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A Preliminary Evaluation of Articles Related to Fluoride Cited by the Fluoride Action Network (FAN) as Objections to the Sulfuryl Fluoride Pesticide Tolerance Rule

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A preliminary evaluation was conducted of articles cited by the Fluoride Action Network (FAN) as objections to the Sulfuryl Fluoride Pesticide Tolerance Rule (see Tables 1a,b). Additional articles received up until September 30, 2003 from FAN via email were also considered (see Attachment 3). The purpose of this evaluation was to determine and document which submitted studies are of sound scientific quality and included measurements of endpoints and dose effects that may be considered in evaluating potential human health risks and that may add new information regarding the adequacy of the current MCL for fluoride. The US MCL is 4 ppm based on skeletal fluorosis and a secondary standard (SMCL) of 2 ppm for dental fluorosis. EPA requires public notification at 2 ppm. The World Health Organization has recommended 1.5 ppm for dental fluorosis. Canada, Mexico, and the United Kingdom also uses the 1.5 ppm level for dental fluorosis. It should be noted that the EPA recently requested that the National Academy of Sciences review new toxicologic, epidemiologic and clinical data on fluoride and advise the Agency on the adequacy of the MCL and SMCL to protect children and others from adverse effects. The NAS review is anticipated in 2005. Thus, this current evaluation should not be interpreted as a comprehensive and detailed analysis/risk assessment. Rather the intent is to consider the studies submitted by the FAN and identify those that may raise concern for the adequacy of the current MCL for fluoride.

Hard copies of all citations listed in Tables 1a,b were obtained. In proceeding with this review, criteria were established by which to judge the science quality and merit of the studies (i.e., original data, sufficient description of study design and results, and inclusion of fluoride levels associated with potential human health effects). These criteria were applied in an initial screen. It was concluded from this screen that the majority of references cited by the FAN did not require a detailed review prior to completion of the NAS review. Papers fell in this category for a variety of reasons, which are listed below:

- Abstracts that lack sufficient detail on methods and results
- Studies that evaluated high doses of fluoride which were well above the current EPA MCL
- Papers that did not contain any data on fluoride
- Surveys on dental caries and water fluoridation (i.e., no data on dental fluorosis)
- *In vitro* studies that can not be used for dose-effect extrapolation to humans
- Laboratory animal studies using inappropriate routes of administration for dose-effect extrapolation
- Commentaries by others (e.g., on the benefits of water fluoridation or why dental fluorosis should be considered adverse)
- Papers on fluoride but not pertaining to health effects following fluoride exposure (e.g., chemistry papers)
- Exposure studies on fluoride with no toxicity or health effects data
- Studies with confounding factors that render results inconclusive

- Older literature previously evaluated and considered by NAS or EPA.
- Reviews or assessments of the fluoride literature conducted by other government or science organizations (e.g., by Canada, ATSDR, CDC, NRC, WHO) which did not contain original fluoride data.

Nine studies were identified that contained dose response data on fluoride. Five of these were human studies and four were studies conducted in laboratory animals. (summarized in attachments 1 and 2). Expert epidemiologists from EPA's National Health and Environmental Effects Laboratory and OPP's Health Effects Division were asked to review the epidemiology papers (Attachment 1), and senior toxicologists within OPP, OSCP, and OW reviewed the toxicology studies. These papers were on various health effects including the dental fluorosis and bone fractures, as well as other potential health outcomes such as neurological, thyroid, and reproductive effects. In general, due to methodological problems or lack of reproducibility of results, these studies would not contribute to evaluating the adequacy of the MCL. For example, the epidemiologic studies were found to be limited because of inadequate exposure assessment, potential biases associated with misclassification of exposure, limited statistical analysis, and/or internal inconsistencies.

With respect to the cited literature by FAN on health effects other than skeletal or dental fluorosis, there does not appear to be a sufficient scientific foundation to permit confident conclusions or estimates of potential health risk. This conclusion is consistent with the recent conclusions draw by the Medical Research Council (MRC, Working Group Report: Water Fluoridation and Health, 2002) and the York Review (McDonagh et al., The NHS Centre for Reviews and Dissemination, University of York, York England 2000). Although several papers deal with the effects of fluoride on thyroid functioning, overall there is no compelling information to indicate that fluoride produces significant effects on the thyroid. The MRC (2002) indicated that the studies on thyroid and fluoride effects needed to be treated with caution because of study design and report deficiencies and further work on this aspect is of a low priority. Bürgi et al.(1984 *Fluorine and thyroid gland function: A review of the literature*; Klin Wochenschr. 62:564-569; Not cited by FAN) summarizes much of the animal thyroid data, and the York review addresses the epidemiologic studies.

In addition, there is insufficient evidence to establish a causal link between fluoride exposure and effects on reproductive function in humans. Adverse effects on reproduction are primarily reported from animal studies (e.g., Reference #14 Chinoy et al. 1988). Although the Chinoy paper reported adverse effects in the testes, these effects were not reproduced by other laboratories[1] (articles not cited by FAN).

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[1]For example see: Collins TF, Sprando RL, et al, Multigenerational evaluation of sodium fluoride in rats Food Chem Toxicol. 2001 Jun;39(6):601-13.; Sprando RL, Collins TF, et al., Black TN, Rorie J, Ames MJ, O'Donnell M, Testing the potential of

Additionally, the recent Medical Research Council Working Group Report (MRC 2002), concluded that the "plausibility of fluoride affecting the reproductive capacity of humans at the intakes experience from fluoridated drinking water is low".

Although there are animal studies (e.g., Reference #58 Mullinix et al., 1995) that suggest an association between fluoride treatment and effect on the developing brain, more research is need to address the significance of fluoride exposure and neurological effects. Literature on fluoride exposure and reduction of children's intelligence are inconclusive or have not reported an association (Also, see York and MRC reviews). Effects reported on the pineal gland (Reference #52, Luke, 1997) are limited.

Conclusion

In summary, a preliminary evaluation of the articles cited by FAN listed in Attachments 1-3, did not heighten the concern that there is an association between exposure to fluoride and adverse effects on human health at the levels of exposure under consideration by OPP. Thus, the articles cited by FAN do not provide a compelling basis to depart from OPP's use of the current Agency MCL and secondary MCL in pesticide risk assessments at this time. This conclusion is supported by the recent York Review (2000) an d the Medical Research Council Working Group Report (2002). When the NAS review is available, EPA will revisit this conclusion.

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sodium fluoride to affect spermatogenesis in the rat., Food Chem Toxicol. 1997 Sep;35(9):881-90; Li Y, Dunipace and Stookey, Effects of fluoride on the mouse sperm morphology test, J Dent Res. 1987 Sep;66(9):1509-11.

ATTACHMENT 1

Reference #49: *Li, Yiming, Liang, Chaoke, Slemenda, C.W., Ji, R., Sun, S., Cao, J., Emsley, C.L., Ma, F., Wu, Y., Ying, P., Zhang, Y., Gao, S., Zhang, W., Katz, B.P., Niu, S., Cao, S., and Johnston, C.C., 2001. Effect of Long-Term Exposure to Fluoride in Drinking Water on Risks of Bone Fractures. J. of Bone and Mineral Research 16:932-939.*

Reviewed by: Timothy J. Wade Ph.D MPH, and Rebecca L. Calderon, PhD, MPH, USEPA/NHEERL/HSD

Summary:
This semi-ecological study tested the hypothesis that exposure to fluoride in drinking water is associated with bone fractures. Subjects 50 years of age and older were recruited from six rural Chinese populations. The authors state that there were few other dietary and non-dietary fluoride exposures in this population and confirmed this with a 10% sampling for analyses of dietary items. Subjects residing in the area of the highest fluoride drinking water concentration (4.32-7.97 ppm) were at significantly higher risk of overall fractures (OR=1.47, p=0.01), and for hip fracture (OR=3.26, p=0.02), compared to the group with 1.00-1.06 ppm fluoride in their drinking water. The authors report a “U” shaped relationship was observed for overall fractures, but not hip fractures, with a significant risk of overall fractures also occurring in the lowest exposure, 0.25-0.34 ppm, (OR=1.50, p=0.01). Fluoride exposure appeared to be determined by the range reported in the community, but individuals were interviewed about their history of fractures, and X-rays were obtained to confirm the majority of the fractures. Information was also collected on some covariates.

Review:
A study was conducted in a series of rural Chinese populations to evaluate the effects of long term exposure to fluoride in drinking water. The studied populations were ideal for this type of study because of the minimal migration and little if any exposure to fluoride outside their drinking water. The investigators took great care to confirm outcome (fractures) with three different methodologies. It is unlikely that misclassification based on health effect occurred. The authors complied with both US and International guidelines regarding the conduct of human studies.

Inadequate exposure assessment, potential biases associated with misclassification of exposure, the failure to examine fractures other than hip separately, and limited statistical analysis were the most problematic aspect of this study.

It was unclear on what basis communities were selected for inclusion into the study. Was it based on prior knowledge of fluoride levels in water? If so, from where were the fluoride measures obtained? It was unclear what types of water supplies the

communities relied on and whether or not there were alternate sources of water available. Similarly, it was unclear how fluoride concentrations for specific communities were determined. Was it based on the investigator's own water sampling investigations or from previous records? Details about how water samples were collected and where they were collected from was inadequate, making interpretation of the fluoride exposure categories difficult. It seems plausible that fluoride concentrations may have significantly changed throughout the risk period (greater than 30 years) but no data or discussion is provided to confirm or refute this. Furthermore, selection of wells for analysis may have introduced an uncertainty of exposure classification. Many rural areas in China have multiple wells per village and it is often difficult to assign a specific well to a particular family. No data is also available on individual water consumption and usage. A positive interaction between amount of water consumption and fluoride concentration would have been expected. Biomarkers of fluoride exposure (e.g., urinary or blood measures) would have confirmed classification of exposure.

No mention is made of the training of interviewers and whether or not they were aware of the hypotheses under investigation. Since results were self-reported, interviewers who were aware of the goals of the study could have knowingly or unknowingly influenced the study results. Likewise, it was not clear whether or not study subjects were aware of the research questions being investigated.

The community with the highest fluoride concentration was significantly younger, and a considerably higher percentage male. Although these factors were adjusted for in the logistic regression, these differences cause great concern that there may be "residual confounding" and that there may be additional unmeasured factors that differed among the groups. In other words, there is evidence that these groups truly represent different samples, complicating the interpretation of the result.

Physical activity is a known risk factor for fractures. The authors cite a standardized method for assessing this, however, it is unknown how well this relates to activities associated with fractures such as accidents, occupation, nutrition and underlying illness. It is difficult to assess the reliability of this measure. This could be a significant source of bias in this study, particularly given other data that suggests these groups represent different samples.

A 10% sample of the individuals were assessed for dietary sources of fluoride and overall nutritional status. Very little discussion of this methodology (random, stratified sampling etc) and why they think the sample is representative of the study population was provided. This would have been a stronger study had this been explained in more detail.

The results for a "U" shaped exposure-response function for the relationship between fluoride and overall fractures is inadequately explained, and is inconsistent with the

results for hip fractures (not internally consistent). It is unclear why this relationship would be present only in overall fractures, but not hip fractures. Presumably, hip fractures are part of overall fractures, but the data for fractures other than hip are not shown. When crude calculations are conducted based on the data provided, the unadjusted OR for fractures other than the hip it is apparent that the results seen for overall fractures were heavily influenced by hip fractures. For fractures other than the hip there is less evidence of the same association seen for hip and overall fractures. The OR (unadjusted) for the highest exposure category is 1.33 (95% CI: 0.96-1.84). Note that adjusted ORs reported in the hip and overall categories tended to be smaller (i.e. closer to one), and that adjustment generally will widen confidence intervals, so it can be presumed that adjustment would result in a smaller OR with wider confidence intervals. For fractures other than hip, there was a stronger relationship at the lowest exposure category (OR=1.52, 95% CI 1.10-2.13). This should have been examined more fully by the authors. (Note: confidence intervals done by reviewers).

The statement of a "U" shaped exposure-response function is based on point estimates for prevalence and OR in this analysis. Given the large population numbers (~1000 per group), the authors should have provided confidence intervals to allow the reader to assess the robustness of the data. While there may have been significant differences between point estimates, wide confidence intervals would suggest potential biases in the data. Narrow confidence intervals would have been evidence for the robustness of the point estimates.

This is the first time a "U" shaped exposure-response function has been reported for fluoride exposures. No biological hypotheses are offered for this phenomenon. Furthermore, the authors state the usual "protective" effect at low levels seen in other studies was not see here.

The removal of gender from the analysis of the results in Table 4 is puzzling, despite not having a significant effect, it is clearly related to community and the risk of fracture. Gender may have affected the point estimate for the coefficients in the logistic regression. Removing this from the model may have inflated the coefficients for the highest fluoride group since this group was predominantly male, and males were at greater risk for fracture. Moreover, males were more likely to smoke which was moderately, but not significantly associated with increased risk of bone fracture.

Summary:
The results of this study suggests an elevated risk of fracture with fluoride drinking water levels above 4.32 ppm, and below 0.34 ppm in communities with apparently little other exposure to fluoride. This study took advantage of a non migratory population that facilitated the conduct of a study to evaluate the effects associated with long-term low levels of fluoride in water. This study suggests that a more hypothesis specific study would be feasible in the study population. Future studies should address

problems in exposure assessment. Exposure assessment in this study was not adequate enough to support the conclusions. The results for overall fractures seem to be mostly influenced by a strong association in hip fractures. The statistical significance of the highest exposure level is not evident when fractures other than hip fractures are considered, although risk remains associated with the fluoride exposure at the lowest exposure level for fractures other than hip. It is unclear if risk for fractures is greater for those with very low exposure or those in the higher exposures, and whether or not this "U" shape response is consistent with what scientists would expect or could be an indication of bias with the study design. Furthermore, the ecological design cannot account for potential differences among the exposure groups, which seem indicated by imbalances in both gender and age. Conclusions about exposure-response are difficult due to the lack of individual exposure data.

Reviews by Dr. J. Blondell:
Reference #1: *Alarcon-Herrera MT et al (2001). Well water fluoride, dental fluorosis, and bone fractures in the Guadaine Valley of Mexico. Fluoride, 24(2): 138-148*.

Background

Fluorosis is mottling (discoloration) of tooth enamel. It can occur in children who drink water with > 1 ppm fluoride during the period of tooth development. The enamel changes can range from irregular whitish opaque areas to severe brown discoloration of the entire crown with a roughened surface. Such teeth have a high resistance to dental caries. Severe dental fluorosis weakens the enamel resulting in surface pitting.

Bony changes that can occur in skeletal fluorosis include osteosclerosis (hardening of bone) and exostoses (bony growth of surface of bone) usually seen after prolonged high intake in adults. Communities with high intakes above 10 ppm can be affected.

Background from "The Merck Manual of Diagnosis and Therapy, Fifteenth Edition" edited by R. Berkow (Merck, Sharp & Dohme Research Laboratories, Rahway, NJ) 1987.

Study review

Study sampled population for occurrence of dental fluorosis, non-traumatic bone fractures, and water supply fluoride levels in the Guadiana Valley of Mexico. A multistage, cluster sampling technique selected some 77-80 families made up of 1,437 individuals from five areas with different levels of fluoride. Fluoride was determined by analysis of all 74 wells in both the rural and city areas of the valley. The following results were obtained:

Fluoride level (ppm)	Number of wells	Population	Children sampled	Percent children with fluorosis that was: none	moderate/severe	Percent fractures in children/adults	
nd* - 1.5	12	9,428	97	24	7	5	3
1.51-4.99	52	400,591	112	14	13	9	8
5.0-8.49	6	3,823	38	5	3	3	9
8.5 - 11.9	2	4,270	27	15	19	11	7
> 12	2	392	59	0	59	8	6
	74	418,504	333				

* nd = non detectable

The table above appears to show some evidence of non-traumatic fractures in children associated with the two highest fluoride levels. However, the two highest levels occurred only in rural areas where children are likely to be more active out-of-doors where rough and tumble play might lead to fractures without any immediate cause being apparent. The authors do not mention this important confounder and the percentages are based on a total of 10 fractures, a relatively small number on which to base conclusions. On the other hand, the authors also performed a regression analysis between the Dean Index of fluorosis and fractures which demonstrated a very high correlation ($R^2 > 0.93$) for both adults and children. Both children and adults with severe fluorosis had three times the percent of fractures (about 15%) as children and adults with no evidence of fluorosis. The authors did not explain why fluorosis but not level of fluoride in the water was correlated so highly with occurrence of non-traumatic fractures. The authors note that fluoride "may increase bone quality, but it can also decrease bone quality and strength . . . The correlation of bone fractures and dental fluorosis seems to be linear, but bone fractures and fluoride water concentrations show a third order polynomial curve. We have no explanation for this finding, although a clear tendency was observed, and this must be analyzed in future studies." It appears likely that exposure to other sources of fluoride may have influenced the results of this study. In addition, mobility within the valley could have led to exposure misclassification. The occurrence of 7% fluorosis in the lowest exposure group strongly suggest this possibility. Therefore, it is not possible to determine, from this study, what the contribution of fluoride in drinking water is to increasing risk for non-traumatic fractures.

Reference #34: *Galletti P-M, Joyet G. 1958. Effect of fluorine on thyroidal iodine metabolism in hyperthyroidism. J. Clin. Endocr. Metab. 18:1120-1123.*

Sodium fluoride was administered three times daily in oral doses of 2-10 mg to 15 adult patients with hyperthyroidism for a period ranging from 20 to 245 days. Signs and symptoms of hyperthyroidism disappeared in 4-8 weeks for 6 of the 15 patients. Some patients received injected doses of 20 mg. Younger patients with Graves disease did not respond at all. The present reviewer is not qualified to comment on the mechanistic hypotheses and relationships advanced to explain the interaction of fluoride and iodine and their effect on thyroid function. The authors were not attempting to test fluoride as a medical treatment for hyperthyroidism but rather "to elucidate the inhibitory effect of chronic administration of fluoride upon thyroid function in cases of hyperthyroidism". They concluded "such an action appears only occasionally among persons subjected to massive doses of this substance." Other reviewers have concluded that "no fluoride toxicity was seen in these patients", but given the aim of this study, the limited focus to thyroid effects, and the absence of a listing of toxic effects or any evidence that they were measured; this statement is not warranted.

Reference #39: *Heller, KE, Ecklund, SA, and BA Burt. 1997. Dental caries and dental fluorosis at varying water fluoride concentrations. J Public Health Dent 57(3):136-143.*

Using the National Survey of US School-children, 18,755 children with a single continuous residence were identified for the study of the effect of fluoride exposure on dental caries and dental fluorosis. Fluoride levels of school water were used as an indicator of children's fluoride exposure from water. The use of fluoride drops, tablets, and other sources was ascertained by questionnaire. Appropriate precautions were taken to ensure reliability of fluorosis measurements by testing interrator comparability and requiring an agreement within one point on the Dean's Fluorosis Index.

To this reviewer, the most important limitation of this study was the reliance on a single 500 ml sample from each school as an indicator of fluoride exposure for all children in that school. Most water is likely to be consumed at home and while tap water is likely to come from the same source (water treatment plant) as the school's, this will not always be the case. The use of bottled water, soft drinks, soups, and juices might easily account for 50% or more of a child's fluid intake. The variation and potential bias from these other sources is not known to the present reviewer. Do charcoal filters or other water treatment devices used in schools and homes have any effect on fluoride bioavailability? This reviewer is not qualified to say. The authors note that another limitation of their study, that reliance on children residing in a single residence their whole lives "necessarily reduces the generalizability and external validity of the findings". In general, the authors do a good job of discussing the strengths and weaknesses of their analysis.

The authors conclude “it is likely that some degree of fluorosis will occur at even low levels of fluoride exposure. Therefore, any level of water fluoridation will necessarily involve a trade-off between obtaining desired caries reduction with an acceptable level of concomitant fluorosis.” The authors advocate using the lowest level of fluoride to achieve the desired therapeutic effect and argue that this level is now around 0.7 ppm rather than 1-1.2 ppm suggested by earlier research. They acknowledge that this change may be due to the increased exposure to fluoride from sources other than drinking water. In a letter to the editor, it was pointed out that the inclusion of older school children (up to 22 years of age) introduces potential recall bias and measurement error concerning exposure in the first six years of life. Use of a single water sample for middle and high school children may not be representative of earlier exposures, though this likely would result in additional random error rather than systematic error.

Reference # 56: *Investigations of soft tissue functions in fluorotic individuals of North Gujarat Michael M, Barot VV, Chinoy NJ Fluoride 29:63-71. 1996.*

This is a survey of clinical chemistries among residents in a region of India with high levels of fluoride in drinking water (500 people from 52 villages; 1.0 - 6.5 ppm fluoride) compared to persons in a ‘normal’ fluoride level city (mean 0.6 ppm). The number of individuals used to compute summary data in the paper varied from 40 - 76 in the high fluoride group and 15 or 22 in the normal fluoride group. Dental and Skeletal Fluorosis were common in the high fluoride residents: 74% had slight to severe tooth mottling; 59% had stiff spine. There was a correlation between water fluoride and serum fluoride concentrations. Serum fluoride also showed a significant age relationship.

Differences were found for some clinical chemistries in the high fluoride group compared to the normal group. Significant decreases were noted for protein and calcium concentrations. Significant increases were noted for SGOT, SGPT, sodium, potassium, adrenalin, noradrenalin, cholesterol and T4. No changes were noted for T3, TSH, hemoglobin and testosterone.

Reference #76: *Williams JE et al. (1990). Community Water Fluoride Levels, Preschool Dietary Patterns, and The Occurrence of Fluoride Enamel Opacities. J of Pub Health Dent; 50:276-281.*

A study of fluorosis was conducted in Augusta, Georgia and nearby Richmond County among 374 12-14 year old children. Two city and two county middle schools with a population of 3,000 students received a request and questionnaire to participate in the study. A total of 1,003 questionnaires were returned for a response rate of 33.4 percent and 990 consented to an examination. Of these, 408 students were life-long residents of the area. However, just 374 appeared in time for the examination and

were not excluded for reasons such as wearing orthodontic appliances. City residents were consistently exposed to water fluoridated at 0.9 to 1.2 ppm and county residents were exposed to levels between 0.2 and 0.9 ppm. Fluorosis measurements emphasized the front of the teeth to focus more on esthetic appearance and, hence the overall rate of fluorosis may have been underestimated. One individual who received training by two other examiners performed all the examinations. The questionnaire included assessment of whether children had additional exposure to fluoride from food, drink, vitamins, or toothpaste

Higher scores indicating fluorosis were more prevalent in city children than in county children. High scores for fluorosis were observed in 14.0% of city children and 1.4% of county children. Low to moderate scores were seen in 66.9% of city children and 52.5% of county children. There were no significant associations with gender, race, milk, food, or beverage ingestion, vitamin and fluoride drop supplementation, or toothpaste swallowing. The authors attribute the relatively high rate of fluorosis to additional sources besides drinking water including: fluoride supplements (fluoride/vitamin drops) which were reported by 80.5% of parents, ingestion or absorption from dentifrices and mouth rinse, and fluorides in infant formulas, foods and beverages.

As noted above, the authors make a key point, fluoride exposure comes from several sources and drinking water alone is likely not responsible for the fluorosis observed in the current study. Rather drinking water levels need to be set taking into account background levels and exposure to other sources of fluoride. The authors recommend that the association between "these multiple sources and the occurrence of fluoride enamel opacities should be explored further." The authors warn against generalizing their results to other locations "because of the fluctuations in the fluoride content in Richmond County and the unusually high frequency of inappropriate fluoride-vitamin drop supplement use in the local area." Another serious limitation which the authors did not discuss was the very low response rate of only 33% of initial questionnaires returned. They did not, but should have mentioned whether this response rate differed significantly between city and county children. Presence or absence of dental problems, socio-economic factors, and other unknown factors which might have biased participation in the study were not discussed. On the whole, however, it does not seem likely that most of these factors would differentially affect participation in city versus county children in this reviewer's opinion. Nonetheless, this is an important caution that should prevent over interpretation of these results along with the factors the authors pointed out themselves which are listed above.

Reference #112: *Takahashi K, Akiniwa K, Narita K. 2001. Regression analysis of cancer incidence rates and water fluoride in the U.S.A. based on IACR/IARC (WHO) data (1978-1992). Journal of Epidemiology 11:170-179*.

The three authors of this study apparently include 2 dentists and one member formerly on the faculty of medicine at Tokyo University (the lead author). None of the authors appear to have sufficient background in epidemiology or statistics. They compared cancer incidence rates for 36 sites in 3 states and 6 metro areas with a water fluoridation index. Regression analysis found that 64% of the 36 sites (e.g., lung, colon, etc.) had some positive correlations with fluoridation index.

The only other contributing variable considered was percent sunlight which is not as important a determinant of skin or lip cancer, as is latitude, which is a much better measure of time out-of-doors with exposed skin. So the only confounder measured was the wrong one! The cities and states in this study are in no sense a sample of the USA, they are locations that happen to have a cancer registry. Other factors known to affect cancer such as urban/rural gradient (at least for the three states) were totally ignored. Five locations were excluded: "New York City which does not separate black people; New Mexico where the experiments with the Atomic Bomb were conducted; Alameda CA for which the population density of the Metropolitan area could not be found; Alaska and Hawaii for which conditions are far different from the other States of the USA". Neither the New Mexico or Alameda exclusions are justified. There is no evidence that the atomic bomb tests influenced cancer rates for the whole state during the period studied and the authors did not measure the effects of population density in any case.

Many factors besides fluoride undoubtedly vary among the nine diverse locations but none (other than % sunlight) were measured. Even more important, nine is too small a number of locations, even if they do represent over 20 million people. Nine data points that were not sampled (but rather selected based on available data) could easily be subject to a variety of distribution problems. The authors claim that a logalithmic (sic) transformation was required to normalize the cancer incidence data, but no data is supplied to confirm this. And this procedure is not normally used in studies of this type. The concluding sentence of the methods section states "The cancer incidence ratios FD at the level of 100%/1% (CIR-100) were defined as the magnitude when the exposure of the inhabitants to fluoride increased hundred times from 1 to 100%." This sentence appears to contain incorrect terminology (cancer incidence rates not ratios) and does not make any logical sense.

The authors performed a log transformation on the percents for the nine locations that received water adjusted to 1 ppm fluoride or with naturally occurring fluoride levels of 0.7 ppm. No justification is provided for these particular levels. Normally, arcsin transformation is the appropriate transformation for percents. An arcsin or angle transformation of the percent receiving a certain level of fluoride has the effect of stretching out both tails of the distribution curve, which may help stabilize the variance and provide a better fit. The angle transform enables linear regression analysis of data when one of the variables is a proportion. The authors claim "These

two logalithmic transformation (sic) confirms the linearity of the regression curve" but provide no evidence that either transformation was warranted or appropriate.

Given the extreme methodological shortcomings of this study, no conclusions can be drawn from this paper

ATTACHMENT 2

Reference #14: *Chinoy, N and E. Sequeria. 1988. Effects of fluoride on the histoarchitecture of reproductive organs of the male mouse. Repro Toxicol 3:261-267.*

Groups of 40 mature male mice were treated via feeding tube with doses of 0, 4.5 or 9 mg/Kg F as NaF for 30 days. Two additional groups of 40 mice were given doses of 4.5 mg/kg F as NaF for 30 days after which treatment was stopped for a period of 30 or 60 days before sacrifice. No information was provided on the fluoride content of the diet. After sacrifice the testes, caput epididymis, cauda epididymis, vas deferens, prostate and seminal vesicles from the control and treated mice were removed and prepared for histological examination.

Fluoride treatment was associated with changes to the architecture of the testes. Lydig cells, prostate, and seminal vesicles did not appear to be affected by treatment. However, there were observed changes in the seminiferous tubules, caput- and cauda epididymis, and vas deferens. The germinal epithelial cell height was significantly reduced in both exposed groups but the decrease was not dose-related. Changes in the caput- and cauda epididymis included disorganization of the secretory epithelium, nuclear pyknosis and an absence of spermatozoa in the lumen. Epithelial cell height was also significantly reduced for both cell types in both treatment groups, but as with the testes, the decrease was not dose-related. An increase of the lamina propria of the vas deferens was observed and the muscle layers were reduced. In both cases the measured changes were not dose-related.

Histological evaluation of the tissues from the animals sacrificed after recovery periods of one and two months indicated that the changes in the testes were reversible. The fluoride treatment did not appear to cause any permanent structural testicular alterations

There are several studies conducted by other investigators that have not been able to reproduce the findings of Chinoy and Sequeira. For example, the US Food and Administration conducted a multigeneration study in rats. (Collins TF, Sprando RL, et al, Multigenerational evaluation of sodium fluoride in rats Food Chem Toxicol. 2001 Jun;39(6):601-13.). In this study, the effects of sodium fluoride ingestion at 0, 25, 100, 175 or 250 ppm in drinking water measured in rats throughout three generations, and no cumulative effects on reproduction were observed. FDA also reported (Sprando RL, Collins TF, et al., Black TN, Rorie J, Ames MJ, O'Donnell M, Testing the potential of sodium fluoride to affect spermatogenesis in the rat., Food Chem Toxicol. 1997 Sep;35(9):881-90) that sodium fluoride did not affect spermatogenesis and endocrine function (LH, FSH and serum testosterone were measured) in P and F1 generation male rats exposed in their drinking water at one of four concentrations (25, 100, 175, 250 ppm). Li Y, Dunipace and Stookey (Effects of fluoride on the mouse sperm

morphology test, J Dent Res. 1987 Sep;66(9):1509-11) did not find any spermatogenic influence of sodium fluoride (NaF) by means of the sperm morphology test when mice were intubated up to a maximally tolerated dose of NaF (70 mg/kg).

Reference #52: *Luke, Jennifer. The Effect of Fluoride on the Physiology of the Pineal Gland. Ph.D. Dissertation at the Univ of Surrey, Guildford, England. March 27, 1997*

There are high levels of F in the human pineal gland and this thesis explores the relationship between F and basic physiology of the pineal gland using the gerbil as the test species. The pineal glad is outside of the blood brain barrier.

In the actual experiment, there were 2 groups of 24 gerbils (12 of each sex).One group(HF) received approximately 2.3 mg/ kg/ day 5 days a week by the oral route from birth to weaning at day 24. After that, they received a high F diet with 37 mg/kg daily dose. The low F gerbils (LF) did not received any F by oral gavage and at weaning ate a regular diet containing 7 mg/kg/day.

There was a decrease in excretion of melatonin's urinary metabolite in the HF gerbils as compared to the LH group. The author discussed the hypothesis that depressed melatonin levels in the blood may hasten the onset of puberty and, in a pilot study, indicated that this was seen in HF females. However, as the author states, this interpretation is "conjectural". Because this data is the result of a single study with limited number of animals with only two dose levels, the findings should be confirmed by other studies.

Reference #58: *Mullinix, PJ, Denbesten, PK, Schunior, A and WJ Kernan. 1995. Neurotoxicity of sodium fluoride in rats. Neurotox. and Teratology 17 (2):169-177.*

Changes in behavior were monitored in groups of Sprague Dawley rats after exposure to fluoride at three different developmental stages. Body weight, plasma, and brain fluoride levels were also measured. Behavioral patterns such as sitting, grooming, head turns, standing, etc. were recorded by video camera and analyzed for their frequency and duration. Pairs of control and experimental animals were observed at the same time. Several of the statistical methods used to evaluate the data, e.g. the RS statistic and K(t) values, were not fully described by the authors.

In the first group, pregnant dams were exposed by subcutaneous injection to 0.13 mg/kg sodium fluoride on gestational days 14-18 (n = 7) or days 17-19 (n = 9) two or three times daily with at least 4 hours between injections. Nine weeks after delivery, the pups (10/litter) were observed for body weight, plasma fluoride and behavior. The K(t) values were depressed for the exposed animals as opposed to the controls. K values are described as measures of time structure changes. The author reported that fluoride exposure appeared to effect behavior-time structure but not behavior initiation

or total time. The rats received no fluoride treatment other than that during gestation. There were no significant differences in body weight or plasma fluoride at 9 weeks.

The second group of animals were exposed immediately after weaning to 0, 75, 100, 125, or 175 ppm fluoride as sodium fluoride in their drinking water for 6 or 20 weeks. The diet contained 10 ppm fluoride. The 175 ppm group was discontinued after 10 days because of dehydration and death of some of the animals. No measurements of drinking water consumption or dose were reported. Significant deficits in body weight were noted in the 125 ppm dose group and plasma fluoride levels were greater than those for the controls in all dose groups. In the group exposed for six weeks, the plasma fluoride levels did not show a dose-response trend and were higher for the 125 ppm females than the 100 ppm group. In the males the plasma fluoride level was greater for the 75 ppm group than the 125 ppm group.

The behavioral RS statistic was directly proportional to plasma fluoride levels but not to dose. The RS statistic is described as a statistic that differentiates low level behavioral effects from noise. Some decreases in specific behavioral activities were reported for the 100 and 125 ppm females and the 125 ppm males when compared to controls. However, there was also a fair amount of variability in the scores for the control animals and the lack of data for the 75 ppm females and the 75 and 100 ppm males makes it difficult to assess the dose-response.

The last group of animals were given either 0 or 100 ppm fluoride as sodium fluoride in their drinking water for 5 to 6 weeks. Testing was initiated when the animals were 10 weeks old. The animals were given a low fluoride diet. Plasma fluoride levels after 6 weeks were lower adults than in the young rats after a similar exposure period. There were no observed effects on body weight. The RS statistic indicated that the behavior of the females, but not the males, was significantly different from that for the controls.

Brain fluoride levels were measured in the juvenile (post weaning) rats after 20 week exposures to 125 ppm and in the adult rats after 6 week exposures to 100 ppm fluoride. Only one set of control values was reported even though there were two distinct experiments conducted. The control values are reported as pooled values for the 100 ppm and 125 ppm groups. It is assumed that this means that half the control animals were exposed for 20 weeks starting as juveniles and half as adults for six weeks. No significant differences were seen in the adults except for in the hippocampus in females and the medulla oblongata in males where the level in the treated animals were elevated. In the juvenile rats, the fluoride concentrations were increased significantly in most regions of the brain for both males and females. The authors concluded that the results of this study demonstrate the potential for fluoride exposures to influence the developing brain.

The conclusions reached by Mullenix et al. (1995) are not supported due to a number of problems with their study. There have been no systematic studies comparing the Mullenix method for measuring neurobehavioral effects with the standard neurotoxicology battery, which has undergone extensive and international validation studies. There is no published record of validation of the Mullenix method. Also, the numerous T-Tests performed by these authors can lead to significance of results based on chance alone. Finally, there is no scientific basis to imply that motor changes are surrogate of cognitive deficits, as the authors do so in this paper.

Reference #81: *Zhao, W., Zhu, Z, Yu, Z, Aoki, K, Misumi, J, and X Zhang. 1998. Long-term effects of various iodine and fluorine doses on thyroid and fluorosis in mice. Endocrine Reg 32:63-70.*

The authors of this report studied the interactions of fluoride and iodine in Kunmin mice. The animals were divided into nine groups of 32 animals which received different combinations of iodine and fluoride in their drinking water. Iodine was administered as potassium iodate and classified as iodine deficient (ID , 0 µg/L), iodine normal (IN, 20 µg/L) or iodine excess (IE, 2500 µg/L). The drinking water fluoride concentrations (NaF) were: fluoride deficient (FD, 0 mg/L), fluoride normal (FN, 0.6, mg/L) or fluoride excess (FE, 30 mg/L). A special low iodine and low fluoride chow was fed the animals. The nine combinations of nutrients were ID/FD, ID/FN, ID/FE, IN/FD, IN/FN, IN/FE, I.E./FD, I.E./FN, and I.E./FE. The animals were treated for 150 days and the following parameters were measured, incisor fluorosis at two-weeks, radioiodine uptake at 100 and 150 days, Serum T3 and T4, histological evaluation of the thyroid, fluoride content of the bones.

For many of the nutrient combinations there were no apparent interactions between fluoride and iodine. Fluoride produced the expected incisor fluorosis at two weeks in the FE group. There was no incisor fluorosis in the FD or FN groups. An ID increased the severity of fluorosis in the FE group. The fluoride content of bones was dramatically increased in the FE group. Surprisingly it was higher in the FD/ID group than in the FN/ID group. Otherwise the FD and FN groups with IN or IE exposures has fairly similar bone F levels.

Under ID conditions increasing F intake appeared to be associated with a decrease in thyroid weight. At 100 days the incidence of goiter in the ID group increased with increasing F intake. When the iodine was in excess, the incidence of goiter decreased with increasing F intake. Increased T3 levels were seen in the ID/FE groups and T4 was decreased in the ID/FD and ID/FN groups. FE inhibited radiolabeled iodine uptake in the ID and IN groups.

The mechanistic basis for the interactions between fluoride and iodine observed in this study is not clear. The authors mention that their T3/T4 results are not

consistent with other studies and acknowledge that they can not offer any plausible explanation between their results and those of other investigators. The authors also state “it is generally believed that fluorine does not influence either thyroid function or structure at the amount (about 1ppm in water) used to prevent dental caries.” The authors suggest the possibility of interactions between the two minerals, particularly when deficiency of one is combined with excess of the other. The mechanistic basis for the interactions between fluoride in iodine observed in this study is not clear.

ATTACHMENT 3

UNITED STATES ENVIRONMENTAL PROTECTION AGENCY

WASHINGTON, D.C. 20460

OFFICE OF
PREVENTION, PESTICIDES
AND TOXIC SUBSTANCES

October 1, 2003

MEMORANDUM

SUBJECT: Review of Five Recent Papers on Fluoride Submitted by the Fluoride Action Network

FROM: Vicki L. Dellarco, Ph.D., Senior Scientist
Health Effects Division

TO: Debra Edwards, Ph.D., Director
Registration Division

The Health Effects Division (Drs. William Burnam, Karl Baetcke, Jerome Blondell, and Vicki Dellarco) has reviewed the following five papers on fluoride:

1. A. Shashit, Histopathological investigation of fluoride-induced neurotoxicity in rabbits; Fluoride Vol 36, 95-105, 2003
2. L. Li, The biochemistry and physiology of metallic fluoride: action, mechanism, and implications; Crit. Rev. Oral Biol. Med 14: 100-114, 2003
3. D. Ortiz-Perez et al., Fluoride-induced disruption of reproductive hormones in men; Environ. Res. 93:20-30, 2003
4. Q Xiang et al., Effect of fluoride in drinking water on children's intelligence; Fluoride 36:84-94, 2003.
5. E.H. Goh and A.W. Neff, Effects of fluoride on Xenopus embryo development; Food and Chemical Toxicology 41:1501-1508, 2003

Based on our review, we find that these papers contain many limitations (e.g., in study design, analysis of data, and/or reporting of results). One paper was a review and did not contain new information. Thus, the articles cited by the Fluoride Action Network do not provide a compelling basis to depart from the EPA's current MCL based on the preliminary review of these five additional references. A summary of each paper is provided below.

6. A. Shashit, Histopathological investigation of fluoride-induced neurotoxicity in rabbits; Fluoride Vol 36, 95-105, 2003

This publication is a report of an investigation of the neuropathological and functional effects in rabbits treated subcutaneously with sodium fluoride. The doses administered were 0, 5, 10, 20, or 50 mg/kg/day. Slight neuropathological changes were observed in rabbits treated with 10 mg/kg/day and more pronounced neuropathological changes were reported with increasing doses. Neurological symptoms, including hemiplegia, spastic paraplegia, seizures, tremors, and partial or complete paralysis were reported to occur in rabbits treated with 20 mg/kg/day or higher. The effects on functional behavior were attributed by the authors to neuropathology of the brain tissue.

The Materials and Methods section of the publication contains details on the animals used and housing conditions, preparation of dosage solutions, treatment groups, and preparation (emersion fixation) of brain tissue for histopathological evaluation. No details are provided on the protocol followed for functional evaluations.

There are deficiencies in the study that preclude using the information in this publication in a weight of evidence evaluation of the potential for sodium fluoride to induce neuropathology. For example, perfusion of brain tissue is the preferred method for the fixation of brain tissue because emersion fixation may result in artifacts or render the interpretation of lesions problematic. Also, the terminology used by the author is, in some cases, inconsistent with that which is generally accepted. The author states, for example, that the neuroglial cells exhibited chromatolysis but this is not a architectural feature of glial cells. Although the author attributes the functional changes observed in some treatment groups to the neuropathological effects of sodium fluoride, this cause and effect relationship can not be established. This is because histopathology was performed at study termination only and it is not possible to determine whether functional effects preceded or coincided with the neuropathology. Furthermore, it is possible that the neuropathology was secondary to toxicity induced in other tissues (no histopathology was performed on tissue other than brain). For example, effects on pulmonary function can result in neuropathology. Convulsions and paralysis can also lead to neuropathology. Finally, the use of subcutaneous injection as the route of administration confounds dose-response comparisons with oral dose levels that may lead to adverse effects.

Because of the confounding factors (e.g., use of emersion fixation, questionable terminology, absence of information on the methods followed for functional observations and absence of information on incidence and time of onset), the results reported in this study do not add to the weight of the evidence for the potential of sodium fluoride to induce adverse effects in humans that are exposed to sodium fluoride at levels established by the current MCL.

7. L. Li, The biochemistry and physiology of metallic fluoride: Action, mechanism, and implications; Crit. Rev. Oral Biol. Med 14: 100-114, 2003

This publication is a review of information provided in the literature on the biochemistry and potential mechanism of action of fluoride complexes, particularly aluminum fluoride. The major focus is on the interaction of aluminum fluoride with G-protein and the enzymes associated with G-protein activation. The review does not provide new information on the levels of fluoride or fluoride complexes that might be associated with adverse effects in laboratory animals or humans and thus, although potentially useful for hazard assessment, would not provide information useful for evaluation of dose-levels of fluoride associated with adverse effects.

8. D. Ortiz-Perez et al., Fluoride-induced disruption of reproductive hormones in men; Environ. Res. 93:20-30, 2003

Ortiz-Perez et al. (2003) studied the association between fluoride exposure and subclinical effects related to reproductive hormones. This study looked at 133 men aged 20-50 years with occupational exposure to fluoride and compared to 27 men who were only exposed to fluoride in drinking water. The occupationally exposed group had at least one year in fluoride-related industries and were divided into four subgroups: fluorhydric plant (producing HF and AlF_3), sulfuric plant (where fluorspar is dehydrated), maintenance (either industry), and administration. Based on urinary fluoride levels adjusted for creatinine, occupationally exposed workers were estimated to be exposed to 3.4 to 27.4 mg F/day compared to an exposure of 2.-13 mg F/day for men only exposed from fluoride in drinking water. Questionnaires obtained information on different potential sources of fluoride, sociodemographic characteristics, occupation, reproductive history, alcohol consumption, and tobacco exposure. Laboratory analysis included urinary fluoride, blood lead levels, hormonal analyses, and semen analysis. An index of fluoride exposure was created by considering the time at work multiplied by the urinary fluoride level. The mean urinary fluoride level in the occupational group was twice as high as the low exposure group only exposed from drinking water, however there was considerable overlap. A t-test found a significant association between urinary fluoride levels and four out of seven reproductive hormones (higher FSH and lower levels of inhibin-B, prolactin, and free testosterone). Analysis of covariance and multiple regression examined age, alcohol consumption, tobacco, lead in blood and years working to adjust for their influence on hormone values and semen characteristics. This analysis supported an effect of fluoride on lowering levels of inhibin-B levels and a decreased sensitivity of FSH to inhibin-B resulting in a secondary increase in FSH. No abnormalities were found in the semen parameters associated with fluoride levels. The authors note that workers in this study were also

exposed to aluminum and sulfur dioxide and acknowledged that they could not "rule out their partial participation in the effects observed in the present study". They concluded that "fluoride might be a reproductive toxicant for humans. Nevertheless, it is important to perform more complete studies in those communities heavily exposed to fluoride." The doses reported in this study were higher than in the United States and Canada but lower in Russia and India where fluoride has been associated with other effects on hormones (notably lowered testosterone) and reproductive effects (male infertility).

This study suffers from a poor design and inadequate sampling. Nothing is said about how the 27 workers exposed only to drinking water were selected, other than that all subjects apparently resided in the same town. The only selection criteria for exposed workers appears to be at least one year employment in plants where fluoride exposure occurred. In the occupational group, the authors fail to acknowledge that lower exposed administration workers undoubtedly differed in many lifestyle factors (e.g., diet, access to health care, medical history) from the other more highly exposed workers and therefore, are inadequate for use as an internal comparison. The authors acknowledge that other exposures at the plant may be an explanation for some of their findings. How long workers resided in the town, types of occupations among the reference population, lifestyle differences, environmental exposures, and other factors that might influence hormonal levels are inadequately or barely discussed. The authors fail to acknowledge that all the major factors which influence hormonal levels in men have yet to be identified and their study may have easily missed important factors which influence reproductive hormones. It must be emphasized that all of the effects measured were subclinical and that this study did not find evidence of an adverse reproductive outcome. This study did suggest a possible influence of fluoride manufacturing exposures on a hormone level that may be consistent with effects reported in other studies. However, this study is much too preliminary and no conclusions about the effects of fluoride on hormone levels are warranted until this study is repeated in other populations with a carefully chosen comparison group and sufficient sample size to analyze for the effects of confounding variables.

9. Q Xiang et al., Effect of fluoride in drinking water on children's intelligence; Fluoride 36:84-94, 2003.

A study by Xiang et al. (2003) measured the association between fluoride in drinking water and children's intelligence as measured by intelligence quotient (IQ). The study was conducted in two villages 64 km (40 miles) apart where the primary source of fluoride was from drinking water. Fluoride was measured in drinking water and urine with a fluoride ion selective electrode. Over 90% of the children, aged 8 to 13 years, in both villages (512 total, 222 in Wamiao and 290 in Xinhuai) participated in the study and received the Combined Raven's Test for Rural China which measures IQ. Both children and their parents supplied information on medical history (e.g., head trauma in the child), education level, socioeconomic status, and lifestyle. Urinary

fluoride levels in children the Wamiao village, based on 155 samples, was 3.4 times higher than mean levels for children in the Xinhuai village (based on 135 samples). This difference correlated highly with levels of fluoride in drinking water. Mean IQ was 8 points lower in Wamiao than in Xinhuai, a statistically significant difference that was found in both males and females. "When compared with the children in Xinhuai . . . , the children in Wamiao exhibited, as the level of fluoride in the drinking water increased, a decrease in IQ and an increase in rates of mental retardation (IQ < 70) and borderline intelligence (IQ 70-79)". The correlation between IQ and level of fluoride in urine was statistically significant when measured directly and when adjusted for creatinine. Children in the village with low levels of fluoride showed an unexpected but significant decline of IQ with age. Family income and education level of parents did not correlate with children's IQ.

The study by Xiang et al. (2003) is severely deficient in its discussion of potential biases and confounders. The discussion section mentions the lack of association with family income and education, but does not discuss any other potential confounders. Lifestyle is mentioned in the methods but no results or discussion is presented. No explanation is provided for the unusual decline of IQ with age in the low fluoride exposed village, besides fluoride. No consideration was paid to where children lived within the village and how other factors might affect IQ. Known confounders such as intermarriage between close relatives are not discussed. The well known effect of parent's IQ was not measured except by education level which did not correlate suggesting this measure was inadequate for that purpose. Given the lack of exploration of other possible causes for the pattern of IQ, little weight can be placed on the results from this study. The study does mention 3 other studies which apparently support the current findings. These other studies are inadequately discussed in a single sentence without any mention of their strengths and weaknesses. Though the authors have apparently made a careful statistical analysis, their epidemiologic analysis is too incomplete to warrant any conclusion until other contributing factors, confounders, and biases are fully explored.

10. E.H. Goh and A.W. Neff, Effects of fluoride on Xenopus embryo development; Food and Chemical Toxicology 41:1501-1508, 2003

The effects of sodium fluoride (NaF) on the development of frog embryos were evaluated in this study. The concentrations of NaF used ranged from 0 to 20 mM. The authors of this study concluded that NaF acted as a teratogen and reported that most prominent malformations caused were reduction in the head-tail lengths and dysfunction of the neuromuscular system of tadpoles. It should not be concluded from this study that NaF also acts as a teratogen in mammalian fetuses to cause malformations. It should be noted that in a recent developmental toxicity of NaF by the Food and Drug Administration (Developmental toxicity of sodium fluoride measured during multiple generations. Collins TF, et al., Food Chem Toxicol. 2001 Aug;

39(8):867-76.), no dose-related anomalies or malformations in internal organs were observed. Additionally, numbers of corpora lutea, implants, viable fetuses and fetal morphological development were similar in all treatment groups. The only effect found was decreased ossification of the hyoid bone at 250 ppm (above the current MCL). FDA also evaluated NaF for reproductive effects and found no cumulative effects in three generations. (Multigenerational evaluation of sodium fluoride in rats. Collins TF, et al., Food Chem Toxicol. 2001 Jun;39(6):601-13). Furthermore, this frog embryo assay can not be used for the quantitative extrapolation of human potential risks. The FETAX assay is currently being reviewed by the Interagency Coordinating Committee on the Validation of Alternative Methods (ICCVAM), where it is being proposed only as a “screening method” for evaluating the developmental toxicity potential of chemicals.

Table 1a. Bibliography of Studies/Citations Referenced by the Fluoride Action Network in Response to Various Sulfuryl Fluoride *Federal Register* Notices

RD #	Study Title	Cited in Which Action?			Article does not contain primary quantitative data that can be used directly for dose-effect extrapolation to support the MCL.	Article contains primary quantitative data that may support dose-effect extrapolation evaluation. See text for a preliminary review of study and Attachments 1 & 2
		EUP Proposed Rule	Objections/ Hearing Request	In Response to Dow's §3 NOF		
1	Alarcon-Herrera MT et al (2001). Well water fluoride, dental fluorosis, and bone fractures in the Guadaine Valley of Mexico. Fluoride, 24(2): 138-148.		✔	✔		✔
2	ATSDR 1993. Toxicological profile for fluorides, hydrogen fluoride, and fluorine. Report No. TP-91/17.	✔			Govt. review	
3	Bachinskii PP et al. (1985) Action of the body fluorine of healthy persons and thyroidopathy patients on the function of hypophyseal-thyroid system. Probl Endokrinol (Mosk) 31(6):25-9.		✔	✔	Russian paper; abstract is in English. Without a translation, can not discern from the numberical data the significance of the influence of fluoride levels on thyroid-pituitary function. It is also not known to what extent there may have been fluoride sources other than drinking water. Predates the 1993 NAS report, thus contains no new information.	

RD #	Study Title	Cited in Which Action?			Article does not contain primary quantitative data that can be used directly for dose-effect extrapolation to support the MCL.	Article contains primary quantitative data that may support dose-effect extrapolation evaluation. See text for a preliminary review of study and Attachments 1 & 2
		EUP Proposed Rule	Objections/ Hearing Request	In Response to Dow's §3 NOF		
4	Baylet TA et al. (1990). Fluoride-induced factures: relation to osteogenic effect. J Bone Miner Res. Mar;5 Suppl 1:S217-22.			✔	An osteoporotic study using high doses of fluoride (60 mg/day) which are above the current EPA MCL .	
5	Benes FM et al. (1994). Myelination of a key relay zone in the hippocamal formation occurs in the human brain during childhood, adolescence, and adulthood. Arch Gen Psychiatry; 51: 477-484 (Cited by Filley, 2001).			✔	No data on fluoride.	
6	Brunelle JA and Carlos JP (1990). J. Dent. Res 69 (Special edition), 723-727.			✔	Trends on dental carries and water fluoriation.	
7	Byrd SE et al. (1993). White matter of the brain: maturation and myeliation on magenetic resonance in infants and children. Neuroimaging Clin N Am 3:247-266 (Cited by Filley, 2001).			✔	No data on the effects of fluoride are contained in this article.	

RD #	Study Title	Cited in Which Action?			Article does not contain primary quantitative data that can be used directly for dose-effect extrapolation to support the MCL.	Article contains primary quantitative data that may support dose-effect extrapolation evaluation. See text for a preliminary review of study and Attachments 1 & 2
		EUP Proposed Rule	Objections/ Hearing Request	In Response to Dow's §3 NOF		
8	Calderon J et al. (2000). Influence of fluoride exposures on reaction time and visuospatial organization in children. Epidemiology 11(4): S153.			✔	Abstract	
9	Canada Report 1993. Priority Substances List Assessment Report. Inorganic Fluorides. Government of Canada, Environment Canada, Health Canada, Canadian Environmental Protection Act. ISBN 0-662-21070-9 Cat. No. En40-215/32E-See: http://www.hc-sc.gc.ca/ehp/ehd/catalogue/bch_pubs/cepa/inorganic_fluorides.pdf	✔	✔		Govt. review	
10	Canada Report 1999. BENEFITS AND RISKS OF WATER FLUORIDATION. An Update of the 1996 Federal-Provincial Sub-committee Report. Prepared under contract for: Public Health Branch, Ontario Ministry of Health First Nations and Inuit Health Branch, Health Canada. Submitted by Dr. David Locker, Community Dental Health Services Research Unit, Faculty of Dentistry, University of Toronto, November 15, 1999). Report can be downloaded from: http://www.gov.on.ca/MOH/english/pub/ministry/fluoridation/fluoridation.html	✔	✔	✔	Govt. review	

RD #	Study Title	Cited in Which Action?			Article does not contain primary quantitative data that can be used directly for dose-effect extrapolation to support the MCL.	Article contains primary quantitative data that may support dose-effect extrapolation evaluation. See text for a preliminary review of study and Attachments 1 & 2
		EUP Proposed Rule	Objections/ Hearing Request	In Response to Dow's §3 NOF		
11	CDC 2001. Recommendations for using fluoride to prevent and control dental cariers in the United States. MMWR. August 17, 50(RR14):1-42.		✔	✔	Govt. review	
12	CDC 1999. Achievements in public health, 1990-1999: fluoridation of drinking water to prevent dental caries. MMWR, 48(41):933-940.		✔	✔	Govt. review	
13	Chen CJ (1988). Effects of fluoride on parathyroid hormone secretion and intracellular second messengers in bovine parathyroid cells. J Bone Miner Res 1 Jun; 3(3): 278-88.	✔			In vitro data that can not be used for dose-effect extrapolation	
14	Chinoy NJ, Sequeira E (1988). Effects of fluoride on the histoarchitecture of reproductive organs of the male mouse. Reprod Toxicol, 3(4): 261-7.			✔		✔
15	Chinoy NJ et al. (1991). Microdose vasal injection of sodium fluoride in the rat. Reprod Toxicol; 5(6):505-12.			✔	Direct injection into the vas deferens is not a relevant route of human exposure and thus should not be used for dose-effect extrapolation	

RD #	Study Title	Cited in Which Action?			Article does not contain primary quantitative data that can be used directly for dose-effect extrapolation to support the MCL.	Article contains primary quantitative data that may support dose-effect extrapolation evaluation. See text for a preliminary review of study and Attachments 1 & 2
		EUP Proposed Rule	Objections/ Hearing Request	In Response to Dow's §3 NOF		
16	Chinoy NJ, Narayana MV (1994). In vitro fluoride toxicity in human spermatozoa. Reprod Toxicol, Mar-Apr; 8(2):155-9.			✔	In vitro data that can not be used for dose-effect extrapolation	
17	Cohn PD (1992). An Epidemiologic Report on Drinking Water and Fluoridation. New Jersey Department of Health, Trenton, NJ.			✔	Govt review	
18	Colquhoun J (1997). Why I changed my mind on Fluoridation. Perspectives in Biology and Medicine, 41, 29-44. http://www.fluoride-journal.com/98-31-2/312103.htm			✔	Commentary on why dental fluorosis should be considered adverse.	
19	Colquhoun, J (1987). Studies of Child Dental Health Differences in New Zealand. Community Health Studies. 6(3): 85-90.			✔	Can not correlate fluoride levels and dental decay from the information provided in this paper.	
20	Connett E and Connett P (2002). Comments on Draft Toxicological Profile for Fluorides. Docket Control Number ATSDR-173. Submitted to: Division of Toxicology, Agency for Toxic Substances and Disease Registry, Mailstop E-29, 1600 Clifton Road, NE, Atlanta, Georgnia 30333. (Available at http://www.fluoridealert.org/pesticies/Fluorides.Comments.ATSDR.02.htm)		✔		Commentary on ATSDR document	

RD #	Study Title	Cited in Which Action?			Article does not contain primary quantitative data that can be used directly for dose-effect extrapolation to support the MCL.	Article contains primary quantitative data that may support dose-effect extrapolation evaluation. See text for a preliminary review of study and Attachments 1 & 2
		EUP Proposed Rule	Objections/ Hearing Request	In Response to Dow's §3 NOF		
21	Connett P and Connett M. (2000). The emperor has no clothes: a critique of the CDC's promotion of fluoridation. Waste Not #468. 82 Judson Street, Canton, NY 13617. http://www.fluoridealert.org/cdc.htm		✔	✔	Commentary by FAN	
22	DHHS (1991). Review of fluoride: benefits and risks, Report of the Ad Hoc Committee on Fluoride of the Committee to Coordinate Environmental Health and Related Programs. Department of Health and Human Services, USA.		✔	✔	Govt. review	
23	DHHS (1993). Fluoridation Census 1992. Published by the U.S. Department of Health & Human Services, Public Health Service, Centers for Disease Control and Prevention, National Center for Prevention Services, Division of Oral Health, Atlanta, Georgia 30333.	✔		✔	Govt. review	
24	DenBesten P (1999). Biological mechanism of dental fluorosis relevant to the use of fluoride supplements. Community Dent. Oral Epidemiol., 27, 41-7.			✔	No original data-Secondary data on exposure to fluoride and developmental windows (i.e., dose, duration, and timing)	

RD #	Study Title	Cited in Which Action?			Article does not contain primary quantitative data that can be used directly for dose-effect extrapolation to support the MCL.	Article contains primary quantitative data that may support dose-effect extrapolation evaluation. See text for a preliminary review of study and Attachments 1 & 2
		EUP Proposed Rule	Objections/ Hearing Request	In Response to Dow's §3 NOF		
25	Diesendorf M (1986). The mystery of declining tooth decay. Nature, 322, 125-129. http://www.fluoridealert.org/diesendorf.htm			✔	Commentary on relationship of dental decay and fluoride levels. No original data.	
26	Dyson, Rose and John R. Marier (1977). Environmental fluoride 1977. National Research Council of Canada. NRC Associate Committee on Scientific Criteria for Environmental Quality. NRCC No. 16081. ISSN 0316-0114.		✔		Review	
27	Emamghoreishi M et al (2000). Associated disturbances in calcium homeostatsis and G protein-mediated cAMP signaling in bipolar I disorder. Biol Psychiatry Oct 1; 48(7): 665-73.	✔			In vitro data that can not be used for dose-effect extrapolation.	
28	Emsley J et al.(1981). An unexpectedly strong hydrogen bond: ab initio calculations and spectroscopic studies of amide-fluoride systems. Journal of the American Chemical Society, 103, 24-28.			✔	This is a chemistry paper on hydrogen binding between fluoride and amines.	
29	Fein NJ and Cerlewski FL (2001). Fluoride content of foods made with mechanically separated chicked. J Agric Food Chem. Sep; 49(9):4284-6.		✔		An exposure study with no dose-response data	

RD #	Study Title	Cited in Which Action?			Article does not contain primary quantitative data that can be used directly for dose-effect extrapolation to support the MCL.	Article contains primary quantitative data that may support dose-effect extrapolation evaluation. See text for a preliminary review of study and Attachments 1 & 2
		EUP Proposed Rule	Objections/ Hearing Request	In Response to Dow's §3 NOF		
30	Filley CM (1998). The behavioural neurology of cerebral white matter. Neurology; 50:1535-1540. (Cited by Filley, 2001).			✔	No fluoride data.	
31	Filley CM (2001). The behavioural neurology of white matter. New York: Oxford University Press.			✔	Same as reference #30	
32	Flechsig P (1901). Developmental (myelogenetic) localisation of the cerebral cortex in the human subject. Lancet; 2:1027-1029 (Cited by Filley, 2001).			✔	No fluoride data	
33	Fletcher JM et al. (1992). Cerebral white matter and cognition in hydrocephalic children. Arch Neurol; 49:818-824. (Cited by Filley, 2001).			✔	No fluoride data	
34	Galletti P and Joyet G (1958). Effect of fluorine on thyroidal iodine metabolism in hyperthyroidism. Journal of Clinical Endocrinology; 18:1102-1110. http://www.fluoridealert.org/galletti.htm		✔	✔		✔
35	Graff I et al (1987). Carbachol and sodium fluoride, but not TSH, stimulate the generation of inositol phosphates in the dog thyroid. FEBS Lett Jan 5; 210(2):204-10.	✔			In vitro data that can not be used for dose-effect extrapolation	

RD #	Study Title	Cited in Which Action?			Article does not contain primary quantitative data that can be used directly for dose-effect extrapolation to support the MCL.	Article contains primary quantitative data that may support dose-effect extrapolation evaluation. See text for a preliminary review of study and Attachments 1 & 2
		EUP Proposed Rule	Objections/ Hearing Request	In Response to Dow's §3 NOF		
36	Gutteridge DH et al. (2002). A randomized trial of sodium fluoride (60 mg) Estrogen in Postmenopausal Osteoporotic Vertebral Fractures: Increased Vertebral Fractures and Peripheral Bone Loss with Sodium Fluoride; Concurrent Estrogen Prevents Peripheral Loss, But Not Vertebral Fractures. Osteoporosis International. Vol. 13 No. 2: 158-170.			✔	An osteoporotic study using high doses of fluoride (60 mg/day) which are above current EPA MCL	
37	Hattori Y (2000). Predominant contribution of the G protein-mediated mechanism to NAF-induced vascular contractions in diabetic rats: association with an increased level of G (qalapha) expression. J Parmacol Exp Ther Feb; 292(2):761-8.	✔			In vitro data that can not be used for dose-effect extrapolation	
38	Hedlund LR, Gallagher JC (1989). Increased incidence of hip fracture in osteoporotic women treated with sodium fluoride. J Bone Miner Res Apr; 4(2):223-5.			✔	Increased incidence of hip fracture in osteoporotic women found at fluoride levels (50mg/day) higher than current OW MCL.	
39	Heller KE et al. (1997). Dental caries and dental fluorosis at varying water fluoride concentrations. J of Pub Health Dent, 57; No. 3, 136-143.		✔	✔		✔

RD #	Study Title	Cited in Which Action?			Article does not contain primary quantitative data that can be used directly for dose-effect extrapolation to support the MCL.	Article contains primary quantitative data that may support dose-effect extrapolation evaluation. See text for a preliminary review of study and Attachments 1 & 2
		EUP Proposed Rule	Objections/ Hearing Request	In Response to Dow's §3 NOF		
40	Hirzy JW (1999). Why the EPA's headquarters union of scientists opposes fluoridation. Press release from National Treasury Employees Union, May 1, 1999. (for text see http://www.fluoridealert.org/hp-epa.htm)			✔	Commentary	
41	Institute of Medicine (NAS) (1997). Dietary reference intakes for calcium, phosphorus, magnesium, vitamin D, and fluoride. Standing Committee on the Scientific Evaluation of Dietary Reference Intakes, Food and Nutrition Board. National Academy Press.			✔	NAS review	
42	Jaumot M et al (2001). Protein phosphatases 1 and 2A promote Raf-1 activation by regulating 14-3-3 interactions. Oncogene Jul 5; 20(30): 3949-58	✔			In vitro data that can not be used for dose-effect extrapolation	
43	Kandel et al. (2000). Principles of neural science. 4th ed. New York: McGraw-Hill. (Cited by Filley, 2001).			✔	No new information on the adverse effects of fluoride	

RD #	Study Title	Cited in Which Action?			Article does not contain primary quantitative data that can be used directly for dose-effect extrapolation to support the MCL.	Article contains primary quantitative data that may support dose-effect extrapolation evaluation. See text for a preliminary review of study and Attachments 1 & 2
		EUP Proposed Rule	Objections/ Hearing Request	In Response to Dow's §3 NOF		
44	Klingberg T et al. (1999). Myelination and organization of the frontal white matter in children: a diffusion tensor MRI study. Neuroreport; 10:2817-2821. (Cited by Filley, 2001).			✔	No fluoride data	
45	Kumar A, Sushella AK (1994). Ultrastructural studies of spermiogenesis in rabbit exposed to chronic fluoride toxicity. Int J Fertil Menopausal Stud, May-Jun; 39(3):164-171.			✔	Study did not demonstrate that oral exposure to fluoride at a relatively high dose (4.5 mg/kg bw) can lead to abnormalities in spermatids and epididymal spermatozoa of rabbits. Only one dose was used. Also, insufficient information on methods and lack of data on the treatment of controls are weaknesses in the report.	
46	Lalumandier JA et al. (1995). The prevalence and risk factors of fluorosis among patients in a pediatric dental practice. Pediatric Dentistry - 17:1, 19-25.			✔	Does not provide any information on fluoride levels in drinking water or in supplements used by subjects, thus study is of limited value in evaluating levels of fluoride that are associated with dental fluorosis.	

RD #	Study Title	Cited in Which Action?			Article does not contain primary quantitative data that can be used directly for dose-effect extrapolation to support the MCL.	Article contains primary quantitative data that may support dose-effect extrapolation evaluation. See text for a preliminary review of study and Attachments 1 & 2
		EUP Proposed Rule	Objections/ Hearing Request	In Response to Dow's §3 NOF		
47	Lewis DW and Limeback H (1996). Comparison of recommended and actual mean intakes of fluoride by Canadians. Journal of the Canadian Dental Association; 62(9):708-709 and 712-715. Can be found at http://www.gov.on.ca/MOH/english/pub/ministry/fluoridation/fluor.pdf		✔		Review	
48	Li S, Zhi JL, Gao RO (1995). Effect of fluoride exposure on intelligence. Fluoride 28(4): 189-192.		✔		Number of children with IQs below 70 or between 70 to 90 was greater in the medium- and severe-fluorosis areas than in the no- and slight-fluorosis areas. Children or the environment not monitored for exposure to lead, methyl mercury, or other contaminants that can influence mental development. Coal burning can contribute mercury to the environment as well as fluoride. Authors did not correct for potential confounding variables e.g., poverty, parental education, or other characteristics that might have influenced results.	

RD #	Study Title	Cited in Which Action?			Article does not contain primary quantitative data that can be used directly for dose-effect extrapolation to support the MCL.	Article contains primary quantitative data that may support dose-effect extrapolation evaluation. See text for a preliminary review of study and Attachments 1 & 2
		EUP Proposed Rule	Objections/ Hearing Request	In Response to Dow's §3 NOF		
49	Li Y, C Liang et al. (2001). Effect of long-term exposure to fluoride in drinking water on risks of bone factures. J Bone Miner Res. May 16(5):932-9.		✔	✔		✔
50	Lin FF et al. (1991). The relationship of a low-iodine and high fluoride environment to subclinical cretinism in Zinjiang. Iodine Deficiency Disorder Newsletter 7.			✔	Confounding factors–low iodine could have resulted in cretinism. No data on diet.	
51	Lu Y et al. (2000). Effect of high-fluoride water on intelligence of children. Fluoride, 33, 74-78.		✔		Other environmental exposures that can influence mental development were not accounted for in this study (e.g., exposure to lead, methyl mercury, other pollutants). The authors also did not account for confounding factors/variables, e.g., poverty, parental education.	
52	Luke J (1997). The effect of fluoride on the physiology of the pineal gland. Ph.D. Thesis. University of Surrey, Guilford.		✔	✔		✔
53	Luke J (2001). Fluoride deposition in the aged human pineal gland. Caries Res. 35:125-128.	✔	✔	✔	No data associating fluoride exposure with adverse effects. Data from cadavers measuring fluoride deposition in pineal gland	

RD #	Study Title	Cited in Which Action?			Article does not contain primary quantitative data that can be used directly for dose-effect extrapolation to support the MCL.	Article contains primary quantitative data that may support dose-effect extrapolation evaluation. See text for a preliminary review of study and Attachments 1 & 2
		EUP Proposed Rule	Objections/ Hearing Request	In Response to Dow's §3 NOF		
54	Marcus W (1990). Memorandum from Dr. William Marcus to Alan B. Hais, Acting Director Criteria & Standards Division ODW, US EPA, DATED MAY 1, 1990, and subsequent memos. These can be viewed on the web at http://www.fluoridealert.org/marcus.htm			✔	Commentary	
55	McDonagh MS (2000): Systematic review of water fluoridation. BMJ 2000; 321:855-859 (7 October). York Review. NHS Center for Reviews and Dissemination, University of York, September 2000.		✔	✔	Review, no original data	
56	Michael M, Barot VV, Chinoy NJ (1996). Investigations of soft tissue functions in fluorotic individuals of North Gujarat. Fluoride; 29:2;63-71.			✔		✔

RD #	Study Title	Cited in Which Action?			Article does not contain primary quantitative data that can be used directly for dose-effect extrapolation to support the MCL.	Article contains primary quantitative data that may support dose-effect extrapolation evaluation. See text for a preliminary review of study and Attachments 1 & 2
		EUP Proposed Rule	Objections/ Hearing Request	In Response to Dow's §3 NOF		
57	Morgan L et al. (1998). Investigation of the possible associations between fluorosis, fluoride exposure, and childhood behavior problems. Pediatric Dentistry - 20:4, 244-252.			✔	Although refutes reference #58-by reporting no association between fluorosis, fluoride exposure and childhood behavior problems, this study alone can not resolve the issue of potential neurobehavioral problems. This study is not adequate to evaluate dental fluorosis and fluoride levels because it does not contain exposure data.	
58	Mullenix PJ et al. (1995). Neurotoxicity of sodium fluoride in rats. Neurotoxicology and Teratology. 17:2, 169-177.		✔	✔		✔
59	National Academy of Sciences (1997). Drinking water and health. National Academy Press, Washington, DC, pp. 388-389.			✔	NAS review	
60	National Research Council (1993). Health effects of ingested fluoride. National Academy Press, Washington, DC. Page 49.			✔	NAS review	

RD #	Study Title	Cited in Which Action?			Article does not contain primary quantitative data that can be used directly for dose-effect extrapolation to support the MCL.	Article contains primary quantitative data that may support dose-effect extrapolation evaluation. See text for a preliminary review of study and Attachments 1 & 2
		EUP Proposed Rule	Objections/ Hearing Request	In Response to Dow's §3 NOF		
61	National Toxicology Program (1990). Toxicology and carcinogenesis studies of sodium fluoride in F344/N rats and B6C3fl mice. Techical report Series No. 393. NIH Publ. No 91-2848. National Institute of Environmental Health Sciences, Research Triangle Park, N.C. The results of this study are summarized in the Department of Health and Human Services report (DHHS, 1991).		✔	✔	NTP cancer bioassay study which will not effect current MCL . This study was considered by the 1993 NAS Report.	
62	Nolte J (1999). The human brain. 4th ed. St. Louis: Mosby. (Cited by Filley, 2001).			✔	No data on fluoride	
63	Okajima F et al (1989). P2-purinergic agonists activate phospholiphase C in a guanine nucleotide- and Ca2+ dependent manner in FRTL-5 thyroid cell membranes. FEBS Lett Aug 14; 253(1-2):132-6.	✔			In vitro data that can not be used for dose-effect extrapolation	
64	Sayeski PP et al (2000). The role of Ca2+ mobilization and heterotrimeric G protein activation in mediating tyrosine phosphorylation signaling patterns in vascular smooth muscle cells. Mol Cell Biochem Sep; 212(1-2): 91-8.	✔			In vitro data that can not be used for dose-effect extrapolation.	

RD #	Study Title	Cited in Which Action?			Article does not contain primary quantitative data that can be used directly for dose-effect extrapolation to support the MCL.	Article contains primary quantitative data that may support dose-effect extrapolation evaluation. See text for a preliminary review of study and Attachments 1 & 2
		EUP Proposed Rule	Objections/ Hearing Request	In Response to Dow's §3 NOF		
65	Schlesinger ER et al. (1956). Newburgh-Kingston caries-fluorine study XIII. Pediatric findings after ten years. Journal of the American Dental Association, 52.		✔	✔	Historical study that has been considered by many groups, and thus will not impact MCL. This study was considered by the 1993 NAS report.	
66	Spak CJ et al (1983). Fluoride in human milk. Acta Paediatrica Scandinavica; V72, No 5, pp 699-701; September.	✔			An exposure study on fluoride concentrations in human milk. No dose-response data. This study was considered by the 1993 NAS report	
67	Strunecka A and Patocka J (1999). Pharmacological and toxicological effects of aluminofluoride complexes. Fluoride, 32, 230-242.	✔	✔	✔	Review on aluminofluoride complexes, no original data on fluoride	

RD #	Study Title	Cited in Which Action?			Article does not contain primary quantitative data that can be used directly for dose-effect extrapolation to support the MCL.	Article contains primary quantitative data that may support dose-effect extrapolation evaluation. See text for a preliminary review of study and Attachments 1 & 2
		EUP Proposed Rule	Objections/ Hearing Request	In Response to Dow's §3 NOF		
68	Susheela AK, Jethanandani P (1996). Circulating testosterone levels in skeletal fluorosis patients. J Toxicol Clin Toxicol, 34(2): 183-9.			✔	An epidemiological study comparing testosterone levels in 3 groups: fluorosis patients with 3.9 ppm F in their water, controls with high F in their water(4.5 ppm) and controls with low F in their water.(0.5 ppm). According to the authors, fluorosis patients had the lowest serum testosterone, with the high water F controls. However, confounding factors (e.g., age, diet, health status, exposure to other chemicals) were not accounted for that could affect testosterone levels.	
69	Sutton P (1959). Fluoridation: errors and omissions in experimental trials. Melbourne University Press. First Edition.			✔	Predates the 1993 NAS report	
70	Sutton P (1960). Fluoridation: errors and omissions in experimental trials. Melbourne University Press. Second Edition.			✔	A view on the benefits of drinking fluoridation and prevention tooth decay.	
71	Sutton P (1996). The Greatest Fraud: Fluoridation. Lorne, Australia: Kurunda Pty. Ltd.			✔	A view on the benefits of drinking fluoridation and prevention tooth decay.	

RD #	Study Title	Cited in Which Action?			Article does not contain primary quantitative data that can be used directly for dose-effect extrapolation to support the MCL.	Article contains primary quantitative data that may support dose-effect extrapolation evaluation. See text for a preliminary review of study and Attachments 1 & 2
		EUP Proposed Rule	Objections/ Hearing Request	In Response to Dow's §3 NOF		
72	Tezelman S (1994). Desensitization of adenylate cyclase in Chinese hamster ovary cells transfected with human thyroid-stimulating hormone receptor. Endocrinology Mar; 134(3):1561-9.	✔			In vitro data that can not be used for dose-effect extrapolation.	
73	Thrane EV et al (2001). Fluoride-induced apoptosis in epithelial lung cells involves activation of MAP kinases p38 and possibly JNK. Toxicol Sci May; 61(1):83-91.	✔			In vitro data that can not be used for dose-effect extrapolation.	
74	van der Knaap et al. (1991). Myelination as an expression of the functional maturity of the brain. Deb Med Child Neurol; 33:849-857. (Cited by Filley, 2001).			✔	No data on fluoride.	

RD #	Study Title	Cited in Which Action?			Article does not contain primary quantitative data that can be used directly for dose-effect extrapolation to support the MCL.	Article contains primary quantitative data that may support dose-effect extrapolation evaluation. See text for a preliminary review of study and Attachments 1 & 2
		EUP Proposed Rule	Objections/ Hearing Request	In Response to Dow's §3 NOF		
75	Varner JA et al. (1998). Chronic administration of aluminum-fluoride or sodium-fluoride to rats in drinking water; alterations in neuronal and cerebrovascular integrity. Brain Research, 784, 284-298.	✔	✔	✔	Before the results of this study are used for drawing inferences regarding human risk, this study needs to be repeated due to several major limitations, e.g., absence of dose-response assessment and evaluation of functional impairments, and lack of assessment of fluoride in the animal chow. With respect to the renal effects, it is important to distinguish those caused by aging versus fluoride treatment.	
76	Williams JE et al. (1990). Community Water Fluoride Levels, Preschool Dietary Patterns, and The Occurrence of Fluoride Enamel Opacities. J of Pub Health Dent; 50:276-281.			✔	This study was considered by the 1993 NAS report	✔
77	WHO (Online). WHO Oral Health Country/Area Profile Programme. Department of Noncommunicable Diseases Surveillance/Oral Health. WHO Collaborating Centre, Malmo University, Sweden. http://www.whocollab.od.mah.se/euro.html		✔	✔	WHO review-survey on fluoridation in different countries and benefits	

RD #	Study Title	Cited in Which Action?			Article does not contain primary quantitative data that can be used directly for dose-effect extrapolation to support the MCL.	Article contains primary quantitative data that may support dose-effect extrapolation evaluation. See text for a preliminary review of study and Attachments 1 & 2
		EUP Proposed Rule	Objections/ Hearing Request	In Response to Dow's §3 NOF		
78	Yakovlev PI, Lecours AR (1967). The myelogenetic cycles of regional maturation of the brain. In: Minkowki A, ed. Regional development of the brain in early life. Oxford: Blackwell Scientific Publications, 3-79. (Cited by Filley, 2001).			✔	No data on fluoride	
79	Yiamouyiannis JA (1990). Water fluoridation and tooth decay: Results from the 1986-1987 national survey fo U.S. schoolchildren. Fluoride, 23, 55-67.			✔	Survey of dental records from 84 areas throughout the US. No actual exposure data on fluoride levels, simply reported as fluoridation, partial fluoridation versus non-fluoridation.	
80	Zhao LB et al. (1996). Effect of high-fluoride water supply on children's intelligence. Fluoride, 29, 190-192.		✔	✔	There were more children with IQs less than 80 in the high fluoride village (25) than in the low fluoride village (9). As was the case with the study by Li et al.(1995), the potential for exposures to chemicals e.g., lead and methyl mercury which have a demonstrated effect on IQ was not assessed and the data were not corrected for possible confounding variables	

RD #	Study Title	Cited in Which Action?			Article does not contain primary quantitative data that can be used directly for dose-effect extrapolation to support the MCL.	Article contains primary quantitative data that may support dose-effect extrapolation evaluation. See text for a preliminary review of study and Attachments 1 & 2
		EUP Proposed Rule	Objections/ Hearing Request	In Response to Dow's §3 NOF		
81	Zhao W (1998). Long-term effects of various iodine and fluorine doses on the thyroid and fluorosis in mice. Endocr Regul 32(2):63-70.			✔		✔

Table 1b. SULFURYL FLUORIDE - SUPPLEMENTARY REFERENCES

RD #	STUDY TITLE	Cited in Which Action			Article does not contain primary quantitative data that can be used directly for dose-effect extrapolation to support the MCL.	Article contains primary quantitative data that may support dose-effect extrapolation evaluation. See text for a preliminary review of study and Attachments 1 -3
		EUP Proposed rule	Objections/ Hearing request	In response to Dow's §3 NOF		
82	Ast, DB, Smith DJ, Wachs B, et al. (1956). Newburgh-Kingston caries-fluorine study: XIV. Combined clinical and roentgenographic dental findings after ten years of fluoride experience. J. Am Dent. Assoc. 52:314-325.			✔	Historical paper considered by the 1993 NAS	
84	BNA. (2000). http://www.fluoridealert.org/alum-fluoride.htm (cited by Connett - 07/OPP#PF-1068 page 10/26)			✔	web page not available 10/29/03: commentary	

RD #	STUDY TITLE	Cited in Which Action			Article does not contain primary quantitative data that can be used directly for dose-effect extrapolation to support the MCL.	Article contains primary quantitative data that may support dose-effect extrapolation evaluation. See text for a preliminary review of study and Attachments 1 -3
		EUP Proposed rule	Objections/ Hearing request	In response to Dow's §3 NOF		
85	Caffey J. 1956.(cited in: National Academy of Sciences. 1977. Drinking water and health. National Academy Press. pp. 388-389.) {Work probably reported in Schlesinger 1956 [ref 65]. "In light of the concern today about the possibility that fluoride might increase the risk of osteosarcoma in young males, an intriguing piece of history occurred in 1956 when Dr. Caffey examined the health data collected in the 1945-55 Newburg vs Kingston fluoridation trial [Schlesinger, 1956]. He noticed a greater percentage of cortical bone defects in the bones of the children in fluoridated Newberg compared to unfluoridated Kingston."}			✔	Historical paper considered by the 1977 NAS	
87	CDC. Chart 1, p. xx, **Fluoridation Census** (1992). Centers for Disease Control and Prevention, National Center for Prevention Services, Division of Oral Health, Atlanta, Georgia 30333.	✔			No health or toxicological data	

RD #	STUDY TITLE	Cited in Which Action			Article does not contain primary quantitative data that can be used directly for dose-effect extrapolation to support the MCL.	Article contains primary quantitative data that may support dose-effect extrapolation evaluation. See text for a preliminary review of study and Attachments 1 -3
		EUP Proposed rule	Objections/ Hearing request	In response to Dow's §3 NOF		
89	Freni SC. (1994). Exposure to high fluoride concentrations in drinking water is associated with decreased birth rates. Journal of Toxicology and Environmental Health, 42:109-12.			✔	Study did not measure factors affecting fertility ,e.g., contraceptive practices. Cohort effect prior to 1980 not measured by use of single point estimates for key measures of sociodemographic status. An ecologic study that measured county levels of fluoride rather than individual level and cannot be used to show a causal association. Author acknowledges need for follow up study on individual women to verify results.	
90	Full, C.A. and Parkins, F.M. (1975). Effect of cooking vessel composition on fluoride. J. Dent. Res. 54: 192.	✔			Exposure article with no toxicology data on Fluoride	
91	Goh EH, Neff AW. (2003) Effects of fluoride on Xenopus embryo development. Food Chem Toxicol. 2003 Nov;41(11):1501-8.					The frog embryo assay can not be used for the quantitative extrapolation of human potential risks. Other laboratories in well conducted studies in mammalian have not found fluoride to be a teratogen. See Attachment 3

RD #	STUDY TITLE	Cited in Which Action			Article does not contain primary quantitative data that can be used directly for dose-effect extrapolation to support the MCL.	Article contains primary quantitative data that may support dose-effect extrapolation evaluation. See text for a preliminary review of study and Attachments 1 -3
		EUP Proposed rule	Objections/ Hearing request	In response to Dow's §3 NOF		
92	Guan ZZ, Y.N. Wang, K.Q. Xiao, D.Y. Dai, Y.H. Chen, J.L. Liu, P. Sindelar and G. Dallner. (1998). Influence of chronic fluorosis on membrane lipids in rat brain. Neurotoxicology and Teratology 20 537-542.			✔	No brain histopathology and changes in membrane lipids above current MCLs	
93	http://www.fluoridealert.org/pesticides/Sulfuryl.F.Mar.2002comments.htm Table 2: Results from a March 18, 2002, search of the EPA Office of Pesticide Program site for "White Matter."			✔	No original data, citations from EPA website	
94	http://www.nofluoride.com/presentations/presentations.htm (cited by L001/OPP#PF-1068 p 1 of 3. "toxicity charts which have been presented to the congressional committee on science by your own EPA scientists and representatives from the NRDC." site not available 10/31/03)			✔	Presentation-commentary	
95	http://www.sarep.ucdavis.edu/BIFS/bifs01/annual.htm#walnut (re % of California walnut crop that is exported. cited in Ellen Connett submission dated September 29, 2001. site not available 10/29/03)			✔	Commentary	

RD #	STUDY TITLE	Cited in Which Action			Article does not contain primary quantitative data that can be used directly for dose-effect extrapolation to support the MCL.	Article contains primary quantitative data that may support dose-effect extrapolation evaluation. See text for a preliminary review of study and Attachments 1 -3
		EUP Proposed rule	Objections/ Hearing request	In response to Dow's §3 NOF		
96	Li L. (2003). The biochemistry and physiology of metallic fluoride: action, mechanism, and implications. Crit Rev Oral Biol Med. 14(2):100-14.					This review does not provide new information on the levels of fluoride or fluoride complexes that might be associated with adverse effects in laboratory animals or humans dose-levels of fluoride associated with adverse effects. See Attachment 3
100	Matsuo S, Kiyomiya K, Kurrebe M. (1998). Mechanism of toxic action of fluoride in dental fluorosis: whether trimeric G proteins participate in the disturbance on intracellular transport of secretory ameloblast exposed to fluoride. Arch Toxicol 1998 Dec; 72(12); 798-806.			✔	Mechanism paper on dental fluorosis using doses higher than the current secondary MCL	
102	Ortiz-Perez D, et al. (2003). Fluoride-induced disruption of reproductive hormones in men. Environmental Research 93(1):20-30.					This study suffers from a poor design and inadequate sampling. See Attachment 3

RD #	STUDY TITLE	Cited in Which Action			Article does not contain primary quantitative data that can be used directly for dose-effect extrapolation to support the MCL.	Article contains primary quantitative data that may support dose-effect extrapolation evaluation. See text for a preliminary review of study and Attachments 1 -3
		EUP Proposed rule	Objections/ Hearing request	In response to Dow's §3 NOF		
104	Rathwell PJ. (1997). Per capita fruit consumption continues to increase. Clemson University. Outlook Update 332. Oct.9, 1997. http://cherokee.agecon.clemson.edu/otlk332.htm (re raisin consumption as % of dried fruit consumed in US. Cited in Ellen Connett submission dated September 29, 2001.)			✔	Exposure article with no toxicological data on fluoride	
107	Shames R, Shames KH. (2002). **Thyroid Power: Ten Steps to Total Health** HarperResource			✔	Opinion article with no original data on fluoride	
108	Shashit A. (2003). Histopathological investigation of fluoride-induced neurotoxicity in rabbits. Fluoride 36: 95-105.					Because of confounding factors, the results in this study do not add to the weight of the evidence for the potential of sodium fluoride to induce adverse effects in humans (see Attachment 3)

RD #	STUDY TITLE	Cited in Which Action			Article does not contain primary quantitative data that can be used directly for dose-effect extrapolation to support the MCL.	Article contains primary quantitative data that may support dose-effect extrapolation evaluation. See text for a preliminary review of study and Attachments 1 -3
		EUP Proposed rule	Objections/ Hearing request	In response to Dow's §3 NOF		
110	Staub M et al. (1998) Deoxycytidine kinase can be also potentiated by the G-protein activator NaF in cells. Adv Exp Med Biol 1998; 431:425-8			✔	Because the effects of fluoride on biochemical parameters were determined in vitro, it is not possible to determine the dose that would lead to the effects in vivo.	
111	Susa M. (1999). Heterotrimeric G proteins as fluoride targets in bone (review). Int J Mol Med. Feb; 3(2):115-26.			✔	Review article, not an original study	
112	Takahashi K, Akiniwa K, Narita KJ. (2001). Regression analysis of cancer incidence rates and water fluoride in U.S.A. based on IACR/IARC (WHO) data (1978-1992). Epidemiol 2001 Jul; 11(4):170-9.	✔				Given the extreme methodological shortcomings of this study, no conclusions can be drawn from this paper. See Attachment 1
113	Vincent S et al. (1998). Evidence for distinct mechanisms of transition state stabilization of GTPases by fluoride. Proc Natl Acad Sci USA 1998 Mar 3; 95(5):2210-5.			✔	The effects of fluoride on biochemical parameters were determined in vitro, thus, it is not possible to determine the dose that would lead to the effects in vivo.	
114	Wasner HK et al. (2000). Two different mechanisms for activation of cyclic PIP synthase by a G protein or by protein tyrosine phosphorylation. Biol Chem 2000 Feb; 381(2):145-53.			✔	The effects of fluoride on biochemical parameters were determined in vitro, thus, it is not possible to determine the dose that would lead to the effects in vivo.	

RD #	STUDY TITLE	Cited in Which Action			Article does not contain primary quantitative data that can be used directly for dose-effect extrapolation to support the MCL.	Article contains primary quantitative data that may support dose-effect extrapolation evaluation. See text for a preliminary review of study and Attachments 1 -3
		EUP Proposed rule	Objections/ Hearing request	In response to Dow's §3 NOF		
115	Weber S, Lemoine H, Wasner HK. (2000). Prostaglandin deficiency promotes sensitization of addenylyl cyclase. Biol Chem 2000 May-June; 381(5-6):525-9			✔	Because the effects of fluoride on biochemical parameters were determined in vitro, it is not possible to determine the dose that would lead to the effects in vivo. Furthermore, no association between the dose that induced the biochemical changes in vitro and toxic effects was shown. This report does not provide information for evaluating any in vivo toxicity that may result from perturbations of AMP synthesis or cyclic AMP activity following exposures to fluoride.	
116	Xiang Q, et al. (2003). Effect of fluoride in drinking water on children's intelligence. Fluoride 36: 84-94.					This study is severely deficient in its discussion of potential biases and confounders. See Attachment 3