fluorides in waters. *Pub. Hlth. Rep.* 48:1219-1222.
- Dean HT, Elvove E. (1935). Studies on the minimal threshold of the dental sign of chronic endemic fluorosis (Mottled enamel). *Pub. Hlth. Rep.* 50:1719-1729.
- Megregian S, Maier FJ. (1952). Modified zirconium alizarin reagent for determination of fluoride in water. *J. Am. Water Works Assn.* 44:239-246.

Charge Question 3. Are the strengths and weaknesses of Dean (1942) study (selected as critical) fully characterized?

<u>Strengths</u>:

In general, the reviewers believe that the strengths of the Dean (1942) study were well-characterized in the draft dose-response report.

Dr. Cauley noted that additional strengths of the study include: (1) the wide range of fluoride concentrations, although there were fewer subjects in the high fluoride categories; (2) the dose-response relationship illustrated by the data showing increasing risk of severe skeletal fluorosis with increasing fluoride; and (3) the consistency of the findings across several different communities.

Dr. Jackson doubted that anyone would disagree that the Dean (1942) study has been and will continue to be a benchmark study in the dental literature, as well as the much broader literature related to public health and epidemiology. He noted that Dean's study was performed when confounding fluoride sources were not available and, thus, probably gives a very clear picture of the prevalence of the relationship of fluoride ingestion and the subsequent development of dental fluorosis.

Weaknesses:

The reviewers expressed some concern about the characterization of the weaknesses of the study.

Dr. Cauley and **Dr. Jackson** noted that the Dean (1942) study does not provide information on race/ethnicity, and given the probable characteristics of the region, the population of subjects examined was likely not diverse in terms of racial or cultural characteristics.

Dr. Cauley noted that the data were collected the late 1930-40s. Although confounding by use of other fluoride products would be minimal, there are many other cohort differences between children exposed to fluoride in the late 1930-40s as compared to now. For example, dental hygiene, dietary intakes (e.g., less water and more carbonated beverages), and body weight are very different in today's children compared to those in the 1930s. Additionally, **Dr. Cauley** questioned whether puberty and/or hormonal changes may influence fluoride effects, which may be important because age of menarche has been decreasing. Another weakness of the studies reviewed is that there appears to be no information on exposure duration (e.g., How long did these children live in each community? Did the inclusion criteria include a minimum time of residence?).

Dr. Jackson further noted that it has been postulated that genetic factors may impact the expression of dental fluorosis at identical levels of ingestion. The fact that data were collected in what may have been an exclusively white population appears to limit its applicability for use as a benchmark.

Dr. Whitford believes that the weaknesses of the study were not necessarily fully characterized. One example given in his discussion of how the weaknesses of the Dean (1942) study were not fully characterized, was the lack of review of the publications presented in his response to Question #2 (Elvove, 1933). He also had the following specific comments:

1. A major weakness of the Dean (1942) report is the chemical method used for the determination of fluoride concentrations in water (Elvove, 1933). The zirconium-alizarin method is rarely, or probably never, used today because of its relative insensitivity, several interfering substances, and lack of specificity for fluoride. Please see **Dr. Whitford's** final comment for Question 3 in Appendix B for a critical review of the Elvove (1933) method.
2. The study population in the Dean (1942) may not have been continuously exposed to the community's communal water supply. Dean (1942, page 25) listed two major requisites for quantitative evaluation of the dental effects of ingesting water containing fluoride. One of these requisites was "a population continuously exposed throughout life to the variable under investigation (the communal water supply)." For a more thorough

discussion of this issue, please see **Dr. Whitford's** final comment for Question 3 in Appendix B. **Dr. Whitford** recommended that the original papers summarized in Dean (1942) be examined to determine the extent to which the children met the requisite cited above and that the information be included in the Dose-Response Analysis document. If such information is not available, then the document should note this and discuss the implications in its conclusions.

Dr. Whitford further commented that the appropriateness of the LOAEL (2.2 ppm) and the calculated reference dose (RfD) (0.07 mg F/kg bw/day) reported in the OW's draft dose-response report are based largely on the accuracy of the water fluoride concentrations shown in Dean (1942), as well as on several other variables that may have affected the outcomes of the epidemiological studies. He indicated that his preceding comments draw attention to several shortcomings of the chemical method used and other limiting aspects of the studies summarized by Dean (1942) and recommended that the uncertainties associated with these factors be discussed wherever appropriate and certainly in the "Uncertainty Factors" section.

Charge Question 4. Are some teeth more susceptible to severe fluorosis than others? If so, should the OW make modifications to the age range identified as the period of concern?

The reviewers had differing opinions on whether some (and which) teeth are more susceptible to severe fluorosis.

Dr. Cauley thought age may be a factor in the susceptibility to severe fluorosis and suggested that a more direct discussion of the age at risk is needed and that this topic should be highlighted in a separate section. She suggested that, in particular, a table summarizing the ages of the children in each study and the age range which appeared to be at highest risk would be helpful. She recommended that the writers add this to Table 3-16.

Dr. DenBesten thought that the report was clear in showing that posterior teeth also are susceptible, and the case for including children up to age 14 was clear and compelling.

Neither **Dr. Jackson** nor **Dr. Whitford** has come across any literature that indicates that some teeth are more susceptible to the development of severe dental fluorosis. **Dr. Jackson** believes that the susceptibility is the same assuming that the exposure is constant and taking into consideration how long the developing tooth is exposed to higher levels of fluoride. He stated that, for example, maxillary third molars may present greater evidence of severe fluorosis (pitting, staining, etc) because they take longer to develop (12-16 years-of-age) and erupt as opposed to a maxillary central incisor (5 years-of-age). **Dr. Whitford** hypothesized that the posterior teeth may be more susceptible since their development is more protracted than that of the anterior teeth.

Charge Question 5. Are the data on cavities as collected and graphically presented by the OW consistent with the hypothesis that there is an increased risk for cavities in susceptible individuals with severe dental fluorosis?

Dr. Cauley, Dr. DenBesten, and **Dr. Jackson** agreed that the data on cavities presented in the report are consistent with the stated hypothesis.

Dr. Cauley noted that the data summarized in Table 3-40 to 3-44 are convincing that dental caries are more common in subjects with severe fluorosis. The data suggest a U-shaped relationship with fluoride: a higher risk in those with very low intakes and intakes >2-3 mg/L.

Dr. Jackson noted that the OW may want to consider examiner bias in the methodology of these studies. Although it would be impossible to eliminate this bias, it is something that should at least be mentioned as a possible confounder. He also described the findings of the following study (see Appendix B for a description of the study findings):

- Jackson R, Kelly S, Katz B, Hull J, Stookey G. (1995). Dental fluorosis and caries prevalence in children residing in communities with different levels of fluoride in the water. *J Publ Health Dent.* 88:79-84.

Dr. Whitford disagreed with the other reviewers, noting that the available data present an unclear relationship. He discussed conflicting results from several studies in his final comments in Appendix B. He noted that overall, and as summarized in Figure 3-7 on page 71, the relationship between the severity of dental fluorosis and the risk of caries is suggestive, but not convincing. He believes that this subject requires more study with control for variables that are known risk factors for caries before a reasonably firm conclusion can be drawn about the possibility of an association between severe dental fluorosis and an increased risk of caries.

Charge Question 6. Are there recent data that would impact the Institute of Medicine (IOM; 1997) Adequate Intake value of 0.05 mg/kg/day for fluoride that the OW should consider in this assessment?

Dr. Cauley, Dr. Jackson, and Dr. Whitford were unaware of any recent data that could influence the adequate intake (AI) value.

Dr. Cauley suggested that the writers mention life stage and the upper limit of toxicity. She pointed out that the American Dental Association website may have updated their fluoride document in 2006. In her final comment for Question 6 (see Appendix B), she provided two tables: Criteria and Dietary Reference Intake Values for Fluoride by Life Stage Group and Tolerable Upper Intake Levels by Life Stage Group.

Although **Dr. Whitford** did not know of any additional data, he commented on the interpretation of the IOM's values. He noted that the IOM's AI value represents the amount of intake of any substance "needed to maintain a defined nutritional state or criterion of adequacy in essentially all members of a specific healthy population." In the case of fluoride, the AI (see page 301, Dietary Reference Intakes, 1997) "is based on estimated intakes that have been shown to reduce the occurrence of dental caries maximally in a population without causing unwanted side effects

including moderate dental fluorosis." He noted that this does not mean that intakes somewhat higher than 0.05 mg/kg/day increase risk of moderate dental fluorosis. In fact, the IOM's estimate for the threshold for that risk is 0.10 mg/kg/day.

Dr. DenBesten disagreed with the presumption that the IOM's value is sufficient. The weight of evidence indicates that the primary mechanism by which fluoride protects against tooth decay is a topical effect. Therefore, she noted that the IOM's recommendation of an adequate intake value, at least relating to tooth decay, should be reassessed.

Charge Question 7. Can you suggest an approach to transform the water concentration data from the Dean (1942) study to units of dose for the population susceptible to severe dental fluorosis other than that used by the OW?

Dr. Cauley suggested that writers use more generalizable data from the National Health and Nutrition Examination Survey (NHANES) for body weight. She noted that if this approach were used, the writers could still utilize the water intake estimates from the Erchow and Cantor (1989) paper.

Dr. DenBesten and Dr. Jackson noted several difficulties in developing an approach for transforming the water concentration data from the Dean study to units of dose for the population susceptible to severe dental fluorosis.

Dr. DenBesten stated that serum fluoride levels would be the most useful measurement, but these levels are not available. She believes that using Dean's data as a starting point to quantify total ingestion is the best option. She recommended that future studies include random sampling of serum fluoride levels to strengthen future decision-making relative to fluoride intake.

Dr. Jackson noted that the U.S. marketplace is constantly changing and ingestion of tap water continues to decline. U.S. EPA's (2004) report stated that bottled water accounted for only 13% of water consumption in the United States. More recent trade manufacturing data indicates that bottled water consumption in the United States exceeds this percentage by a wide margin, and bottled water consumption may surpass tap water consumption in the near future. Other published data suggest that among Hispanic individuals, tap water is commonly perceived as "unhealthy" and again bottled water is almost exclusively consumed. Another point that should be further explored is the possible "halo effect" of imported foods and beverages into the United States and the fluoride content of these consumables.

Charge Question 8. Can you provide input concerning the strengths and weaknesses of the approach utilized by the OW to identify a lower bound dose for severe dental fluorosis?

Dr. Cauley and **Dr. DenBesten** both listed strengths and weaknesses to the approach utilized by OW to identify a lower bound dose for severe dental fluorosis.

Strengths:

Dr. Cauley said that the main strengths of the approach were that: (1) conservative estimates were carried out with and without the outlier community; (2) several different models were considered; and (3) sensitivity analysis showed that even eliminating three highest data points had little effect on the model goodness of fit or on the BMDL.

Dr. DenBesten added that: (1) the data used were limited to studies found in the Dean report where the only exposure to fluoride was drinking water; and (2) a careful assessment was done regarding tap water consumed and mean body weights.

Weaknesses:

Dr. Cauley said that the inherent weakness of the approach relates primarily to the weaknesses previously cited related to the parent Dean (1942) study. The writers may consider adding a formal test for trend in data.

Dr. DenBesten added that the assumption that 0.05 mg/kg/day is a required amount of fluoride and that dose estimates must be above this level could be considered a weakness because the purpose of U.S. EPA's analysis was to determine risk, not to conduct risk benefit analyses. In addition, she noted that the assumption that the small number of children who displayed severe fluorosis were those who had excess exposure to fluoride could also be considered a weakness. This assumes that these children drank significantly more water since water was the only source of fluoride. It is more likely that genetic or other causes are responsible for this small outlier group.

Dr. Whitford expressed additional concerns about the approach utilized by the OW, particularly in reference to the determination of the LOAEL. He proposed that the variability among published studies regarding the relationship between estimated fluoride intakes and the risk of severe dental fluorosis may be due to the relative altitude of the cities that were studied. He provided a detailed discussion of this issue in his final comment for Question 8 (see Appendix B). He suggested that based on the variability among published studies regarding the relationship between estimated fluoride intakes and the risk of severe dental fluorosis, consideration should be given to establishing a lower bound dose other than 1 percent.

Charge Question 9. Are you aware of dose estimates other than those from IOM (1997) and the World Health Organization (WHO; 2002) that are appropriate critical doses for skeletal effects and/or can you suggest a different approach that the OW might use to estimate the fluoride dose associated with skeletal fractures using available data?

Dr. Cauley referred to her response to Question 2, which included additional references that would address this question. She expressed some concern about the inconsistencies across the age groups. The significance of the differences in results across age could have occurred by chance. In addition, she noted that referring to the "radius" (one of the bones of the forearm) as a whole is problematic because if it is the distal radius (the end of the radius closest to the wrist), it is primarily trabecular bone mineral density (BMD), whereas the mid radius is predominantly

cortical BMD. She added that it is very problematic if the effects of fluoride differ by bone type. Lastly, she indicated that in the instance of randomized trials, the compliance/adherence to the Na F is a limitation because it is poorly tolerated. She questioned whether they adhered to the intent to treat principle.

Both **Dr. DenBesten** and **Dr. Jackson** noted that they were not aware of additional dose estimates that would be appropriate for determining critical dosage levels and their possible skeletal effects. **Dr. Jackson** added that he could not think of an alternative to the approach taken to estimate fluoride intake associated with skeletal fractures.

Dr. Whitford noted that as in the case of dental fluorosis, the critical doses for skeletal effects are difficult to establish with a reasonable degree of certainty. The IOM's estimate (page 307) of fluoride exposures that may result in clinical signs of the "milder forms" of skeletal fluorosis (preclinical and perhaps stage I) is 10 mg/day for 10 or more years. There are published exceptions suggesting that higher exposure levels and durations are required; these are noted on the same page of the IOM report. In addition, chronic consumption of large amounts of tea and chronic exposure to aluminum can alter the skeletal response to fluoride. **Dr. Whitford** included a discussion of recent case reports of "tea fluorosis" in the U.S. in his final comments (see Appendix B). In addition, he stated that much of the data relating bone fluoride concentrations to the stages of skeletal fluorosis comes from studies of workers in aluminum processing factories. High, chronic exposures to aluminum result in skeletal changes that share some features in common with skeletal fluorosis; this makes it difficult to attribute the skeletal changes only to fluoride. He believes that this subject is worthy of further exploration.

Charge Question 10. Are you aware of any data that can be used to demonstrate that protection of the secondary teeth from severe dental fluorosis will also protect the primary teeth?

Dr. Cauley, Dr. Jackson, and Dr. Whitford said that they are not aware of any additional data that demonstrate that protection of the secondary teeth from severe dental fluorosis also will protect primary teeth. **Dr. Whitford** added that there are a few reports suggesting that dental fluorosis in the primary teeth may correlate with the condition in secondary teeth.

Dr. DenBesten stated that fluorosis in both primary and permanent teeth is caused by ingested fluoride, and there is no data to suggest that primary teeth are more susceptible to fluorosis than permanent teeth. Therefore, a measure that would protect permanent teeth would require limiting ingestion of fluoride. These same measures would protect primary teeth.

Charge Question 11. Are there factors other than those discussed in Section 3.1.4 that increase sensitivity to fluoroisis of the teeth or bones?

All four reviewers suggested additional factors that may have some effect on an individual's sensitivity to fluorosis of the teeth and/or bones.

Dr. Cauley questioned whether race/ethnicity and pubertal stage are important. Likewise, **Dr. Whitford** noted that some researchers have suggested that African-Americans are more susceptible to dental fluorosis but less susceptible to skeletal fluorosis.

Dr. DenBesten said that there seem to be individuals who are uniquely sensitive to the effects of fluoride on enamel formation. These individuals may have other more "hidden" enamel defects that are exacerbated by the effects of fluoride. Little is currently known as to why some individuals seem to be more fluoride sensitive.

In addition, **Dr. Jackson** cited a study by Hong et al (2004) that appears to indicate that the use of amoxicillin could play a contributing role in the development of primary tooth fluorosis, especially for children exposed to lower levels of fluoride. The full citation is:

- Hong L, Levy S, Warren J, Bergus G, Dawson D, Wefel J, Broffitt B. (2004) Primary tooth fluorosis and amoxicillin use during infancy. *J Publ Health Dent*. 64: 38-44.

Dr. Jackson also indicated that there have been studies (i.e., Vieira et al., 2005; Yan et al., 2007) in mice that suggest there may be a varying genetic response to identical levels of fluoride ingestion, and these responses have been identified in both tooth and bone formation. He noted that Vieira et al. (2005) found that genetic influences have a direct bearing on the biomechanical properties of the teeth. Furthermore, Yan et al. (2007) found strain-specific effects, like increased osteoclastogenesis, when mice were exposed to physiological level of fluoride. While he was unable to find comparable human trials, he recommended that this area be further explored as the technological means become available. The full citations are:

- Vieira A, Hanocock R, Eggertsson H, Everett E, Grynpas M. (2005). Tooth quality in dental fluorosis: genetic and environmental factors. *Calcified Tissue International.* 76:17-25.
- Yan D, Gurumurthy A, Wright M, Pfeiler T, Loboa E, Everett E. (2007). Genetic background influences fluoride's effects on osteoclastogenesis. *Bone.* 41:1036-1044.

Dr. Whitford cited published data that indicate that the susceptibility to dental fluorosis (Everett et al., J Dent Res 81: 794-698, 2002) and the mechanical properties in the bone (Mousny et al,. Bone 39: 1283-1289, 2006) are different among strains of mice. He noted that the differences presumably are due to genetic differences among the strains.

Charge Question 12. Do you support the OW's conclusion that an RfD of 0.07 mg/kg/day will be protective for severe dental fluorosis in children and skeletal effects in adults while still providing for the beneficial effects of fluoride?

In general, **Dr. Cauley, Dr. Jackson,** and **Dr. Whitford** support the use of the 0.07 mg/kg/day RfD. However, all three of these reviewers stated that there is a considerable degree of uncertainty regarding the estimate.

Dr. Jackson indicated that he does not think that a claim can be made that no single individual will be completely immune from the development of severe fluorosis even at this recommended RfD. As a result of an individual's potential genetic predisposition, he does not think that the possibility of developing severe fluorosis can be totally ruled out. Additionally, he noted that his concern is not as much about the amount of fluoride that is ingested through the public water supply, but the other well-known sources of fluoride (see his final comment for Question 12 in Appendix B for other sources) that have an additive effect to that derived from consuming fluoridated drinking water.

Dr. Whitford added that, as estimated by the IOM (1997), the RfD may be closer to 0.10 mg/kg/day for moderate (not severe) dental fluorosis and substantially higher than that for clinically significant skeletal effects in the United States.

On the other hand, **Dr. DenBesten** did not agree with this recommendation because it is based on limiting severe fluorosis to 1% of the population and the IOM's recommended adequate intake level. She suggested that the level be lowered to eliminate severe fluorosis. She noted that one percent of the population represents a relatively large number of individuals, and these individuals are most likely uniquely sensitive to fluoride. She recommended that the data be analyzed without taking the IOM's recommended adequate levels into account. Then, a secondary analysis could be conducted to include the IOM's recommendations.

Appendix A: Pre-Meeting Responses to the Charge Questions from Reviewers (Organized Alphabetically by Reviewer)

Jane A. Cauley, Dr.P.H
Professor, Department of Epidemiology, University of Pittsburgh

1. Was the document clear and transparent in its presentation of data and explanation of the analytical approaches used to characterize the concentration- response and dose-response relationships for severe dental fluorosis? If not, do you have any suggestions that will enable the authors to improve its clarity?

Response

The document could be strengthened by reorganizing it as a manuscript. The Introduction should provide background and a scientific rationale for this analysis. The specific objectives should be listed.. The methods for identifying all the literature should be transparent and consistently applied to both the fluorosis and fracture sections. This is typical of meta analyses which also apply certain quality scores to each paper.
I think it might also be helpful if I understand who the audience for this report is. I found it a confusing because I do not understand the roles of the EPA, OW and the NRC : how will each agency find this information helpful.? What prompted this report?

2. Are you aware of any significant publications related to severe dental fluorosis or the skeletal effects of fluoride that are not included in the noncancer assessment document?

Response

The dental fluorosis literature included studies with a wide range of fluoride levels while the skeletal/ fracture studies were limited to those with fluoride levels $\geq$ 4mg/l. You might consider adding some of the literature on fractures at lower levels of fluoride including the following:

Cauley JA, Murphy PA, Riley TJ, Buhari AM. Effects of fluoridated drinking water on bone mass and fractures: the study of osteoporotic fractures. J Bone Miner Res. 1995 Jul;10(7):1076-86.

Phipps, KR, Orwoll, ES, Mason, JD, Cauley, J A. Community water fluoridation, bone mineral density, and fractures: prospective study of effects in older women. BMJ 2000 321(7265):860-4.

In addition, Phipps KR et al published a study comparing bone mineral density in Deming, NM (0.7mg/L) and Lordsburg, NM (3.5mg/L). The citation is: Phipps KR, Burt BA. Water-borne fluoride and cortical bone mass: a comparison of two communities. J Dent Res. 1990 Jun;69(6):1256-60.

It appeared to me that this section is overall less thorough than the dental fluorosis section. Additional Selections include the following. I would pay particular attention to the papers by Kathy Phipps, Cyrus Cooper, S Jacobsen and T Hillier. These are high quality in my opinion.

Demos LL, Kazda H, Cicuttini FM, Sinclair MI, Fairley CK.
Water fluoridation, osteoporosis, fractures--recent developments.
Aust Dent J. 2001 Jun;46(2):80-7; quiz 143.
PMID: 11491235 [PubMed - indexed for MEDLINE]

Fabiani L, Leoni V, Vitali M.
Bone-fracture incidence rate in two Italian regions with different fluoride concentration levels in drinking water.
J Trace Elem Med Biol. 1999 Dec;13(4):232-7.
PMID: 10707346 [PubMed - indexed for MEDLINE]

Kleerekoper M.
Fluoride and the skeleton.
Crit Rev Clin Lab Sci. 1996 Apr;33(2):139-61. Review.
PMID: 8744520 [PubMed - indexed for MEDLINE]

Raheb J.
Water fluoridation, bone density and hip fractures: a review of recent literature.
Community Dent Oral Epidemiol. 1995 Oct;23(5):309-16. Review. No abstract available.
PMID: 8529346 [PubMed - indexed for MEDLINE]

Søgaard CH, Mosekilde L, Schwartz W, Leidig G, Minne HW, Ziegler R.
Effects of fluoride on rat vertebral body biomechanical competence and bone mass.
Bone. 1995 Jan;16(1):163-9.
PMID: 7742076 [PubMed - indexed for MEDLINE]

Kröger H, Alhava E, Honkanen R, Tuppurainen M, Saarikoski S.
The effect of fluoridated drinking water on axial bone mineral density--a population-based study.
Bone Miner. 1994 Oct;27(1):33-41.
PMID: 7849544 [PubMed - indexed for MEDLINE]

Ripa LW.
A half-century of community water fluoridation in the United States: review and
commentary.
J Public Health Dent. 1993 Winter;53(1):17-44. Review.
PMID: 8474047 [PubMed - indexed for MEDLINE]

Gordon SL, Corbin SB.
Summary of workshop on drinking water fluoride influence on hip fracture on bone
health. (National Institutes of Health, 10 April, 1991)
Osteoporos Int. 1992 May;2(3):109-17. No abstract available.
PMID: 1627897 [PubMed - indexed for MEDLINE]

Colquhoun J.
Water fluoride and fractures.
N Z Med J. 1991 Aug 14;104(917):343. No abstract available.

PMID: 1876343 [PubMed - indexed for MEDLINE]

McNeill KG, Coote GE, Hitchman AJ.
Uptake of fluorine in cortical and trabecular bone.
J Bone Miner Res. 1991 Aug;6(8):859-64.
PMID: 1785375 [PubMed - indexed for MEDLINE]

Arnala I, Alhava EM, Kivivuori R, Kauranen P.
Hip fracture incidence not affected by fluoridation. Osteofluorosis studied in
Finland.
Acta Orthop Scand. 1986 Aug;57(4):344-8.
PMID: 3788501 [PubMed - indexed for MEDLINE]

Ericsson Y, Luoma H, Ekberg O.
Effects of calcium, fluoride and magnesium supplementations on tissue mineralization in calcium- and magnesium-deficient rats.
J Nutr. 1986 Jun;116(6):1018-27.
PMID: 3723198 [PubMed - indexed for MEDLINE]

Simonen O, Laitinen O.
Does fluoridation of drinking-water prevent bone fragility and osteoporosis?
Lancet. 1985 Aug 24;2(8452):432-4.
PMID: 2863455 [PubMed - indexed for MEDLINE]

Madans J, Kleinman JC, Cornoni-Huntley J.
The relationship between hip fracture and water fluoridation: an analysis of national data.
Am J Public Health. 1983 Mar;73(3):296-8.
PMID: 6824115 [PubMed - indexed for MEDLINE]

Schamschula RG, Barmes DE.
Fluoride and health: dental caries, osteoporosis, and cardiovascular disease.
Annu Rev Nutr. 1981;1:427-35. Review.
PMID: 6764723 [PubMed - indexed for MEDLINE]

Alhava EM, Olkkonen H, Kauranen P, Kari T.
The effect of drinking water fluoridation on the fluoride content, strength and
mineral density of human bone.
Acta Orthop Scand. 1980 Jun;51(3):413-20.
PMID: 7446020 [PubMed - indexed for MEDLINE]

Stein ID, Granik G.
Human vertebral bone: relation of strength, porosity, and mineralization to fluoride content.
Calcif Tissue Int. 1980;32(3):189-94.
PMID: 6775787 [PubMed - indexed for MEDLINE]

Hegsted DM.
Osteoporosis and fluoride deficiency.
Postgrad Med. 1967 Jan;41(1):A49-53. No abstract available.
PMID: 6036214 [PubMed - indexed for MEDLINE]

Bernstein DS, Sadowsky N, Hegsted DM, Guri CD, Stare FJ.
Prevalence of osteoporosis in high- and low-fluoride areas in North Dakota.

JAMA. 1966 Oct 31;198(5):499-504. No abstract available.
PMID: 5953273 [PubMed - indexed for MEDLINE]

Hillier S, Cooper C, Kellingray S, Russell G, Hughes H, Coggon D.
Fluoride in drinking water and risk of hip fracture in the UK: a case-control
study.
Lancet. 2000 Jan 22;355(9200):265-9.
PMID: 10675073 [PubMed - indexed for MEDLINE]

Hillier S, Inskip H, Coggon D, Cooper C.
Water fluoridation and osteoporotic fracture.
Community Dent Health. 1996 Sep;13 Suppl 2:63-8. Review.
PMID: 8897754 [PubMed - indexed for MEDLINE]

Jacobsen SJ, Goldberg J, Cooper C, Lockwood SA.
The association between water fluoridation and hip fracture among white women and
men aged 65 years and older. A national ecologic study.
Ann Epidemiol. 1992 Sep;2(5):617-26.
PMID: 1342313 [PubMed - indexed for MEDLINE]

Cooper C, Wickham C, Lacey RF, Barker DJ.
Water fluoride concentration and fracture of the proximal femur.
J Epidemiol Community Health. 1990 Mar;44(1):17-9.
PMID: 2348142 [PubMed - indexed for MEDLINE]

1: Phipps KR, Orwoll ES, Mason JD, Cauley JA.
Community water fluoridation, bone mineral density, and fractures: prospective
study of effects in older women.
BMJ. 2000 Oct 7;321(7265):860-4.
PMID: 11021862 [PubMed - indexed for MEDLINE]

Phipps KR, Orwoll ES, Bevan L.
The association between water-borne fluoride and bone mineral density in older
adults.
J Dent Res. 1998 Sep;77(9):1739-48.
PMID: 9759671 [PubMed - indexed for MEDLINE]

Phipps K.
Fluoride and bone health.
J Public Health Dent. 1995 Winter;55(1):53-6. Review.
PMID: 7776293 [PubMed - indexed for MEDLINE]

Phipps KR, Burt BA.
Water-borne fluoride and cortical bone mass: a comparison of two communities.
J Dent Res. 1990 Jun;69(6):1256-60.
PMID: 2355118 [PubMed - indexed for MEDLINE]

3. Are the strengths and weaknesses of Dean (1942) study (selected as critical) fully characterized?

Response:

Additional strengths include the wide range of fluoride concentrations, although there were fewer subjects in the high fluoride categories. The dose response in the data showing increasing risk of severe skeletal fluorosis with increasing fluoride is also a strength. Finally the consistency of the findings across several different communities.

Other weaknesses: No information is provided on race/ethnicity. The data were collected the late 1930-40s. Although confounding by use of other fluoride products would be minimal, there are many other cohort differences between children exposed to fluoride in the late 1930-40s compared to now. For example, dental hygiene, dietary intakes (less water… more carbonated beverages), body weight are very different in today's children compared to children in the 30s.. Do puberty/ hormonal changes influence fluoride effects? This may be important because age of menarche has been decreasing. Hormones have been shown to influence dental health. Another weakness of the studies reviewed is that there appears to be no information on duration of exposure. How long did these children live in each community? Did the inclusion criteria include a minimum time of residence? If so this point needs to be made in the document.

4. Are some teeth more susceptible to sever fluorosis than others? If so, should the OW make modifications to the age range identified as the period of concern?

Response

This should be highlighted in a separate section. A more direct discussion of the age at risk is needed. A table summarizing the ages of the children in each study and the age range which appeared to be at highest risk would be helpful. You could add this to Table 3-16.

5. Are the data on cavities as collected and graphically presented by the OW consistent with the hypothesis that there is an increased risk for cavities in susceptible individuals with severe dental fluorosis?

Response

The data summarized in Table 3-40 to 3-44 are convincing that dental caries are more common in subjects with severe fluorosis. The data suggest a U shaped relationship with fluoride: a higher risk in those with very low intakes and intakes >2-3mg/L.

6. Are there recent data that would impact the IOM (1997) Adequate Intake value of 0.05 mg/kg/day for fluoride that the OW should consider in this assessment?

Response

I do not know of any more recent data. Perhaps you want to mention life stage and Upper limit of toxicity. The ADA website looks like they updated their Fluoride document in 2006.

TABLE S-5
Criteria and Dietary Reference Intake Values for Fluoride by Life Stage Group

Life Stage Group		Criterion	AI (mg/day)[a]	
			Male	Female
	0 through 6 months	Human milk content	0.01	0.01
	7 through 12 months	Caries prevention	0.5	0.5
	1 through 3 years	Caries prevention	0.7	0.7
	4 through 8 years	Caries prevention	1	1
	9 through 13 years	Caries prevention	2	2
	14 through 18 years	Caries prevention	3	3
	19 through 30 years	Caries prevention	4	3
	31 through 50 years	Caries prevention	4	3
	51 through 70 years	Caries prevention	4	3
>	70 years	Caries prevention	4	3
Pregnancy				
<=	18 years	Caries prevention	--	3
	19 through 50 years	Caries prevention	--	3
Lactation				
<=	18 years	Caries prevention	--	3
	19 through 50 years	Caries prevention	--	3

[a]AI = Adequate Intake. For healthy infants fed human milk, AI is the mean intake. The observed estimate of nutrient intake that reduces the incidence of dental caries maximally in a group of healthy people. The AI is used if the scientific evidence is not available to derive an EAR. The AI is believed to cover their

needs, but lack of data or uncertainty in the data prevent being able to specify with confidence the percentage of individuals covered by this intake.

TABLE S-6
Tolerable Upper Intake Levels (UL[a]), by Life Stage Group

Life Stage Group		Calcium (g/day)	Phosphorus (g/day)	Magnesium (mg/day)[b]	Vitamin D (µg/day)[c]	Fluoride
	0 through 6 months	ND[d]	ND	ND	25	0.7
	7 through 12 months	ND	ND	ND	25	0.9
	1 through 3 years	2.5	3	65	50	1.3
	4 through 8 years	2.5	3	110	50	2.2
	9 through 18 years	2.5	4	350	50	10
	19 through 70 years	2.5	4	350	50	10
>	70 years	2.5	3	350	50	10
Pregnancy						
<=	18 years	2.5	3.5	350	50	10
	19 through 50 years	2.5	3.5	350	50	10
Lactation						
<=	18 years	2.5	4	350	50	10
	19 through 50 years	2.5	4	350	50	10

[a]UL = the maximum level of daily nutrient intake that is likely to pose no risk of adverse effects to members of the healthy general population. Unless specified otherwise, the UL represents total intake from food, water, and supplements.

[b]The UL for magnesium represents intake from pharmacological agents only and does not include intake from food and water.

[c]As cholecalciferol. 1 µg cholecalciferol = 40 IU vitamin D.

[d]ND. Not determinable due to lack of data of adverse effects in this age group and concern with regard to lack of ability to handle excess amounts. Source of intake should be from food only in order to prevent high

levels of intake.

7. Can you suggest an approach to transform the water concentration data from the Dean (1942) study to units of dose for the population susceptible to severe dental fluorosis other than that used by the OW?

Response

To estimate the RFD of 0.07 mg/Kg/day, the OW attempted to obtain detailed analysis of average body weights and water intakes during the time the Dean data was collected. Data from the Erchow and Cantor were used (1989). Why not use more generalizable data from NHANES at least for body weight? You could still use the water intake estimates from the Cantor paper.

8. Can you provide input concerning the strengths and weaknesses of the approach utilized by the OW to identify a lower bound dose for severe dental fluorosis?

Response

Strengths: Conservative estimates carried out with and without the outlier community; several different models were considered, sensitivity analysis showed that even eliminating three highest data points had little effect on the model goodness of fit or on the BMDL.

Weakness: Inherent weakness of the approach relate primarily to the weaknesses cited related to the parent Dean Study. You might consider adding a formal test for trend in data.

9. Are you aware of dose estimates other than those from IOM (1997) and WHO (2002) that are appropriate critical doses for skeletal effects and/or can you suggest a different approach that the OW might use to estimate the fluoride dose associated with skeletal fractures using available data?

Response

See answer to #2 for additional references.

Some concern about the inconsistencies across the age groups. The significance of the differences in results across age could have occurred by chance alone.

Referring to the" radius" is problematic because if it s the distal radius, it is primarily trabecular BMD and mid radius is predominantly cortical BMD. If the effects of fluoride differs by type of bone, this is very problematic.

Randomized trials: please note the compliance/adherence to the Na F because it is poorly tolerated. This is a limitation to theses studies. Did they adhere to the intent to treat principle?

10. Are you aware of any data that can be used to demonstrate that protection of the secondary teeth from severe dental fluorosis will also protect the primary teeth?

Response

No, I am not aware of any additional data.

11. Are there factors other than those discussed in Section 3.1.4 that increase sensitivity to fluoroisis of the teeth or bones?

Response

I am not sure but I question whether race/ ethnicity and pubertal stage is important.

12. Do you support the OW's conclusion that an RfD of 0.07 mg/kg/day will be protective for severe dental fluorosis in children and skeletal effects in adults while still providing for the beneficial effects of fluoride?

Response

Yes, I am in support of the 0.07 mg/kg/day but I would admit the uncertainty that surrounds this estimate..

Pamela Denbesten, D.D.S., M.S.
Professor and Chair, Division of Pediatric Dentistry, Department of Orofacial Sciences, University of California at San Francisco

1. Was the document clear and transparent in its presentation of data and explanation of the analytical approaches used to characterize the concentration- response and dose-response relationships for severe dental fluorosis? If not, do you have any suggestions that will enable the authors to improve its clarity?

Response:
The document was for the most part clear and transparent in its presentation. To improve clarity, I suggest the following:

Page 13. Please indicate the purpose of the Secondary Maximum Contaminant Level, and what this was used for.

Page 15, section 2.1.1. The statement with regards to fluoroapatite, at the end of paragraph 2.1.1 is not accurate. The mineral formed in tooth enamel exposed to higher fluoride levels is fluoride-containing carbonated apatite, rather than fluoroapatite. In fluorotic enamel, fluoride levels are increased to about 200 ppm rather than the 10-100 ppm fluoride in normal enamel below the surface, while fluorapatite is about 30,000 ppm fluoride. The fluoride-substituted apatite, formed with increased levels of ingested fluoride, has some increased resistance to bacterial acids that cause tooth decay. However, the primary function of fluoride in drinking water to reduce tooth decay is topical, primarily by the enhancement of remineralization.

References:
1. Featherstone JDB. Prevention and reversal of dental caries: role of low level fluoride. Community Dent and Oral Epidemiol 1999;27:31-40.
2. Featherstone JDB. The science and practice of caries prevention. JADA 2000;131:887-99.
3. Fejerskov O, Thylstrup A, Larsen MJ. Rational use of fluorides in caries prevention. Acta Odont. Scand. 1981;39:241-49.
4. LeGeros RZ. Calcium Phosphates in Enamel, Dentin and Bone. In: Myers HM, editor. Calcium Phosphates in Oral Biology and Medicine. Basel: Karger; 1991. p. 108-29.
5. Robinson C, Kirkham J, Weatherell JA. Fluoride in teeth and bone. In: Fejerskov O, Ekstrand J, Burt BA, editors. Fluoride in Dentistry. 2nd ed. Copenhagen: Munksgaard; 1996. p. 69-87.
6.Ten Cate JM, Featherstone JDB. Mechanistic aspects of the interactions between fluoride and dental enamel. CRC Critical Reviews in Oral Biology 1991;2:283-96.

Page 19 section 2.2. Determine whether in fact the apatite formed in bone is fluoroapatite or like in tooth, is a fluoride-substituted hydroxyapatite.

Page 21 section 3. Change "…are preferred for evaluating the potential effects of fluoride in drinking water", to the more accurate statement, "…are preferred for evaluating the potential effects of ingested fluoride".

Page 57; paragraph 1. Statements relating fluoride in the drinking water to anti-caries benefit from studies conducted after 1980, should be qualified in that other sources of fluoride may be confounding factors. For example, fluorosis incidence in more recent studies may decrease as consumption of tap water is altered by consumption of bottled beverages.

In general for all of section 3.2.2, when fluoride effects on dental caries are discussed, the data should be divided into studies before and after 1980 when fluoride was widely available in toothpaste, and perhaps before and after the late 1990s when bottled beverages became widely used.

Page 86; Table 3-52. What does "complete" and "total" refer to?

Page 94; section 4.4. Explain with the "NOAEL/LOAEL" approach is, or at least spell out the acronmym.

Page 97 paragraph 1. The importance of fluoride as a nutrient may need to be reassessed, given that its primary function in caries prevention is topical. It would seem more appropriate focus on the upper limits for ingestion of this caries preventive agent.

Page 98. The statement describing the timing incisor tooth formation is incorrect. The secondary incisors and molars begin development in utero, and continue with crown formation complete at about 3 years of age.

Page 101. Please justify the rationale for setting the BMDL at an incidence of 1% severe fluorosis, rather than 0% severe fluorosis.

Page 103. As stated above, I question the concept of a recommended fluoride intake.

Page 104, paragraph 1. The statement that "…fluoroapatite crystals disrupt the hydroxyapatite crysal lattice…" is incorrect and should be deleted.

2. Are you aware of any significant publications related to severe dental fluorosis or the skeletal effects of fluoride that are not included in the noncancer assessment document?

Response:
No I am not

3. Are the strengths and weaknesses of Dean (1942) study (selected as critical) fully characterized?

Response:
Yes, to me they are.

4. Are some teeth more susceptible to severe fluorosis than others? If so, should the OW make modifications to the age range identified as the period of concern?

Response:
The OW report was clear in showing that posterior teeth are also susceptible, and the case for including children up to age 14 was clear and compelling.

5. Are the data on cavities as collected and graphically presented by the OW consistent with the hypothesis that there is an increased risk for cavities in susceptible individuals with severe dental fluorosis?

Response
Yes

6. Are there recent data that would impact the IOM (1997) Adequate Intake value of 0.05 mg/kg/day for fluoride that the OW should consider in this assessment?

Response.
I do not know on what basis the IOM recommended that fluoride levels at 0.05 mg/kg/day be ingested. However the weight of evidence is that the primary mechanism by which fluoride protects against tooth decay is topical.

I would agree with the recent NRC report that states; "The recommended optimal fluoride intake for children to maximize caries prevention and minimize the occurrence of dental fluorosis is often stated as being 0.05-0.07 mg/kg/day (Levy1994; Heller et al. 1999, 2000). Burt (1992) attempted to track down the origin of the estimate of 0.05-0.07 mg/kg/day as an optimum intake of fluoride but was unable to find it. He interpreted the available evidence as suggesting that 0.05-0.07 mg/kg/day (from all sources) remains a useful upper limit for fluoride intake in children".

I agree with Burt's interpretation, and do not believe that the EPA should consider that there is a particular desired fluoride intake. For further discussion on the relative importance of fluoride ingestion in caries prevention, please see the response to question 1.

7. Can you suggest an approach to transform the water concentration data from the Dean (1942) study to units of dose for the population susceptible to severe dental fluorosis other than that used by the OW?

Response:
No I can not. Of course serum fluoride levels would be the most useful measurement, but these levels are not available. I believe the using Dean's data as a starting point to quantify total ingestion is the best that we can do. I suggest that the suggestion be given that future studies include random sampling of serum fluoride levels to strengthen future decision making relative to fluoride intake.

8. Can you provide input concerning the strengths and weaknesses of the approach utilized by the OW to identify a lower bound dose for severe dental fluorosis?

Response:

9. Are you aware of dose estimates other than those from IOM (1997) and WHO (2002) that are appropriate critical doses for skeletal effects and/or can you suggest a different approach that the OW might use to estimate the fluoride dose associated with skeletal fractures using available data?
Response: No I can not.

10. Are you aware of any data that can be used to demonstrate that protection of the secondary teeth from severe dental fluorosis will also protect the primary teeth?

Response:
Primary teeth are known to be less affected by fluoride ingestion, most likely because they are formed in utero with a dilution of maternal serum fluoride levels as fluoride in the placenta. This suggests that minimizing the risk of severe dental fluorosis in secondary teeth will also protect the primary teeth

11. Are there factors other than those discussed in Section 3.1.4 that increase sensitivity to fluoroisis of the teeth or bones?

Response:
There seem to be individuals who are uniquely sensitive to the effects of fluoride on enamel formation. These individuals may have other more "hidden" enamel defects that are exacerbated by the effects of fluoride. Little is currently know as to why some individuals seem to be more fluoride sensitive.

12. Do you support the OW's conclusion that an RfD of 0.07 mg/kg/day will be protective for severe dental fluorosis in children and skeletal effects in adults while still providing for the beneficial effects of fluoride?

Response: I do not agree with this recommendation as it is based on limiting severe fluorosis to 1% of the population. I suggest that the level be lowered to eliminate severe fluorosis. One percent of the population represents a relatively large number of individuals, and given the data showing the primarily topical effects of fluoride I do not see a rationale for acceptance of the 1% severe fluorosis

Richard D. Jackson, D.M.D.
Assistant Professor, Preventive and Community Dentistry, School of Dentistry, Indiana University, Oral Health Research Institute

1. Was the document clear and transparent in its presentation of data and explanation of the analytical approaches used to characterize the concentration- response and dose-response relationships for severe dental fluorosis? If not, do you have any suggestions that will enable the authors to improve its clarity?

I felt that the document clearly described what is available in the literature and presented the information in an understandable format. I am glad individual tables summarizing some of the studies were included for reference. As an aside, I would have preferred that the studies be described chronologically as opposed to alphabetically by author.

2. Are you aware of any significant publications related to severe dental fluorosis or the skeletal effects of fluoride that are not included in the noncancer assessment document?

Jackson R, Kelly S, Katz B, Brizendine E, Stookey G. Dental fluorosis in children residing in communities with different fluoride levels in the water: 33 month follow-up. Pediatric Dentistry 21:248-254.

Examination of 357 school-age children in 1994 from three Indiana communities with varying levels of fluoride in the water (0.2, 1.0 and 4.0 ppm). Severe fluorosis using the TSIF (score 5, 6 or 7) was noted in 9% of the children examined in the 4.0 ppm community. No scores of this magnitude were seen in the other two communities. Based on comparison to previous prevalence data in the same communities in 1992, the prevalence of fluorosis had increased in each community but primarily in the categories rated as a 1, 2 or 3 using the TSIF.

3. Are the strengths and weaknesses of Dean (1942) study (selected as critical) fully characterized?

I think the strengths of the investigation conducted by Dean (1942) are fully characterized. I doubt that anyone would disagree that Dean (1942) has been and will continue to be a benchmark study in the dental literature as well as the much broader literature related to public health and epidemiology. It is true that Dean's study was performed when confounding fluoride sources were not available and thus in all probability gives a very clear picture of the prevalence of the relationship of fluoride ingestion and the subsequent development of dental fluorosis. However, one point of weakness that could be argued is that the population of subjects examined was not diverse in terms of racial or cultural characteristics. As it has been postulated that genetic factors may impact on the expression of dental fluorosis at identical levels of ingestion, the fact that the data was collected in an exclusively white population appears to limit its applicability for use as a benchmark.

4. Are some teeth more susceptible to severe fluorosis than others? If so, should the OW make modifications to the age range identified as the period of concern?

I was a little unclear as to the intent of the question. My initial reaction was to answer "No". I have not come across any literature that indicates that some teeth are more susceptible to the development of severe dental fluorosis assuming that the exposure is constant and taking into consideration how long the developing tooth is exposed to higher levels of fluoride. For example, maxillary third molars may present with greater evidence of severe fluorosis (pitting, staining, etc) based on the fact that they take longer to develop (Cc 12-16 years-of-age) and erupt as opposed to a maxillary central incisor (Cc 5 years-of-age).

5. Are the data on cavities as collected and graphically presented by the OW consistent with the hypothesis that there is an increased risk for cavities in susceptible individuals with severe dental fluorosis?

It appears that the data are consistent with and support this hypothesis. A point of discussion may be made concerning examiner bias in the methodology of these studies. It is obvious that it would be impossible to eliminate bias as the most severely affected teeth would be evident by the caries examiner when determining the DMFT/DMFS of the subject. I don't see any solution for this dilemma with the use of a single examiner or multiple examiners but it is something that should at least be mentioned as a possible confounder.

Jackson et al (1995) also examined children residing in communities with varying levels of fluoride in their public water supplies. No child in the negligibly-fluoridated community or the optimally-fluoridated community was found to have TSIF scores greater than 2. However, the DMFT and DMFS in the 7-10 year-olds residing in the 4X optimum community were not significantly different from those residing in the 1.0 ppm community. For 11-14 year-olds DMFS was significantly lower than in the negligibly fluoridated community.

6. Are there recent data that would impact the IOM (1997) Adequate Intake value of 0.05 mg/kg/day for fluoride that the OW should consider in this assessment?

I am not aware of any new data that could influence the value of 0.5 mg/kg/day as the AI value.

7. Can you suggest an approach to transform the water concentration data from the Dean (1942) study to units of dose for the population susceptible to severe dental fluorosis other than that used by the OW?

No, using the data available the approach seems to be logical. The problem, as I see it. is the US marketplace is constantly changing and ingestion of tap water continue to decline. The EPA (2004) report stated that bottled water accounted for only 13% of water consumption in the United States. More recent trade manufacturing data indicates that bottled water consumption in the United States exceeds this percentage by a wide margin and bottled water consumption may surpass tapwater consumption in the near future. Other published data suggest that among Hispanic individuals, tap water is commonly perceived as "unhealthy" and again bottled water is consumed almost exclusively. Another point that should be further explored is the possible "halo effect" of imported foods and beverages into the United States and the fluoride content of these consumables.

8. Can you provide input concerning the strengths and weaknesses of the approach utilized by the OW to identify a lower bound dose for severe dental fluorosis?

9. Are you aware of dose estimates other than those from IOM (1997) and WHO (2002) that are appropriate critical doses for skeletal effects and/or can you suggest a different approach that the OW might use to estimate the fluoride dose associated with skeletal fractures using available data?

I am not aware of additional dose estimates that would be appropriate for determining critical dosage levels and their possible skeletal effects.

IOM.

Recommended total dietary fluoride intake

Age	Reference weight*		Adequate intake†	Tolerable upper intake§
	kg	lb	mg/day	mg/day
0–6 months	7	16	0.01	0.7
6–12 months	9	20	0.5	0.9
1–3 years	13	29	0.7	1.3
4–8 years	22	48	1.1	2.2
>9 years	40/76	88/166	2.0/3.8	10.0

*** Values based on data collected during 1988–1994 as part of the third National Health and Nutrition Examination Survey.**
† Intake that maximally reduces occurrence of dental caries without causing unwanted side effects, including moderate enamel fluorosis.
§ Highest level of nutrient intake that is likely to pose no risks for adverse health effects in almost all persons.
Source: Adapted from Institute of Medicine. Fluoride. In: Dietary reference intakes for calcium,phosphorus, magnesium, vitamin D, and fluoride. Washington, DC: National Academy Press,1997:288–313.

BONE FRACTURE AND BONE DEVELOPMENT PROBLEMS
There were 29 studies included on the association between bone fracture and bone development problems and water fluoridation. Other than fluorosis, bone effects (not including bone cancers) were the most studied potential adverse effect. These studies had a mean validity score of 3.4 out of 8. All but one study were of evidence level C. These studies included both cohort and ecological designs, some of which included analyses controlling for potential confounding factors. Observer bias could potentially play a role in bone fracture studies, depending on how the study is conducted.
The evidence on bone fracture can be classified into hip fracture and other sites because there are more studies on hip fracture than any other site. Using a qualitative method of analysis (Figure 8.1), there is no clear association of hip fracture with water fluoridation. Theevidence on other fractures is similar. Overall, the findings of studies of bone fracture effects showed small variations around the 'no effect' mark. A meta-regression of bone fracture studies also found no association with water fluoridation.

10. Are you aware of any data that can be used to demonstrate that protection of the secondary teeth from severe dental fluorosis will also protect the primary teeth?

I am not aware of any data that supports this possibility.

11. Are there factors other than those discussed in Section 3.1.4 that increase sensitivity to fluorosis of the teeth or bones?

Section 3.1.4. states that climate and altitude of place of residence; dietary habits, nutritional status, physiological state, and certain pathological conditions may affect the occurrence and severity of fluoride-induced dental fluorosis. The section further mentions that certain genetic conditions such as amelogenesis imperfecta may lead to defective development of enamel. There have been studies (Vieira et al, 2005; Yan et al, 2007) that suggest, in mice, that there may be a varying genetic response to identical levels of fluoride ingestion. These changes have been identified in both tooth and bone formation. Vieira et al found that genetic influences have a direct bearing on the biomechanical properties of the teeth. Yan et al. found strain-specific effects of physiological level of fluoride with increased osetoclastogenesis in some mouse strains. While I was unable to find comparable human trials, this area should be further explored as the technological means become available.

A study by Hong et al (2004) appears to indicate that the use of amoxicillin could play a contributing role in the development of primary tooth fluorosis, especially for children exposed to lower levels of fluoride.

12. Do you support the OW's conclusion that an RfD of 0.07 mg/kg/day will be protective for severe dental fluorosis in children and skeletal effects in adults while still providing for the beneficial effects of fluoride?

References

Hong L, Levy S, Warren J, Bergus G, Dawson D, Wefel J, Broffitt B. Primary tooth fluorosis and amoxicillin use during infancy. Journal of Public Health Dentistry 64: 38-44 (2004).

Vieira A, Hanocock R, Eggertsson H, Everett E, Grynpas M. Tooth quality in dental fluorosis: genetic and environmental factors. Calcified Tissue International 76:17-25 (2005).

Yan D, Gurumurthy A, Wright M, Pfeiler T, Loboa E, Everett E. Genetic background influences fluoride's effects on osteoclstogenesis. Bone 41:1036-1044 (2007).

Jackson R, Kelly S, Katz B, Hull J, Stookey G. Dental fluorosis and caries prevalence in children residing in communities with different levels of fluoride in the water. Journal of Public Health Dentistry 88: 79-84 (1995).

A systematic review of public water fluoridation
NHS Centre for Review and Dissemination
University of York 2000
EXECUTIVE SUMMARY

This systematic review has been commissioned by the Chief Medical Officer of the Department of Health to 'carry out an up to date expert scientific review of fluoride and health' (Paragraph 9.20, Our Healthier Nation).

Overall, the aim has been to assess the evidence on the positive and negative effects of population wide drinking water fluoridation strategies to prevent caries. To achieve this aim five objectives were identified:

Objective 1: What are the effects of fluoridation of drinking water supplies on the incidence of caries?
Objective 2: If water fluoridation is shown to have beneficial effects, what is the effect over and above that offered by the use of alternative interventions and strategies?
Objective 3: Does water fluoridation result in a reduction of caries across social groups and between geographical locations, bringing equity?
Objective 4: Does water fluoridation have negative effects?
Objective 5: Are there differences in the effects of natural and artificial water fluoridation?

Methods

A search of 25 electronic databases (with no language restrictions) and the world-wide-web was undertaken. Relevant journals and indices were hand searched and attempts were made to contact authors for further information.

Quality inclusion criteria were based on a pre-defined hierarchy of evidence (A, B, and C). Studies of efficacy were included if they were of evidence level A or B. In order to allow the broadest search for evidence on potential adverse effects, studies of all levels of evidence were included. Objective specific inclusion criteria, based on selection of participants, intervention, outcomes assessed, and study design appropriate for a given objective were then applied. Study validity was formally assessed using a published checklist modified for this review (CRD Report 4, 1996).

Inclusion criteria were assessed independently by at least two reviewers. Extraction of data from, and validity assessment of, included studies was independently performed by two reviewers, and checked by a third reviewer. Disagreements were resolved through consensus.

Where the data were in a suitable format, measures of effect and 95% confidence intervals (CI) were plotted. Heterogeneity was investigated by visual examination and statistically using the Q-statistic. Where no evidence of heterogeneity was found a meta-analysis was conducted to produce a pooled estimate of the measure of effect. Statistically significant heterogeneity was investigated using meta-regression. Multiple regression analysis was used to explore the relationship between fluoridation and fluorosis.

Results

214 studies met full inclusion criteria for one or more of the objectives. No randomised controlled trials of the effects of water fluoridation were found. The study designs used included 45 ‘before and after’ studies, 102 cross-sectional studies, 47 ecological studies, 13 cohort (prospective or retrospective) studies and 7 case-control studies. Several studies were reported in multiple papers over a number of years.

Results by Objective

Objective 1

A total of 26 studies of the effect of water fluoridation on dental caries were found. For this objective, the quality of studies found was moderate (no level A studies). A large number of studies were excluded because they were cross-sectional studies and therefore did not meet the inclusion criteria of being evidence level B or above. All but three of the studies included were before-after studies, two included studies used prospective cohort designs, and one used a retrospective cohort design. All before-after studies located by the search were included. The most serious defect of these studies was the lack of appropriate analysis. Many studies did not present an analysis at all, while others only did simple analyses without attempting to control for potentially confounding factors. While some of these studies were conducted in the 1940’s and 50’s, prior to the common use of such analyses, studies conducted much later also failed to use methods that were commonplace at the time of the study.

Another defect of many studies was the lack of any measure of variance for the estimates of decay presented. While most studies that presented the proportion of caries-free children contained sufficient data to calculate standard errors, this was not possible for the studies that presented dmft/DMFT scores. Only four of the eight studies using these data provided estimates of variance.

The best available evidence suggests that fluoridation of drinking water supplies does reduce caries prevalence, both as measured by the proportion of children who are caries free and by

the mean change in dmft/DMFT score. The studies were of moderate quality (level B), but of limited quantity. The degree to which caries is reduced, however, is not clear from the data available. The range of the mean difference in the proportion (%) of caries-free children is -5.0 to 64%, with a median of 14.6% (interquartile range 5.05, 22.1%). The range of mean change in dmft/DMFT score was from 0.5 to 4.4, median 2.25 teeth (interquartile range 1.28, 3.63 teeth). It is estimated that a median of six people need to receive fluoridated water for one extra person to be caries-free (interquartile range of study NNTs 4, 9). The best available evidence from studies following withdrawal of water fluoridation indicates that caries prevalence increases, approaching the level of the low fluoride group. Again, however, the studies were of moderate quality (level B), and limited quantity. The estimates of effect could be biased due to poor adjustment for the effects of potential confounding factors.

Objective 2

To address this objective, studies conducted after 1974 were examined. While only nine studies were included for Objective 2, these would have been enough to provide a confident answer to the objective's question if the studies had been of sufficient quality. Since these studies were completed after 1974, one might expect that the validity assessments would be higher than the earlier studies following the introduction of more rigorous study methodology and analytic techniques. However, the average validity checklist score and level of evidence was essentially the same for studies after 1974 as those conducted prior to 1974. Hence, the ability to answer this objective is similar to that in Objective 1.

In those studies completed after 1974, a beneficial effect of water fluoridation was still evident in spite of the assumed exposure to non-water fluoride in the populations studied. The metaregression conducted for Objective 1 confirmed this finding.

Objective 3

No level A or B studies examining the effect of water fluoridation on the inequalities of dental health between social classes were identified. However, because of the importance of this objective, level C studies conducted in England were included. A total of 15 studies investigating the association of water fluoridation, dental caries and social class in England were identified. The quality of the evidence of the studies was low, and the measures of social class that were used varied. Variance data were not reported in most of these studies, so a statistical analysis was not undertaken.

There appears to be some evidence that water fluoridation reduces the inequalities in dental health across social classes in 5 and 12 year-olds, using the dmft/DMFT measure. This effect was not seen in the proportion of caries-free children among 5 year-olds. The data for the effects in children of other ages did not show an effect. The small quantity of studies, differences between these studies, and their low quality rating, suggest *caution* in interpreting these results.

Objective 4

DENTAL FLUOROSIS

Dental fluorosis was the most widely and frequently studied of all negative effects. The fluorosis studies were largely cross-sectional designs, with only four before-after designs. Although 88 studies of fluorosis were included, they were of low quality. The mean validity score for fluorosis was only 2.8 out of 8. All, but one, of the studies were of evidence level C. Observer bias may be of particular importance in studies assessing fluorosis. Efforts to control for the effects of potential confounding factors, or reducing potential observer bias were uncommon.

As there may be some debate about the significance of a fluorosis score at the lowest level of each index being used to define a person as 'fluorosed', a second method of determining the proportion 'fluorosed' was selected. This method describes the number of children having dental fluorosis that may cause 'aesthetic concern'.

With both methods of identifying the prevalence of fluorosis, a significant dose-response relationship was identified through a regression analysis. The prevalence of fluorosis at a water fluoride level of 1.0 ppm was estimated to be 48% (95% CI 40 to 57) and for fluorosis of aesthetic concern it was predicted to be 12.5% (95% CI 7.0 to 21.5). A very rough estimate of the number of people who would have to be exposed to water fluoride levels of 1.0 ppm for one additional person to develop fluorosis of any level is 6 (95% CI 4 to 21), when compared

with a theoretical low fluoride level of 0.4 ppm. Of these approximately one quarter will have fluorosis of aesthetic concern, but the precision of these rough estimates is low. These estimates only apply to the comparison of 1.0 ppm to 0.4 ppm, and would be different if other levels were compared.

BONE FRACTURE AND BONE DEVELOPMENT PROBLEMS

There were 29 studies included on the association between bone fracture and bone development problems and water fluoridation. Other than fluorosis, bone effects (not including bone cancers) were the most studied potential adverse effect. These studies had a mean validity score of 3.4 out of 8. All but one study were of evidence level C. These studies included both cohort and ecological designs, some of which included analyses controlling for potential confounding factors. Observer bias could potentially play a role in bone fracture studies, depending on how the study is conducted.

The evidence on bone fracture can be classified into hip fracture and other sites because there are more studies on hip fracture than any other site. Using a qualitative method of analysis (Figure 8.1), there is no clear association of hip fracture with water fluoridation. The evidence on other fractures is similar. Overall, the findings of studies of bone fracture effects showed small variations around the 'no effect' mark. A meta-regression of bone fracture studies also found no association with water fluoridation.

CANCER STUDIES

There were 26 studies of the association of water fluoridation and cancer included. Eighteen of these studies are from the lowest level of evidence (level C) with the highest risk of bias. There is no clear association between water fluoridation and overall cancer incidence and mortality. This was also true for osteosarcoma and bone/joint cancers. Only two studies considered thyroid cancer and neither found a statistically significant association with water fluoridation.

Overall, no clear association between water fluoridation and incidence or mortality of bone cancers, thyroid cancer or all cancers was found.

OTHER POSSIBLE NEGATIVE EFFECTS

A total of 33 studies of the association of water fluoridation with other possible negative effects were included in the review. Interpreting the results of studies of other possible negative effects is very difficult because of the small numbers of studies that met inclusion criteria on each specific outcome, and poor study quality. A major weakness of these studies generally was failure to control for any confounding factors.

Overall, the studies examining other possible negative effects provide insufficient evidence on any particular outcome to permit confident conclusions. Further research in these areas needs to be of a much higher quality and should address and use appropriate methods to control for confounding factors.

Objective 5:

The assessment of natural versus artificial water fluoridation effects is greatly limited due to the lack of studies making this comparison. Very few studies included both natural and artificially fluoridated areas, and direct comparisons were not possible for most outcomes. No major differences were apparent in this review, however, the evidence is not adequate to make a conclusion regarding this objective.

Conclusions

This review presents a summary of the best available and most reliable evidence on the safety and efficacy of water fluoridation.

Given the level of interest surrounding the issue of public water fluoridation, it is surprising to find that little high quality research has been undertaken. As such, this review should provide both researchers and commissioners of research with an overview of the methodological limitations of previous research conducted in this area.

The evidence of a benefit of a reduction in caries should be considered together with the increased prevalence of dental fluorosis. The research evidence is of insufficient quality to allow confident statements about other potential harms or whether there is an impact on social inequalities. This evidence on benefits and harms needs to be considered along with the ethical, environmental, ecological, costs and legal issues that surround any decisions about water fluoridation. All of these issues fell outside the scope of this review.

Any future research into the safety and efficacy of water fluoridation should be carried out with appropriate methodology to improve the quality of the existing evidence base.

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1. Was the document clear and transparent in its presentation of data and explanation of the analytical approaches used to characterize the concentration- response and dose-response relationships for severe dental fluorosis? If not, do you have any suggestions that will enable the authors to improve its clarity?

Answer:

Yes, the document was clear and transparent. No, I have no suggestions to improve its clarity.

2. Are you aware of any significant publications related to severe dental fluorosis or the skeletal effects of fluoride that are not included in the noncancer assessment document?

Answer:

The report by Dean and Elvove is relevant to question 3. The reference is:
Dean HT, Elvove E. Studies on the minimal threshold of the dental sign of chronic endemic fluorosis (Mottled enamel). Pub Hlth Rep 50: 1719-1729, 1935.

3. Are the strengths and weaknesses of Dean (1942) study (selected as critical) fully characterized?

Answer:

Not entirely. One weakness of the Dean (1942) report was the chemical method used for the determination of fluoride concentrations in water. The zirconium-alizarin method is rarely used today because of its relative insensitivity, several interfering substances and lack of specificity for fluoride. For the SPANDS colorimetric method, interfering substances include Al (0.1 ppm) and Fe (10 ppm) which reduce absorbance and phosphorus (16 ppm) and sulfate (200 ppm) which increase absorbance. I'm not sure how these ions affect the zirconium-alizarin colorimetric method but Elvove's earlier papers should provide the information.

An indication of the problem is found in the footnote to Table 3-1 on page 24 where it is said (quoting Elvove who was the principal chemist) that "as little as 0.01 mg F/50cc, or 0.2 ppm F, could be differentiated from the control by application of this technique." This appears to mean that Elvove's method could differentiate between water without fluoride and water containing 0.2 ppm fluoride. The magnitude of the error at higher concentrations is not known to me but could be similar. Again, Elvove's earlier papers should provide the information.

One indication of the scatter in analytical results can be seen in the 1933/34 monthly results for water in Colorado Springs (see Table 4 in Dean and Elvove, Studies on the minimal threshold of the dental sign of chronic endemic fluorosis (mottled enamel), Public Hlth Rep 50: 1719-1729, 1935). The average of the 12 results was 2.5 ppm but the range was 1.8 to 3.0 ppm despite the fact that the water came from a single source. While some seasonal variation in water concentrations can expected, this wide range (1.2 ppm) appears excessive. The 12 monthly results from Monmouth ranged from 1.6 to 1.9 ppm, those from Galesburg ranged from 1.8 to 2.0 ppm, and those from Pueblo ranged from 0.3 to 0.7 ppm.

4. Are some teeth more susceptible to sever fluorosis than others? If so, should the OW make modifications to the age range identified as the period of concern?

Answer:

I am not aware of data showing that some teeth more susceptible to severe fluorosis than others. I would guess that the posterior teeth may be more susceptible since their development is more protracted than that of the anterior teeth.

5. Are the data on cavities as collected and graphically presented by the OW consistent with the hypothesis that there is an increased risk for cavities in susceptible individuals with severe dental fluorosis?

Answer:

The available data present an unclear relationship. Driscoll's work (1983, 1986) in Illinois and Iowa did not indicate a relationship between severe fluorosis and caries (pages 46-47). Eklund's report (1987) did not find a relationship when the entire dentition was considered. They found more caries in severely fluorosed anterior teeth and premolars but not in the molars. In their Chinese study, Chen et al (1989) found no difference in caries scores between the group without fluorosis and the group with severe fluorosis. Warnakulasuriya et al (1992) reached a similar conclusion in their Sri Lanka study but the validity of the conclusion was less clear because of the way they grouped the fluorosis categories. On the other hand, Mann et al (1987, 1990) and Olsson (1979) found that DMFS scores were directly related to the severity of fluorosis in Israel as did Wondwossen et al (2004) in Ethiopia. Ermis et al (2003) reported a slightly higher prevalence of caries in moderate-to-severely fluorosed teeth but the relationship was not significant.

Overall and as summarized in Figure 3-7 on page 71, the relationship between the severity of dental fluorosis and the risk of caries is suggestive but not convincing. I think this subject requires more study with control for variables that are known risk factors for caries before a reasonably firm conclusion can be drawn about an increased risk of caries associated with severe dental fluorosis.

6. Are there recent data that would impact the IOM (1997) Adequate Intake value of 0.05 mg/kg/day for fluoride that the OW should consider in this assessment?

Answer:

The IOM's Adequate Intake (AI) represents the amount of intake of any substance "needed to maintain a defined nutritional state or criterion of adequacy in essentially all members of a specific healthy population." In the case of fluoride, the AI "is based on estimated intakes that have been shown to reduce the occurrence of dental caries maximally in a population without causing unwanted side effects including moderate dental fluorosis (page 301, Dietary Reference Intakes, 1997). This does not mean that intakes somewhat higher than 0.05 mg/kg/day increase risk of moderate dental fluorosis. In fact, the IOM's estimate for the threshold for that risk is 0.10 mg/kg/day. I am not aware of data that would call for a change in the AI of 0.05 mg/kg F/day.

7. Can you suggest an approach to transform the water concentration data from the Dean (1942) study to units of dose for the population susceptible to severe dental fluorosis other than that used by the OW?

Answer: Not at this time.

8. Can you provide input concerning the strengths and weaknesses of the approach utilized by the OW to identify a lower bound dose for severe dental fluorosis?

Answer:

I am looking at the Clovis, New Mexico, data in Table 3-1 (page 24). The water fluoride concentration was listed as 2.2 ppm, the lowest concentration at which severe dental fluorosis was recorded. The prevalence was 0.7 percent. This concentration, therefore, was selected as the LOAEL. The prevalence of moderate dental fluorosis (Dean score 3) was 11.0 percent. These prevalence values are markedly higher than those for Elmhurst and Galesburg, Illinois, (about 1.1 percent for moderate and an absence of severe dental fluorosis) where the water fluoride concentrations were listed as 1.8 and 1.9 ppm, respectively, just slightly lower than the concentration in Clovis.

In view of the small differences in the water fluoride concentrations between Clovis and the other two communities, the large differences in fluorosis prevalence values suggest that another factor influenced the appearance of the teeth in Clovis. Unlike Elmhurst and Galesburg, Clovis is located at a relatively high altitude (4300 feet). As summarized elsewhere in the document (pages 39-41), there is evidence from laboratory animal studies and epidemiological studies that residence at high altitude affects amelogenesis in a way that resembles fluorosis and that its effects may be additive to the effects of fluoride exposure. This adds uncertainty regarding the selection of 2.2 ppm fluoride (Clovis) as the LOAEL for severe dental fluorosis.

A similar (but weaker) argument can be made for Colorado Springs where the average water fluoride concentration is listed as 2.6 ppm (but with a wide range, see item 3 above). This city is located at an altitude of 6,035 feet.

In addition to these comments and based on the variability among published studies regarding the relationship between estimated fluoride intakes and the risk of severe dental fluorosis, I think consideration should be given to establishing a LBD-5% rather than a LBD-1%.

9. Are you aware of dose estimates other than those from IOM (1997) and WHO (2002) that are appropriate critical doses for skeletal effects and/or can you suggest a different approach that the OW might use to estimate the fluoride dose associated with skeletal fractures using available data?

Answer:

The IOM (page 307) estimate of fluoride exposures that may result in clinical signs of the milder forms skeletal fluorosis (presumably preclinical and stage I) is 10 mg/day for 10 or more years. There are published exceptions as noted on the same page of the IOM report. Recent case reports of “tea fluorosis” in the U.S. suggest that, at least for some individuals, a much higher chronic intake is tolerated without progression to stage II skeletal fluorosis (Whyte et al, Am J Med 118: 78-82, 2005; Whyte et al, J Bone Min Res, in press). In the former report the intake was estimated at 37-74 mg F/day from tea throughout the patient’s adult life. The intake in the latter report was estimated at more than 40 mg F/day throughout the patient’s adult life. Both patients showed marked osteosclerosis without ligamentous calcifications consistent with stage I skeletal fluorosis. Hallanger-Johnson et al (Mayo Clin Proc 82: 719-724, 2007) reported four cases with axial osteosclerosis with elevated serum fluoride levels due to chronic consumption of large amounts of tea.

In addition to the several variables that can affect the quality and quantity of the skeleton cited in the present document, it is of interest that much of the data relating bone fluoride concentrations to the stages of skeletal fluorosis comes from studies of workers in aluminum

processing factories. High, chronic exposures to aluminum lead to skeletal changes that share some features in common with skeletal fluorosis which makes it difficult to attribute the skeletal changes only to fluoride. I think this subject is worthy of further exploration.

10. Are you aware of any data that can be used to demonstrate that protection of the secondary teeth from severe dental fluorosis will also protect the primary teeth?
Answer:

There are a few reports that suggest that dental fluorosis in the primary teeth may correlate with the condition in secondary teeth. I am not aware of reports suggesting that protection of the secondary teeth from severe dental fluorosis will also protect the primary teeth.

11. Are there factors other than those discussed in Section 3.1.4 that increase sensitivity to fluoroisis of the teeth or bones?
Answer:

As I recall, there are suggestions that African-Americans are more susceptible to dental fluorosis but less susceptible to skeletal fluorosis. Also, published data indicate that there are differences among strains of mice regarding susceptibility to dental fluorosis. It is assumed that the differences are due to genetics. NIDCR has requested applications for further pharmacogenetic studies.

12. Do you support the OW's conclusion that an RfD of 0.07 mg/kg/day will be protective for severe dental fluorosis in children and skeletal effects in adults while still providing for the beneficial effects of fluoride?
Answer:

Yes, 0.07 mg/kg/day will be protective. For reasons listed above, however, I think there is a considerable degree of uncertainty regarding the accuracy of the estimate. It may be closer to 0.10 mg/kg/day for moderate/severe dental fluorosis and substantially higher than that for clinically significant skeletal fluorosis in the United States.

Appendix B: Post-Meeting Responses to the Charge Questions from Reviewers (Organized Alphabetically by Reviewer)

Jane A. Cauley, Dr.P.H
Professor, Department of Epidemiology, University of Pittsburgh

1. Was the document clear and transparent in its presentation of data and explanation of the analytical approaches used to characterize the concentration- response and dose-response relationships for severe dental fluorosis? If not, do you have any suggestions that will enable the authors to improve its clarity?

Response

The document could be strengthened by reorganizing it as a manuscript. The Introduction should provide background and a scientific rationale for this analysis. The specific objectives should be listed.. The methods for identifying all the literature should be transparent and consistently applied to both the fluorosis and fracture sections. This is typical of meta analyses which also apply certain quality scores to each paper.
I think it might also be helpful if I understand who the audience for this report isd . I found it a confusing because I do not understand the roles of the EPA, OW and the NRC : how will each agency find this information helpful.? What prompted this report?

2. Are you aware of any significant publications related to severe dental fluorosis or the skeletal effects of fluoride that are not included in the noncancer assessment document?

Response

The dental fluorosis literature included studies with a wide range of fluoride levels while the skeletal/ fracture studies were limited to those with fluoride levels $\geq$ 4mg/l. You might consider adding some of the literature on fractures at lower levels of fluoride including the following:

Cauley JA, Murphy PA, Riley TJ, Buhari AM. Effects of fluoridated drinking water on bone mass and fractures: the study of osteoporotic fractures. J Bone Miner Res. 1995 Jul;10(7):1076-86.

Phipps, KR, Orwoll, ES, Mason, JD, Cauley, J A. Community water fluoridation, bone mineral density, and fractures: prospective study of effects in older women. BMJ 2000 321(7265):860-4.

In addition, Phipps KR et al published a study comparing bone mineral density in Deming, NM (0.7mg/L) and Lordsburg, NM (3.5mg/L). The citation is: Phipps KR, Burt BA. Water-borne fluoride and cortical bone mass: a comparison of two communities. J Dent Res. 1990 Jun;69(6):1256-60.

It appeared to me that this section is overall less thorough than the dental flourosis section. Additional Selections include the following. I would pay particular attention to the papers by Kathy Phipps, Cyrus Cooper, S Jacobsen and T Hillier. These are high quality in my opinion.

Demos LL, Kazda H, Cicuttini FM, Sinclair MI, Fairley CK.
Water fluoridation, osteoporosis, fractures--recent developments.
Aust Dent J. 2001 Jun;46(2):80-7; quiz 143.
PMID: 11491235 [PubMed - indexed for MEDLINE]

Fabiani L, Leoni V, Vitali M.
Bone-fracture incidence rate in two Italian regions with different fluoride concentration levels in drinking water.
J Trace Elem Med Biol. 1999 Dec;13(4):232-7.
PMID: 10707346 [PubMed - indexed for MEDLINE]

Kleerekoper M.
Fluoride and the skeleton.
Crit Rev Clin Lab Sci. 1996 Apr;33(2):139-61. Review.
PMID: 8744520 [PubMed - indexed for MEDLINE]

Raheb J.
Water fluoridation, bone density and hip fractures: a review of recent literature.
Community Dent Oral Epidemiol. 1995 Oct;23(5):309-16. Review. No abstract available.
PMID: 8529346 [PubMed - indexed for MEDLINE]

Søgaard CH, Mosekilde L, Schwartz W, Leidig G, Minne HW, Ziegler R.
Effects of fluoride on rat vertebral body biomechanical competence and bone mass.
Bone. 1995 Jan;16(1):163-9.
PMID: 7742076 [PubMed - indexed for MEDLINE]

Kröger H, Alhava E, Honkanen R, Tuppurainen M, Saarikoski S.
The effect of fluoridated drinking water on axial bone mineral density--a population-based study.
Bone Miner. 1994 Oct;27(1):33-41.
PMID: 7849544 [PubMed - indexed for MEDLINE]

Ripa LW.
A half-century of community water fluoridation in the United States: review and
commentary.
J Public Health Dent. 1993 Winter;53(1):17-44. Review.
PMID: 8474047 [PubMed - indexed for MEDLINE]

Gordon SL, Corbin SB.
Summary of workshop on drinking water fluoride influence on hip fracture on bone
health. (National Institutes of Health, 10 April, 1991)
Osteoporos Int. 1992 May;2(3):109-17. No abstract available.
PMID: 1627897 [PubMed - indexed for MEDLINE]

Colquhoun J.
Water fluoride and fractures.
N Z Med J. 1991 Aug 14;104(917):343. No abstract available.
PMID: 1876343 [PubMed - indexed for MEDLINE]

McNeill KG, Coote GE, Hitchman AJ.
Uptake of fluorine in cortical and trabecular bone.

J Bone Miner Res. 1991 Aug;6(8):859-64.
PMID: 1785375 [PubMed - indexed for MEDLINE]

Arnala I, Alhava EM, Kivivuori R, Kauranen P.
Hip fracture incidence not affected by fluoridation. Osteofluorosis studied in
Finland.
Acta Orthop Scand. 1986 Aug;57(4):344-8.
PMID: 3788501 [PubMed - indexed for MEDLINE]

Ericsson Y, Luoma H, Ekberg O.
Effects of calcium, fluoride and magnesium supplementations on tissue mineralization in calcium- and magnesium-deficient rats.
J Nutr. 1986 Jun;116(6):1018-27.
PMID: 3723198 [PubMed - indexed for MEDLINE]

Simonen O, Laitinen O.
Does fluoridation of drinking-water prevent bone fragility and osteoporosis?
Lancet. 1985 Aug 24;2(8452):432-4.
PMID: 2863455 [PubMed - indexed for MEDLINE]

Madans J, Kleinman JC, Cornoni-Huntley J.
The relationship between hip fracture and water fluoridation: an analysis of national data.
Am J Public Health. 1983 Mar;73(3):296-8.
PMID: 6824115 [PubMed - indexed for MEDLINE]

Schamschula RG, Barmes DE.
Fluoride and health: dental caries, osteoporosis, and cardiovascular disease.
Annu Rev Nutr. 1981;1:427-35. Review.
PMID: 6764723 [PubMed - indexed for MEDLINE]

Alhava EM, Olkkonen H, Kauranen P, Kari T.
The effect of drinking water fluoridation on the fluoride content, strength and
mineral density of human bone.
Acta Orthop Scand. 1980 Jun;51(3):413-20.
PMID: 7446020 [PubMed - indexed for MEDLINE]

Stein ID, Granik G.
Human vertebral bone: relation of strength, porosity, and mineralization to fluoride content.
Calcif Tissue Int. 1980;32(3):189-94.
PMID: 6775787 [PubMed - indexed for MEDLINE]

Hegsted DM.
Osteoporosis and fluoride deficiency.
Postgrad Med. 1967 Jan;41(1):A49-53. No abstract available.
PMID: 6036214 [PubMed - indexed for MEDLINE]

Bernstein DS, Sadowsky N, Hegsted DM, Guri CD, Stare FJ.
Prevalence of osteoporosis in high- and low-fluoride areas in North Dakota.
JAMA. 1966 Oct 31;198(5):499-504. No abstract available.
PMID: 5953273 [PubMed - indexed for MEDLINE]

Hillier S, Cooper C, Kellingray S, Russell G, Hughes H, Coggon D.
Fluoride in drinking water and risk of hip fracture in the UK: a case-control
study.
Lancet. 2000 Jan 22;355(9200):265-9.
PMID: 10675073 [PubMed - indexed for MEDLINE]

Hillier S, Inskip H, Coggon D, Cooper C.
Water fluoridation and osteoporotic fracture.
Community Dent Health. 1996 Sep;13 Suppl 2:63-8. Review.
PMID: 8897754 [PubMed - indexed for MEDLINE]

Jacobsen SJ, Goldberg J, Cooper C, Lockwood SA.
The association between water fluoridation and hip fracture among white women and
men aged 65 years and older. A national ecologic study.
Ann Epidemiol. 1992 Sep;2(5):617-26.
PMID: 1342313 [PubMed - indexed for MEDLINE]

Cooper C, Wickham C, Lacey RF, Barker DJ.
Water fluoride concentration and fracture of the proximal femur.
J Epidemiol Community Health. 1990 Mar;44(1):17-9.
PMID: 2348142 [PubMed - indexed for MEDLINE]

1: Phipps KR, Orwoll ES, Mason JD, Cauley JA.
Community water fluoridation, bone mineral density, and fractures: prospective
study of effects in older women.
BMJ. 2000 Oct 7;321(7265):860-4.
PMID: 11021862 [PubMed - indexed for MEDLINE]

Phipps KR, Orwoll ES, Bevan L.
The association between water-borne fluoride and bone mineral density in older
adults.
J Dent Res. 1998 Sep;77(9):1739-48.
PMID: 9759671 [PubMed - indexed for MEDLINE]

Phipps K.
Fluoride and bone health.
J Public Health Dent. 1995 Winter;55(1):53-6. Review.
PMID: 7776293 [PubMed - indexed for MEDLINE]

Phipps KR, Burt BA.
Water-borne fluoride and cortical bone mass: a comparison of two communities.
J Dent Res. 1990 Jun;69(6):1256-60.
PMID: 2355118 [PubMed - indexed for MEDLINE]

3. Are the strengths and weaknesses of Dean (1942) study (selected as critical) fully characterized?

Response:

Additional strengths include the wide range of fluoride concentrations, although there were fewer subjects in the high fluoride categories. The dose response in the data showing

increasing risk of severe skeletal fluorosis with increasing fluoride is also a strength. Finally the consistency of the findings across several different communities.

Other weaknesses: No information is provided on race/ethnicity. The data were collected the late 1930-40s. Although confounding by use of other fluoride products would be minimal, there are many other cohort differences between children exposed to fluoride in the late 1930-40s compared to now. For example, dental hygiene, dietary intakes (less water… more carbonated beverages), body weight are very different in today's children compared to children in the 30s.. Do puberty/ hormonal changes influence fluoride effects? This may be important because age of menarche has been decreasing. Hormones have been shown to influence dental health. Another weakness of the studies reviewed is that there appears to be no information on duration of exposure. How long did these children live in each community? Did the inclusion criteria include a minimum time of residence? If so this point needs to be made in the document.

4. Are some teeth more susceptible to sever fluorosis than others? If so, should the OW make modifications to the age range identified as the period of concern?

Response

This should be highlighted in a separate section. A more direct discussion of the age at risk is needed. A table summarizing the ages of the children in each study and the age range which appeared to be at highest risk would be helpful. You could add this to Table 3-16.

5. Are the data on cavities as collected and graphically presented by the OW consistent with the hypothesis that there is an increased risk for cavities in susceptible individuals with severe dental fluorosis?

Response

The data summarized in Table 3-40 to 3-44 are convincing that dental caries are more common in subjects with severe fluorosis. The data suggest a U shaped relationship with fluoride: a higher risk in those with very low intakes and intakes >2-3mg/L.

6. Are there recent data that would impact the IOM (1997) Adequate Intake value of 0.05 mg/kg/day for fluoride that the OW should consider in this assessment?

Response

I do not know of any more recent data. Perhaps you want to mention life stage and Upper limit of toxicity. The ADA website looks like they updated their Fluoride document in 2006.

TABLE S-5
Criteria and Dietary Reference Intake Values for Fluoride by Life Stage Group

Life Stage Group		Criterion	AI (mg/day)[a]	
			Male	Female
	0 through 6 months	Human milk content	0.01	0.01
	7 through 12 months	Caries prevention	0.5	0.5
	1 through 3 years	Caries prevention	0.7	0.7
	4 through 8 years	Caries prevention	1	1
	9 through 13 years	Caries prevention	2	2
	14 through 18 years	Caries prevention	3	3
	19 through 30 years	Caries prevention	4	3
	31 through 50 years	Caries prevention	4	3
	51 through 70 years	Caries prevention	4	3
>	70 years	Caries prevention	4	3
Pregnancy				
<=	18 years	Caries prevention	--	3
	19 through 50 years	Caries prevention	--	3
Lactation				
<=	18 years	Caries prevention	--	3
	19 through 50 years	Caries prevention	--	3

[a]AI = Adequate Intake. For healthy infants fed human milk, AI is the mean intake. The observed estimate of nutrient intake that reduces the incidence of dental caries maximally in a group of healthy people. The AI is used if the scientific evidence is not available to derive an EAR. The AI is believed to cover their needs, but lack of data or uncertainty in the data prevent being able to specify with confidence the percentage of individuals covered by this intake.

TABLE S-6
Tolerable Upper Intake Levels (UL[a]), by Life Stage Group

Life Stage Group		Calcium (g/day)	Phosphorus (g/day)	Magnesium (mg/day)[b]	Vitamin D (µg/day)[c]	Fluoride
	0 through 6 months	ND[d]	ND	ND	25	0.7
	7 through 12 months	ND	ND	ND	25	0.9
	1 through 3 years	2.5	3	65	50	1.3
	4 through 8 years	2.5	3	110	50	2.2
	9 through 18 years	2.5	4	350	50	10
	19 through 70 years	2.5	4	350	50	10
>	70 years	2.5	3	350	50	10
Pregnancy						
<=	18 years	2.5	3.5	350	50	10
	19 through 50 years	2.5	3.5	350	50	10
Lactation						
<=	18 years	2.5	4	350	50	10
	19 through 50 years	2.5	4	350	50	10

[a]UL = the maximum level of daily nutrient intake that is likely to pose no risk of adverse effects to members of the healthy general population. Unless specified otherwise, the UL represents total intake from food, water, and supplements.

[b]The UL for magnesium represents intake from pharmacological agents only and does not include intake from food and water.

[c]As cholecalciferol. 1 µg cholecalciferol = 40 IU vitamin D.

[d]ND. Not determinable due to lack of data of adverse effects in this age group and concern with regard to lack of ability to handle excess amounts. Source of intake should be from food only in order to prevent high levels of intake.

7. Can you suggest an approach to transform the water concentration data from the Dean (1942) study to units of dose for the population susceptible to severe dental fluorosis other than that used by the OW?

Response

To estimate the RFD of 0.07 mg/Kg/day, the OW attempted to obtain detailed analysis of average body weights and water intakes during the time the Dean data was collected. Data from the Erchow and Cantor were used (1989). Why not use more generalizable data from NHANES at least for body weight? You could still use the water intake estimates from the Cantor paper.

8. Can you provide input concerning the strengths and weaknesses of the approach utilized by the OW to identify a lower bound dose for severe dental fluorosis?

Response

Strengths: Conservative estimates carried out with and without the outlier community; several different models were considered, sensitivity analysis showed that even eliminating three highest data points had little effect on the model goodness of fit or on the BMDL.

Weakness: Inherent weakness of the approach relate primarily to the weaknesses cited related to the parent Dean Study. You might consider adding a formal test for trend in data.

9. Are you aware of dose estimates other than those from IOM (1997) and WHO (2002) that are appropriate critical doses for skeletal effects and/or can you suggest a different approach that the OW might use to estimate the fluoride dose associated with skeletal fractures using available data?

Response

See answer to #2 for additional references.

Some concern about the inconsistencies across the age groups. The significance of the differences in results across age could have occurred by chance alone.

Referring to the" radius" is problematic because if it s the distal radius, it is primarily trabecular BMD and mid radius is predominantly cortical BMD. If the effects of fluoride differs by type of bone, this is very problematic.

Randomized trials: please note the compliance/adherence to the Na F because it is poorly tolerated. This is a limitation to theses studies. Did they adhere to the intent to treat principle?

10. Are you aware of any data that can be used to demonstrate that protection of the secondary teeth from severe dental fluorosis will also protect the primary teeth?

Response

No, I am not aware of any additional data.

11. Are there factors other than those discussed in Section 3.1.4 that increase sensitivity to fluoroisis of the teeth or bones?

Response

I am not sure but I question whether race/ ethnicity and pubertal stage is important.

13. Do you support the OW's conclusion that an RfD of 0.07 mg/kg/day will be protective for severe dental fluorosis in children and skeletal effects in adults while still providing for the beneficial effects of fluoride?

Response

Yes, I am in support of the 0.07 mg/kg/day but I would admit the uncertainty that surrounds this estimate..

Pamela Denbesten, D.D.S., M.S.
Professor and Chair, Division of Pediatric Dentistry, Department of Orofacial Sciences, University of California at San Francisco

1. Was the document clear and transparent in its presentation of data and explanation of the analytical approaches used to characterize the concentration- response and dose-response relationships for severe dental fluorosis? If not, do you have any suggestions that will enable the authors to improve its clarity?

Response:
The document was for the most part clear and transparent in its presentation. To improve clarity, I suggest the following.

Page 13. State the purpose of the Secondary Maximum Contaminant Level

Page 15. The statement with regards to fluoroapatite, at the end of paragraph 2.1.1 is not accurate. The mineral formed in tooth enamel exposed to higher fluoride levels is fluoride containing carbonated apatite. Fluoride levels in subsurface fluorotic enamel are about 200 ppm rather than the 10-100 ppm fluoride in normal enamel, whereas fluorapatite is about 30,000 ppm fluoride. Precipitation of fluoride mineral salts at the surface of enamel results in high surface level, though this also is not fluoroapatite. This fluoride-substituted apatite has some increased resistance to bacterial acids that cause tooth decay. However, the primary function of fluoride in drinking water in reducing tooth decay is topical, primarily by the enhancement of remineralization.

Page 19 section 2.2. The apatite formed in bone is also a fluoride-substituted hydroxyapatite rather than a fluoroapatite.

Page 21 section 3. Change "…are preferred for evaluating the potential effects of fluoride in drinking water", to the more accurate statement, "…are preferred for evaluating the potential effects of ingested fluoride".

Page 57 paragraph 1. It should be made clear that studies on water fluoridation conducted after 1980, are confounded by additional sources of fluoride, and changes in use of tap water. For example, decreasing fluorosis in more recent studies may be related to reduced consumption of tap water as use of bottled water increases. In general for all of section 3.2.2, when fluoride effects on dental caries are discussed, the data should be divided into studies before and after 1980 when fluoride became widely available in toothpaste, and perhaps before and after the late 1990s when bottled beverages became widely used.

Page 86; Table 3-52. Please indicate what does "complete" and "total" refer to?

Page 94 section 4.4. Explain with the "NOAEL/LOAEL" approach is, or at least spell out the acronym.

Page 97 paragraph 1. The importance of fluoride as a nutrient may need to be reassessed, given that its primary function in caries prevention is topical. It would seem more appropriate

focus on the upper limits for ingestion of this caries preventive agent, and leave it to future panels to assess the relative importance of the IOMs recommended intake of fluoride and risk of severe fluorosis.

Page 98, paragraph 2, sentence 3. The statement as to the timing secondary incisor tooth formation is incorrect. The secondary incisors and molars begin development in utero. Change "development" to "mineralization".

Page 101. What is the rationale for setting the BMDL at an incidence of 1% severe fluorosis? I recommend setting the BMDL at an incidence of 99% moderate fluorosis, which would show an intent to eliminate the adverse effect of severe fluorosis secondary to fluoride added to drinking water.

Page 103. As stated above, I question a recommended fluoride intake, and feel that this document should focus only on the dose response analysis.

Page 104, paragraph 1. The statement that "…fluoroapatite crystals disrupt the hydroxyapatite crysal lattice…" is incorrect and should be deleted.

2. Are you aware of any significant publications related to severe dental fluorosis or the skeletal effects of fluoride that are not included in the noncancer assessment document?

Response:
No I am not

3. Are the strengths and weaknesses of Dean (1942) study (selected as critical) fully characterized?

Response:
Yes, to me they are.

4. Are some teeth more susceptible to severe fluorosis than others? If so, should the OW make modifications to the age range identified as the period of concern?

Response:
The OW report was clear in showing that posterior teeth are also susceptible, and the case for including children up to age 14 was clear and compelling.

5. Are the data on cavities as collected and graphically presented by the OW consistent with the hypothesis that there is an increased risk for cavities in susceptible individuals with severe dental fluorosis?

Response:
Yes
6. Are there recent data that would impact the IOM (1997) Adequate Intake value of 0.05 mg/kg/day for fluoride that the OW should consider in this assessment?

Response.
The weight of evidence is that the primary mechanism by which fluoride protects against tooth decay is a topical effect. Therefore, the recommendation by the IOM that there is an adequate intake value, at least relating to tooth decay should be reassessed.

7. Can you suggest an approach to transform the water concentration data from the Dean (1942) study to units of dose for the population susceptible to severe dental fluorosis other than that used by the OW?

Response:
No I can not. Of course serum fluoride levels would be the most useful measurement, but these levels are not available. I believe the using Dean's data as a starting point to quantify total ingestion is the best that we can do. I suggest a recommendation that future studies include random sampling of serum fluoride levels to strengthen future decision-making relative to fluoride intake.

8. Can you provide input concerning the strengths and weaknesses of the approach utilized by the OW to identify a lower bound dose for severe dental fluorosis?

Response:

Strengths.
1) The data was used limited to Dean's studies where fluoride was only in drinking water.
2) A careful assessment was done of the tap water consumed and mean body weights

Weaknesses
1) The assumption that 0.05 mg/kg/day is a required amount of fluoride and that dose estimates must be above this level. The purpose of the EPA's analysis was to determine risk, not risk benefit analyses.

2) The assumption that the small number of children who displayed severe fluorosis were those who had excess exposure to fluoride. Given the fact that water was the only source of fluoride, this would assume that these children drank significantly more water. It is more likely that genetic or other causes are responsible for this small outlier group.

9. Are you aware of dose estimates other than those from IOM (1997) and WHO (2002) that are appropriate critical doses for skeletal effects and/or can you suggest a different approach that the OW might use to estimate the fluoride dose associated with skeletal fractures using available data?

Response:
No I can not.

10. Are you aware of any data that can be used to demonstrate that protection of the secondary teeth from severe dental fluorosis will also protect the primary teeth?

Response:
Fluorosis in both primary and permanent teeth is caused by ingested fluoride, and there is no data to suggest that primary teeth are more susceptible to fluorosis than permanent teeth. Therefore, measure that would protect permanent teeth would require limiting ingestion of fluoride. These same measures would protect primary teeth.

11. Are there factors other than those discussed in Section 3.1.4 that increase sensitivity to fluoroisis of the teeth or bones?

Response:
There seem to be individuals who are uniquely sensitive to the effects of fluoride on enamel formation. These individuals may have other more "hidden" enamel defects that are exacerbated by the effects of fluoride. Little is currently know as to why some individuals seem to be more fluoride sensitive.

12. Do you support the OW's conclusion that an RfD of 0.07 mg/kg/day will be protective for severe dental fluorosis in children and skeletal effects in adults while still providing for the beneficial effects of fluoride?

Response:
I do not agree with this recommendation as it is based on limiting severe fluorosis to 1% of the population, and the IOM's recommended adequate intake level. I suggest that the level be lowered to eliminate severe fluorosis. One percent of the population represents a relatively large number of individuals. These are the individuals who are most likely uniquely sensitive to fluoride. The data should analyzed without taking the IOM recommended adequate levels into account. A secondary analysis could then be done to include the IOM recommendations.

Richard D. Jackson, D.M.D.
Assistant Professor, Preventive and Community Dentistry, School of Dentistry, Indiana University, Oral Health Research Institute

1. Was the document clear and transparent in its presentation of data and explanation of the analytical approaches used to characterize the concentration- response and dose-response relationships for severe dental fluorosis? If not, do you have any suggestions that will enable the authors to improve its clarity?

I felt that the document clearly described what is available in the literature and presented the information in an understandable format. I am glad individual tables summarizing some of the studies were included for reference. As an aside, I would have preferred that the studies be described chronologically as opposed to alphabetically by author.

2. Are you aware of any significant publications related to severe dental fluorosis or the skeletal effects of fluoride that are not included in the noncancer assessment document?

Jackson R, Kelly S, Katz B, Brizendine E, Stookey G. Dental fluorosis in children residing in communities with different fluoride levels in the water: 33 month follow-up. Pediatric Dentistry 21:248-254.

Examination of 357 school-age children in 1994 from three Indiana communities with varying levels of fluoride in the water (0.2, 1.0 and 4.0 ppm). Severe fluorosis using the TSIF (score 5, 6 or 7) was noted in 9% of the children examined in the 4.0 ppm community. No scores of this magnitude were seen in the other two communities. Based on comparison to previous prevalence data in the same communities in 1992, the prevalence of fluorosis had increased in each community but primarily in the categories rated as a 1, 2 or 3 using the TSIF.

I believe I included this paper in the packet that I sent to Lisa shortly after the meeting was conducted.

3. Are the strengths and weaknesses of Dean (1942) study (selected as critical) fully characterized?

I think the strengths of the investigation conducted by Dean (1942) are fully characterized. I doubt that anyone would disagree that Dean (1942) has been and will continue to be a benchmark study in the dental literature as well as the much broader literature related to public health and epidemiology. It is true that Dean's study was performed when confounding fluoride sources were not available and thus in all probability gives a very clear picture of the prevalence of the relationship of fluoride ingestion and the subsequent development of dental fluorosis. However, one point of weakness that could be argued is that the population of subjects examined was not diverse in terms of racial or cultural characteristics. As it has been postulated that genetic factors may impact on the expression of dental fluorosis at identical levels of ingestion, the fact that the data was collected in what may have been an exclusively white population appears to limit its applicability for use as a benchmark. I was unable to find any information concerning the racial/ethnic composition of the children involved in this investigation.

4. Are some teeth more susceptible to severe fluorosis than others? If so, should the OW make modifications to the age range identified as the period of concern?

I was a little unclear as to the intent of the question. My initial reaction was to answer "No". I have not come across any literature that indicates that some teeth are more susceptible to the development of severe dental fluorosis assuming that the exposure is constant and taking into consideration how long the developing tooth is exposed to higher levels of fluoride. For example, maxillary third molars may present with greater evidence of severe fluorosis (pitting, staining, etc) based on the fact that they take longer to develop (Cc 12-16 years-of-age) and erupt as opposed to a maxillary central incisor (Cc 5 years-of-age).

5. Are the data on cavities as collected and graphically presented by the OW consistent with the hypothesis that there is an increased risk for cavities in susceptible individuals with severe dental fluorosis?

It appears that the data are consistent with and support this hypothesis. A point of discussion may be made concerning examiner bias in the methodology of these studies. It is obvious that it would be impossible to eliminate bias as the most severely affected teeth would be evident by the caries examiner when determining the DMFT/DMFS of the subject. I don't see any solution for this dilemma with the use of a single examiner or multiple examiners but it is something that should at least be mentioned as a possible confounder.

Jackson et al (1995) also examined children residing in communities with varying levels of fluoride in their public water supplies. No child in the negligibly-fluoridated community or the optimally-fluoridated community was found to have TSIF scores greater than 2. However, the DMFT and DMFS in the 7-10 year-olds residing in the 4X optimum community were not significantly different from those residing in the 1.0 ppm community. For 11-14 year-olds DMFS was significantly lower than in the negligibly fluoridated community.

6. Are there recent data that would impact the IOM (1997) Adequate Intake value of 0.05 mg/kg/day for fluoride that the OW should consider in this assessment?

I am not aware of any new data that could influence the value of 0.5 mg/kg/day as the AI value.

7. Can you suggest an approach to transform the water concentration data from the Dean (1942) study to units of dose for the population susceptible to severe dental fluorosis other than that used by the OW?

No, using the data available the approach seems to be logical. The problem, as I see it. is the US marketplace is constantly changing and ingestion of tap water continue to decline. The EPA (2004) report stated that bottled water accounted for only 13% of water consumption in the United States. More recent trade manufacturing data indicates that bottled water consumption in the United States exceeds this percentage by a wide margin and bottled water consumption may surpass tapwater consumption in the near future. Other published data suggest that among Hispanic individuals, tap water is commonly perceived as "unhealthy" and again bottled water is

consumed almost exclusively. Another point that should be further explored is the possible "halo effect" of imported foods and beverages into the United States and the fluoride content of these consumables.

8. Can you provide input concerning the strengths and weaknesses of the approach utilized by the OW to identify a lower bound dose for severe dental fluorosis?
I believe that the ideas brought up during the meeting may clarify things somewhat. I really didn't have any additional comments that I thought would benefit how to improve on the approach taken by the OW.

9. Are you aware of dose estimates other than those from IOM (1997) and WHO (2002) that are appropriate critical doses for skeletal effects and/or can you suggest a different approach that the OW might use to estimate the fluoride dose associated with skeletal fractures using available data?

I am not aware of additional dose estimates that would be appropriate for determining critical dosage levels and their possible skeletal effects. From my review of the literature and the discussion the day of the meeting I cant think of an alternative to the approach taken to estimate fluoride intake associated with skeletal fractures.

IOM.

Recommended total dietary fluoride intake

Age	Reference weight*		Adequate intake†	Tolerable upper intake§
	kg	lb	mg/day	mg/day
0–6 months	7	16	0.01	0.7
6–12 months	9	20	0.5	0.9
1–3 years	13	29	0.7	1.3
4–8 years	22	48	1.1	2.2
>9 years	40/76	88/166	2.0/3.8	10.0

*** Values based on data collected during 1988–1994 as part of the third National Health and Nutrition Examination Survey.**
† Intake that maximally reduces occurrence of dental caries without causing unwanted side effects, including moderate enamel fluorosis.
§ Highest level of nutrient intake that is likely to pose no risks for adverse health effects in almost all persons.
Source: Adapted from Institute of Medicine. Fluoride. In: Dietary reference intakes for calcium,phosphorus, magnesium, vitamin D, and fluoride. Washington, DC: National Academy Press,1997:288–313.

BONE FRACTURE AND BONE DEVELOPMENT PROBLEMS
There were 29 studies included on the association between bone fracture and bone development problems and water fluoridation. Other than fluorosis, bone effects (not including bone cancers) were the most studied potential adverse effect. These studies had a mean validity score of 3.4 out of 8. All but one study were of evidence level C. These studies included both cohort and ecological designs, some of which included analyses controlling for potential confounding factors. Observer bias could potentially play a role in bone fracture studies, depending on how the study is conducted.
The evidence on bone fracture can be classified into hip fracture and other sites because there are more studies on hip fracture than any other site. Using a qualitative method of

analysis (Figure 8.1), there is no clear association of hip fracture with water fluoridation. Theevidence on other fractures is similar. Overall, the findings of studies of bone fracture effects showed small variations around the 'no effect' mark. A meta-regression of bone fracture studies also found no association with water fluoridation.

10. Are you aware of any data that can be used to demonstrate that protection of the secondary teeth from severe dental fluorosis will also protect the primary teeth?

I am not aware of any data that supports this possibility.

11. Are there factors other than those discussed in Section 3.1.4 that increase sensitivity to fluorosis of the teeth or bones?

Section 3.1.4. states that climate and altitude of place of residence; dietary habits, nutritional status, physiological state, and certain pathological conditions may affect the occurrence and severity of fluoride-induced dental fluorosis. The section further mentions that certain genetic conditions such as amelogenesis imperfecta may lead to defective development of enamel. There have been studies (Vieira et al, 2005; Yan et al, 2007) that suggest, in mice, that there may be a varying genetic response to identical levels of fluoride ingestion. These changes have been identified in both tooth and bone formation. Vieira et al found that genetic influences have a direct bearing on the biomechanical properties of the teeth. Yan et al. found strain-specific effects of physiological level of fluoride with increased osetoclastogenesis in some mouse strains. While I was unable to find comparable human trials, this area should be further explored as the technological means become available.

A study by Hong et al (2004) appears to indicate that the use of amoxicillin could play a contributing role in the development of primary tooth fluorosis, especially for children exposed to lower levels of fluoride. I believe I sent a copy of this study to Lisa in the packet following the meeting.

12. Do you support the OW's conclusion that an RfD of 0.07 mg/kg/day will be protective for severe dental fluorosis in children and skeletal effects in adults while still providing for the beneficial effects of fluoride?

Based on the data that was presented and the rationales provided I think the OW does make a very convincing argument that 0.07mg/kg/day will be protective against the more severe forms of fluorosis in children and the skeletal effects in adults in a vast majority of the population. I don't think that a claim can be made that no single individual will be completely immune from the development of severe fluorosis even at this recommended RfD. Based on possible a possible genetic predisposition, I don't think that the possibility can be totally ruled out. As always my concern is not so much the amount of fluoride that is ingested through the public water supply but the other well-known sources of fluoride that have an additive effect to that derived from consuming fluoridated drinking water. Dentifrice/fluoridated mouthrinse ingestion by young children, bottled waters which are not tested for their fluoride content, possible inappropriate use of fluoride supplements, ingestion of foods especially imported from other countries are in my mind more likely to be the possible causative factors if severe dental fluorosis occurs. Because the literature contains so few cases of severe skeletal fluorosis occurring in the United States I

am more confident that the RfD of 0.7 mg/kg/day is well within an acceptable safety margin for minimizing the possible development of skeletal fluorosis.

References

Hong L, Levy S, Warren J, Bergus G, Dawson D, Wefel J, Broffitt B. Primary tooth fluorosis and amoxicillin use during infancy. Journal of Public Health Dentistry 64: 38-44 (2004).

Vieira A, Hanocock R, Eggertsson H, Everett E, Grynpas M. Tooth quality in dental fluorosis: genetic and environmental factors. Calcified Tissue International 76:17-25 (2005).

Yan D, Gurumurthy A, Wright M, Pfeiler T, Loboa E, Everett E. Genetic background influences fluoride's effects on osteoclstogenesis. Bone 41:1036-1044 (2007).

Jackson R, Kelly S, Katz B, Hull J, Stookey G. Dental fluorosis and caries prevalence in children residing in communities with different levels of fluoride in the water. Journal of Public Health Dentistry 88: 79-84 (1995).

A systematic review of public water fluoridation
NHS Centre for Review and Dissemination
University of York 2000
EXECUTIVE SUMMARY

This systematic review has been commissioned by the Chief Medical Officer of the Department of Health to 'carry out an up to date expert scientific review of fluoride and health' (Paragraph 9.20, Our Healthier Nation).

Overall, the aim has been to assess the evidence on the positive and negative effects of population wide drinking water fluoridation strategies to prevent caries. To achieve this aim five objectives were identified:

Objective 1: What are the effects of fluoridation of drinking water supplies on the incidence of caries?

Objective 2: If water fluoridation is shown to have beneficial effects, what is the effect over and above that offered by the use of alternative interventions and strategies?

Objective 3: Does water fluoridation result in a reduction of caries across social groups and between geographical locations, bringing equity?

Objective 4: Does water fluoridation have negative effects?

Objective 5: Are there differences in the effects of natural and artificial water fluoridation?

Methods

A search of 25 electronic databases (with no language restrictions) and the world-wide-web was undertaken. Relevant journals and indices were hand searched and attempts were made to contact authors for further information.

Quality inclusion criteria were based on a pre-defined hierarchy of evidence (A, B, and C). Studies of efficacy were included if they were of evidence level A or B. In order to allow the broadest search for evidence on potential adverse effects, studies of all levels of evidence were included. Objective specific inclusion criteria, based on selection of participants, intervention, outcomes assessed, and study design appropriate for a given objective were then applied. Study validity was formally assessed using a published checklist modified for this review (CRD Report 4, 1996).

Inclusion criteria were assessed independently by at least two reviewers. Extraction of data from, and validity assessment of, included studies was independently performed by two reviewers, and checked by a third reviewer. Disagreements were resolved through consensus.

Where the data were in a suitable format, measures of effect and 95% confidence intervals

(CI) were plotted. Heterogeneity was investigated by visual examination and statistically using the Q-statistic. Where no evidence of heterogeneity was found a meta-analysis was conducted to produce a pooled estimate of the measure of effect. Statistically significant heterogeneity was investigated using meta-regression. Multiple regression analysis was used to explore the relationship between fluoridation and fluorosis.

Results

214 studies met full inclusion criteria for one or more of the objectives. No randomised controlled trials of the effects of water fluoridation were found. The study designs used included 45 'before and after' studies, 102 cross-sectional studies, 47 ecological studies, 13 cohort (prospective or retrospective) studies and 7 case-control studies. Several studies were reported in multiple papers over a number of years.

Results by Objective

Objective 1

A total of 26 studies of the effect of water fluoridation on dental caries were found. For this objective, the quality of studies found was moderate (no level A studies). A large number of studies were excluded because they were cross-sectional studies and therefore did not meet the inclusion criteria of being evidence level B or above. All but three of the studies included were before-after studies, two included studies used prospective cohort designs, and one used a retrospective cohort design. All before-after studies located by the search were included. The most serious defect of these studies was the lack of appropriate analysis. Many studies did not present an analysis at all, while others only did simple analyses without attempting to control for potentially confounding factors. While some of these studies were conducted in the 1940's and 50's, prior to the common use of such analyses, studies conducted much later also failed to use methods that were commonplace at the time of the study.

Another defect of many studies was the lack of any measure of variance for the estimates of decay presented. While most studies that presented the proportion of caries-free children contained sufficient data to calculate standard errors, this was not possible for the studies that presented dmft/DMFT scores. Only four of the eight studies using these data provided estimates of variance.

The best available evidence suggests that fluoridation of drinking water supplies does reduce caries prevalence, both as measured by the proportion of children who are caries free and by the mean change in dmft/DMFT score. The studies were of moderate quality (level B), but of limited quantity. The degree to which caries is reduced, however, is not clear from the data available. The range of the mean difference in the proportion (%) of caries-free children is -5.0 to 64%, with a median of 14.6% (interquartile range 5.05, 22.1%). The range of mean change in dmft/DMFT score was from 0.5 to 4.4, median 2.25 teeth (interquartile range 1.28, 3.63 teeth). It is estimated that a median of six people need to receive fluoridated water for one extra person to be caries-free (interquartile range of study NNTs 4, 9). The best available evidence from studies following withdrawal of water fluoridation indicates that caries prevalence increases, approaching the level of the low fluoride group. Again, however, the studies were of moderate quality (level B), and limited quantity. The estimates of effect could be biased due to poor adjustment for the effects of potential confounding factors.

Objective 2

To address this objective, studies conducted after 1974 were examined. While only nine studies were included for Objective 2, these would have been enough to provide a confident answer to the objective's question if the studies had been of sufficient quality. Since these studies were completed after 1974, one might expect that the validity assessments would be higher than the earlier studies following the introduction of more rigorous study methodology and analytic techniques. However, the average validity checklist score and level of evidence was essentially the same for studies after 1974 as those conducted prior to 1974. Hence, the ability to answer this objective is similar to that in Objective 1.

In those studies completed after 1974, a beneficial effect of water fluoridation was still evident in spite of the assumed exposure to non-water fluoride in the populations studied. The metaregression conducted for Objective 1 confirmed this finding.

Objective 3

No level A or B studies examining the effect of water fluoridation on the inequalities of dental health between social classes were identified. However, because of the importance of this objective, level C studies conducted in England were included. A total of 15 studies investigating the association of water fluoridation, dental caries and social class in England were identified. The quality of the evidence of the studies was low, and the measures of social class that were used varied. Variance data were not reported in most of these studies, so a statistical analysis was not undertaken.

There appears to be some evidence that water fluoridation reduces the inequalities in dental health across social classes in 5 and 12 year-olds, using the dmft/DMFT measure. This effect was not seen in the proportion of caries-free children among 5 year-olds. The data for the effects in children of other ages did not show an effect. The small quantity of studies, differences between these studies, and their low quality rating, suggest *caution* in interpreting these results.

Objective 4

DENTAL FLUOROSIS

Dental fluorosis was the most widely and frequently studied of all negative effects. The fluorosis studies were largely cross-sectional designs, with only four before-after designs. Although 88 studies of fluorosis were included, they were of low quality. The mean validity score for fluorosis was only 2.8 out of 8. All, but one, of the studies were of evidence level C. Observer bias may be of particular importance in studies assessing fluorosis. Efforts to control for the effects of potential confounding factors, or reducing potential observer bias were uncommon.

As there may be some debate about the significance of a fluorosis score at the lowest level of each index being used to define a person as 'fluorosed', a second method of determining the proportion 'fluorosed' was selected. This method describes the number of children having dental fluorosis that may cause 'aesthetic concern'.

With both methods of identifying the prevalence of fluorosis, a significant dose-response relationship was identified through a regression analysis. The prevalence of fluorosis at a water fluoride level of 1.0 ppm was estimated to be 48% (95% CI 40 to 57) and for fluorosis of aesthetic concern it was predicted to be 12.5% (95% CI 7.0 to 21.5). A very rough estimate of the number of people who would have to be exposed to water fluoride levels of 1.0 ppm for one additional person to develop fluorosis of any level is 6 (95% CI 4 to 21), when compared with a theoretical low fluoride level of 0.4 ppm. Of these approximately one quarter will have fluorosis of aesthetic concern, but the precision of these rough estimates is low. These estimates only apply to the comparison of 1.0 ppm to 0.4 ppm, and would be different if other levels were compared.

BONE FRACTURE AND BONE DEVELOPMENT PROBLEMS

There were 29 studies included on the association between bone fracture and bone development problems and water fluoridation. Other than fluorosis, bone effects (not including bone cancers) were the most studied potential adverse effect. These studies had a mean validity score of 3.4 out of 8. All but one study were of evidence level C. These studies included both cohort and ecological designs, some of which included analyses controlling for potential confounding factors. Observer bias could potentially play a role in bone fracture studies, depending on how the study is conducted.

The evidence on bone fracture can be classified into hip fracture and other sites because there are more studies on hip fracture than any other site. Using a qualitative method of analysis (Figure 8.1), there is no clear association of hip fracture with water fluoridation. The evidence on other fractures is similar. Overall, the findings of studies of bone fracture effects showed small variations around the 'no effect' mark. A meta-regression of bone fracture studies also found no association with water fluoridation.

CANCER STUDIES

There were 26 studies of the association of water fluoridation and cancer included. Eighteen of these studies are from the lowest level of evidence (level C) with the highest risk of bias. There is no clear association between water fluoridation and overall cancer incidence and mortality. This was also true for osteosarcoma and bone/joint cancers. Only two studies considered thyroid cancer and neither found a statistically significant association with water

fluoridation.
Overall, no clear association between water fluoridation and incidence or mortality of bone cancers, thyroid cancer or all cancers was found.

OTHER POSSIBLE NEGATIVE EFFECTS

A total of 33 studies of the association of water fluoridation with other possible negative effects were included in the review. Interpreting the results of studies of other possible negative effects is very difficult because of the small numbers of studies that met inclusion criteria on each specific outcome, and poor study quality. A major weakness of these studies generally was failure to control for any confounding factors.
Overall, the studies examining other possible negative effects provide insufficient evidence on any particular outcome to permit confident conclusions. Further research in these areas needs to be of a much higher quality and should address and use appropriate methods to control for confounding factors.

Objective 5:

The assessment of natural versus artificial water fluoridation effects is greatly limited due to the lack of studies making this comparison. Very few studies included both natural and artificially fluoridated areas, and direct comparisons were not possible for most outcomes. No major differences were apparent in this review, however, the evidence is not adequate to make a conclusion regarding this objective.

Conclusions

This review presents a summary of the best available and most reliable evidence on the safety and efficacy of water fluoridation.
Given the level of interest surrounding the issue of public water fluoridation, it is surprising to find that little high quality research has been undertaken. As such, this review should provide both researchers and commissioners of research with an overview of the methodological limitations of previous research conducted in this area.
The evidence of a benefit of a reduction in caries should be considered together with the increased prevalence of dental fluorosis. The research evidence is of insufficient quality to allow confident statements about other potential harms or whether there is an impact on social inequalities. This evidence on benefits and harms needs to be considered along with the ethical, environmental, ecological, costs and legal issues that surround any decisions about water fluoridation. All of these issues fell outside the scope of this review.
Any future research into the safety and efficacy of water fluoridation should be carried out with appropriate methodology to improve the quality of the existing evidence base.

Gary M. Whitford, D.M.D., Ph.D.
Regents' Professor, Department of Oral Biology and Maxillofacial Pathology, Medical College of Georgia

1. Was the document clear and transparent in its presentation of data and explanation of the analytical approaches used to characterize the concentration- response and dose-response relationships for severe dental fluorosis? If not, do you have any suggestions that will enable the authors to improve its clarity?

Answer:

Yes, the document was clear and transparent.

2. Are you aware of any significant publications related to severe dental fluorosis or the skeletal effects of fluoride that are not included in the noncancer assessment document?

Answer:

Each of the following publications contains information pertinent to the reliability of the water fluoride concentrations shown in Dean (1942), the report selected as the "critical study for severe dental fluorosis."

1. Elvove E. Estimation of fluorides in waters. Pub Hlth Rep 48: 1219-1222, 1933.

2. Dean HT, Elvove E. Studies on the minimal threshold of the dental sign of chronic endemic fluorosis (Mottled enamel). Pub Hlth Rep 50: 1719-1729, 1935.

3. Megregian S, Maier FJ. Modified zirconium alizarin reagent for determination of fluoride in water. J Am Water Works Assn 44: 239-246, 1952.

I also recommend that a copy of the book entitled "Fluoride Drinking Waters" be consulted. It is a compilation of many of the early papers, including the three cited above, that dealt with several aspects of fluoride in water. The book is Public Health Service Publication No. 825. It was edited by Frank J. McClure and published in 1962. I am willing to loan my copy if the committee cannot locate the book.

3. Are the strengths and weaknesses of Dean (1942) study (selected as critical) fully characterized?

Answer:

Not entirely. A major weakness of the Dean (1942) report is the chemical method used for the determination of fluoride concentrations in water (Elvove E, 1933). The zirconium-alizarin method is rarely, or probably never, used today because of its relative insensitivity, several interfering substances and lack of specificity for fluoride. In their 1952 report that described improvements to the method, Megregian and Maier (1952) noted that Elvove's original method (1933) had several shortcomings including "non-conformity to the color laws, limited effective fluoride range, and little color change per increment of fluoride." It also appears that Elvove (1933) used the visual method to determine color changes in the zirconium-alizarin reagent (since he referred to "Nessler tubes") which requires subjective judgments and is less accurate that spectrophotometric methods.

Megregian and Maier (1952) also reported the effects of interfering substances on the analytical results. Sulfate at 400 ppm in the water increased the fluoride result by 0.1 ppm as did 1.1 ppm hexametaphosphate. Chloride at 1800 ppm, bicarbonate at 400 ppm and iron at 5 ppm decreased the fluoride result by 0.1 ppm. When the water fluoride concentration was 1.0 ppm, 1.0 ppm aluminum reduced the result by 0.39 ppm and 3.0 ppm aluminum reduced the result by

0.63 ppm. When the fluoride concentration was 2.0 ppm, aluminum at 1.0, 2.0 and 3.0 ppm reduced the results by 0.47, 0.86 and 1.13 ppm, respectively.

The report by Elvove (1933) that described the chemical method used to produce the water fluoride concentrations shown in Table I of Dean's 1942 report made no mention of interfering substances. It can be reasonably assumed that most, if not all, of the substances listed in the preceding paragraph were present in all the water samples analyzed but at unreported or unknown concentrations. However, in his original publication Elvove (1933) shows the concentrations of several ions in water samples obtained from 20 different sources. With the exception of Amarillo, these sources were not those shown in the Dean (1942) report. Among the interfering ions listed in the preceding paragraph, sulfate concentrations were more than 400 ppm in two of the 20 water samples, bicarbonate concentrations were more than 400 ppm in five, and aluminum concentrations were more than 1.0 ppm in five. At these concentrations, each of these ions would have affected the apparent water fluoride results.

It may be possible to access 1930-1940 analytical records from the communities shown in Table I (Dean, 1942) and make a judgment concerning the possible effects of the interfering substances of the reported water fluoride concentrations.

Another indication of the problem with the accuracy of the Elvove method is found in the footnote to Table 3-1 on page 24 of the EPA Dose-Response Analysis where it is said (quoting Elvove who was the principal chemist) that "as little as 0.01 mg F/50cc, or 0.2 ppm F, could be differentiated from the control by application of this technique." This appears to mean that Elvove's method could differentiate between water without fluoride and water containing 0.2 ppm fluoride. The magnitude of the error at higher concentrations is not known to me.

The scatter in the analytical results seen in the 1933/34 monthly results for water in Colorado Springs is of particular interest (see Table 4 in Dean and Elvove, 1935). The average of the 12 results was 2.5 ppm but the range was 1.8 to 3.0 ppm despite the fact that the water came from a single source. While some seasonal variation in water concentrations can expected, this wide range (1.2 ppm) appears excessive. Further, the 12 monthly results from Monmouth ranged from 1.6 to 1.9 ppm, those from Galesburg ranged from 1.8 to 2.0 ppm, and those from Pueblo ranged from 0.3 to 0.7 ppm.

Dean (1942, page 25) listed two major requisites for quantitative evaluation of the dental effects of ingesting water containing fluoride. One was "a population continuously exposed throughout life to the variable under investigation (the communal water supply)." In the Dean and Elvove (1935) paper, the first paragraph in the Discussion contains this sentence: "As a result of checking the water histories as given by the child by a followup recheck with the parents, only about 20 percent of the children in the age group studied and present in the school on the day of the examination were found to have had an unbroken history of residence and constant use of the city water supply." More details, such as how long and/or how frequently the children were away from the home water supply, were not given.

The studies summarized by Dean (1942) were done during or around the time of the Great Depression when large numbers of families were resettling in other locations in search of employment and other necessities. I recommend that the original papers summarized in Dean (1942) be examined to determine the extent to which the children met the requisite cited above and that the information be included in the Dose-Response Analysis document. If such information is not available, then the document should say so and discuss the implications for its conclusions.

Finally, I note that the appropriateness of the LOAEL (2.2 ppm) and the calculated RfD (0.07 mg F/kg bw/day) are based largely on the accuracy of the water fluoride concentrations shown in Dean (1942) but also on several other variables that may have affected the outcomes of the epidemiological studies. The preceding comments draw attention to several shortcomings of the chemical method used and other limiting aspects of the studies summarized by Dean (1942). The uncertainties associated with these factors should be discussed wherever appropriate and certainly in the "Uncertainty factors" section.

4. Are some teeth more susceptible to sever fluorosis than others? If so, should the OW make modifications to the age range identified as the period of concern?

Answer:

I am not aware of data showing that some teeth more susceptible to severe fluorosis than others. I would guess that the posterior teeth may be more susceptible since their development is more protracted than that of the anterior teeth.

5. Are the data on cavities as collected and graphically presented by the OW consistent with the hypothesis that there is an increased risk for cavities in susceptible individuals with severe dental fluorosis?

Answer:

The available data present an unclear relationship. Driscoll's work (1983, 1986) in Illinois and Iowa did not indicate a relationship between severe fluorosis and caries (pages 46-47). Eklund's report (1987) did not find a relationship when the entire dentition was considered. They found more caries in severely fluorosed anterior teeth and premolars but not in the molars. In their Chinese study, Chen et al (1989) found no difference in caries scores between the group without fluorosis and the group with severe fluorosis. Warnakulasuriya et al (1992) reached a similar conclusion in their Sri Lanka study but the validity of the conclusion was less clear because of the way they grouped the fluorosis categories. On the other hand, Mann et al (1987, 1990) and Olsson (1979) found that DMFS scores were directly related to the severity of fluorosis in Israel as did Wondwossen et al (2004) in Ethiopia. Ermis et al (2003) reported a slightly higher prevalence of caries in moderate-to-severely fluorosed teeth but the relationship was not statistically significant.

Overall and as summarized in Figure 3-7 on page 71, the relationship between the severity of dental fluorosis and the risk of caries is suggestive but not convincing. I think this subject requires more study with control for variables that are known risk factors for caries before a reasonably firm conclusion can be drawn about the possibility of an association between severe dental fluorosis and an increased risk of caries.

6. Are there recent data that would impact the IOM (1997) Adequate Intake value of 0.05 mg/kg/day for fluoride that the OW should consider in this assessment?

Answer:

The IOM's Adequate Intake (AI) represents the amount of intake of any substance "needed to maintain a defined nutritional state or criterion of adequacy in essentially all members of a specific healthy population." In the case of fluoride, the AI (see page 301, Dietary Reference Intakes, 1997) "is based on estimated intakes that have been shown to reduce the occurrence of dental caries maximally in a population without causing unwanted side effects including moderate dental fluorosis. This does not mean that intakes somewhat higher than 0.05 mg/kg/day

increase risk of moderate dental fluorosis. In fact, the IOM's estimate for the threshold for that risk is 0.10 mg/kg/day. I am not aware of data that would call for a change in the AI of 0.05 mg/kg F/day.

7. Can you suggest an approach to transform the water concentration data from the Dean (1942) study to units of dose for the population susceptible to severe dental fluorosis other than that used by the OW?
Answer: Not at this time.

8. Can you provide input concerning the strengths and weaknesses of the approach utilized by the OW to identify a lower bound dose for severe dental fluorosis?
Answer:

I am looking at the Clovis, New Mexico, data in Table 3-1 (page 24). The water fluoride concentration was listed as 2.2 ppm, the lowest concentration at which severe dental fluorosis was recorded. The prevalence was 0.7 percent, ie, only one subject out of the 138 examined exhibited what was classified as having severe fluorosis. This concentration, 2.2 ppm, was selected as the LOAEL. Unfortunately, nothing is known about the individual with severe fluorosis including whether he/she was a permanent resident of Clovis or had lived in one or more other communities before moving to Clovis. The prevalence of moderate dental fluorosis (Dean score 3) in Clovis was 11.0 percent. These prevalence values for moderate and severe fluorosis are markedly higher than those for Elmhurst and Galesburg (about 1.1 percent for moderate and an absence of severe dental fluorosis) where the water fluoride concentrations were listed as 1.8 and 1.9 ppm, respectively, just slightly lower than the concentration in Clovis.

In view of the small differences in the water fluoride concentrations between Clovis and the other two communities, the large differences in fluorosis prevalence values suggest that another factor may have influenced the appearance of the teeth in Clovis. Unlike Elmhurst and Galesburg, Clovis is located at a relatively high altitude (4300 feet). As summarized elsewhere in the EPA document under review (pages 39-41), there is evidence from laboratory animal studies and epidemiological studies that residence at high altitude affects amelogenesis in a way that resembles fluorosis and that its effects may be additive to the effects of fluoride exposure. This too adds uncertainty regarding the selection of 2.2 ppm fluoride (Clovis) as the LOAEL for severe dental fluorosis and the appropriateness of the RfD.

A similar (but weaker) argument can be made for Colorado Springs where the average water fluoride concentration is listed as 2.6 ppm (but with a wide range, see item 3 above). This city is located at an altitude of 6,035 feet.

In addition to these comments and based on the variability among published studies regarding the relationship between estimated fluoride intakes and the risk of severe dental fluorosis, I think consideration should be given to establishing a LBD other than a LBD-1%.

9. Are you aware of dose estimates other than those from IOM (1997) and WHO (2002) that are appropriate critical doses for skeletal effects and/or can you suggest a different approach that the OW might use to estimate the fluoride dose associated with skeletal fractures using available data?
Answer:

As in the case of dental fluorosis, the critical doses for skeletal effects will be difficult to establish with a reasonable degree of certainty. The IOM's estimate (page 307) of fluoride

exposures that may result in clinical signs of the "milder forms" of skeletal fluorosis (preclinical and perhaps stage I) is 10 mg/day for 10 or more years. There are published exceptions suggesting that higher exposure levels and durations are required as noted on the same page of the IOM report. Further, recent case reports of "tea fluorosis" in the U.S. suggest that, at least for some individuals, a much higher chronic intake is tolerated without progression to stage II skeletal fluorosis (Whyte et al, Am J Med 118: 78-82, 2005; Whyte et al, J Bone Min Res, in press). In the former report the intake was estimated at 37-74 mg F/day from tea throughout the patient's adult life. The intake in the latter report was estimated at more than 40 mg F/day throughout the patient's adult life. Both patients showed marked osteosclerosis but without ligamentous calcifications which was consistent with stage I skeletal fluorosis and neither patient experienced fractures. Hallanger-Johnson et al (Mayo Clin Proc 82: 719-724, 2007) reported four cases with axial osteosclerosis with elevated serum fluoride levels due to chronic consumption of large amounts of tea.

In addition to the several variables that can affect the quality and quantity of the skeleton cited in the present document, it is of interest that much of the data relating bone fluoride concentrations to the stages of skeletal fluorosis comes from studies of workers in aluminum processing factories. High, chronic exposures to aluminum lead to skeletal changes that share some features in common with skeletal fluorosis which makes it difficult to attribute the skeletal changes only to fluoride. This subject is worthy of further exploration.

10. Are you aware of any data that can be used to demonstrate that protection of the secondary teeth from severe dental fluorosis will also protect the primary teeth?

Answer:

There are a few reports that suggest that dental fluorosis in the primary teeth may correlate with the condition in secondary teeth. I am not aware of reports suggesting that protection of the secondary teeth from severe dental fluorosis will also protect the primary teeth.

11. Are there factors other than those discussed in Section 3.1.4 that increase sensitivity to fluorosis of the teeth or bones?

Answer:

As I recall, there are suggestions that African-Americans are more susceptible to dental fluorosis but less susceptible to skeletal fluorosis. Published data indicate that there are differences among strains of mice regarding susceptibility to dental fluorosis (Everett et al. J Dent Res 81: 794-698, 2002) and mechanical properties of bone (Mousny et al. Bone 39: 1283-1289, 2006). It is assumed that the differences are due to genetic differences among the strains. NIDCR has requested applications for further pharmacogenetic studies.

12. Do you support the OW's conclusion that an RfD of 0.07 mg/kg/day will be protective for severe dental fluorosis in children and skeletal effects in adults while still providing for the beneficial effects of fluoride?

Answer:

Yes. For reasons listed above, however, there is a considerable degree of uncertainty regarding the appropriateness of the estimate. As estimated by the IOM (1997), the RfD may be closer to 0.10 mg/kg/day for moderate (not severe) dental fluorosis and substantially higher than that for clinically significant skeletal effects in the United States.