Assessing and managing fluorosis risk in children and adults in rural Madhya Pradesh, India

Sam Godfrey, Pawan Labhasetwar, Tapas Chakma, Satish Wate, Aditya Swami and Jamie Bartram

ABSTRACT

This paper presents the application of quantitative chemical risk assessment for assessing and managing fluorosis in 19 schools and 6 villages in Madhya Pradesh, India. A longitudinal study was undertaken with a baseline survey in 2005 and an endline in 2007. Household surveys, water quality and food analysis were undertaken to measure the impact of an Integrated Fluorosis Mitigation programme that included water and nutritional interventions. The baseline survey indicated a maximum fluoride content of 7.8 mg/l in food and 3.7 mg/l in water, equating to a maximum fluoride uptake of 4.8 and 3.7 mg/l in food and water respectively. Mean (actual) daily intake of fluoride for all exposure routes was 0.4 mg/kg of combined adult and child body weight. Intake of fluoride through food was more than 40% of total intake. Calculated guideline values for age groups <18 years and >18 years were 1.7 and 1.9 mg/l respectively. Using WHO methodology, the Guideline Value would be 1.7 mg/l. Fluoride dilution was implemented to reduce the fluoride content to below this level. The endline survey indicated reduction in the prevalence of grade 1 fluorosis of 86%, of grade 2 of 77%, of grade 4 of 60% in all children examined.

Key words | fluorosis, health-based targets, quantitative chemical risk assessment

Sam Godfrey (corresponding author)
Water and Environmental Sanitation,
UNICEF,
Maputo,
Mozambique
E-mail: *sgodfrey@unicef.org*

Pawan Labhasetwar
Satish Wate
Aditya Swami
National Environmental Engineering Research Institute,
Nagpur,
India

Tapas Chakma
Regional Medical Research Centre for Tribals,
Jabalpur,
India

Jamie Bartram
Gillings School of Global Public Health,
University of North Carolina at Chapel Hill,
NC,
USA

INTRODUCTION

According to the World Health Organization, fluorosis is prevalent in more than 20 countries around the world and is normally associated with areas with high levels of fluoride in water (Fawell *et al.* 2006). The global burden of disease due to fluoride in drinking water has further been estimated by Fewtrell *et al.* (2006) by combining exposure–response curves for dental and skeletal fluorosis (derived from published data) with model-derived predictions of drinking water fluoride concentrations. This study estimated that approximately 18,197,000 people are affected with dental fluorosis in India alone (Fewtrell *et al.* 2006). Areas requiring fluorosis mitigation are typically identified on the basis of dental and skeletal fluorosis manifestations; they often occur in crystalline biotite/hornblende gneiss geological zones, which are laden with fluorite, tourmaline, sphene and topaz (Driscoll 1986). Mitigation measures include source substitution, blending of high and low fluoride-content waters, and community and household level defluoridation units using diverse media such as ion exchange, activated charcoal and/or charred bone meal (NEERI 2007). However, despite global efforts to combat fluorosis, there is an increase in the prevalence of fluorosis worldwide (Fewtrell *et al.* 2006).

Dental and skeletal fluorosis are common manifestations of excess fluoride intake. Dental fluorosis occurs as the permanent teeth are forming and is characterised by permanent hypo-mineralization (Mullins *et al.* 1998; Murray & Wilson 1948). Very elevated intake of fluoride over prolonged periods of time may result in skeletal fluorosis, i.e. an accumulation of fluoride in the skeletal tissue, associated with pathological bone formation. Fluoride is also an essential nutrient and insufficient fluoride intake is associated with increased rates of dental caries.

Studies by Cao *et al.* (1997, 2006), Cao (2003), Lian Feng (1995) and Mwaniki *et al.* (1994) identified consumption in

doi: 10.2166/washdev.2011.101

food as a potentially significant route of fluoride exposure and state that consumption of certain foods may increase the risk of fluorosis. The International Programme on Chemical Safety has noted that all vegetation contains some fluoride, which is absorbed from soil and water (IPCS 2002). The highest levels in vegetables are found in curly kale (up to 40 mg/kg fresh weight) and endive (0.3–2.8 mg/kg fresh weight) (Slooff 1988). Other foods containing high levels include fish (0.1–30 mg/kg) and tea (Slooff 1988). In India, Chakma *et al.* (2000) reported presence of fluoride in 70 different commonly consumed foodstuffs and habit-forming substances from Mandla District of Madhya Pradesh.

Fluoride in food comes from the plant uptake of fluoride from water including irrigation water. Uptake is greater in tropical climates due to the high level of soil to crop evapotranspiration Singh *et al.* (1995). This can be extremely high in tea: for example, Chakma *et al.* (2000) reported presence of fluoride in a local brand tea of 19.36 μg/g of dry leaves in Mandla District, Madhya Pradesh, India. This is reinforced by Michael *et al.* (2005) who showed that instant tea in distilled water has a fluoride concentration of 3.3 mg/l. Furthermore, brick tea prepared in China, which is fermented and then compressed into bricks, has a fluoride content of 590–780 mg/kg (Sha & Zheng 1994).

The exposure of an individual to fluoride is dependent on the bioavailability of fluoride in food and water. Bioavailability is the proportion of a chemical available for uptake into the systemic circulation of a given target organism following a given mode of exposure. The bioavailability of fluoride in food has been researched by Toth & Sugar (1978), Rao (1984) and Trautner & Einwag (1987). Rao (1984) suggests that fluoride intake from foods in the USA is increasing and it is essential to develop bioavailability profiles for individual foods. Studies by Trautner & Einwag (1987) of factors influencing the bioavailability of fluoride from calcium-rich, health-food products in humans estimated a maximum bioavailability of 60%.The bio-availability of fluoride in water has been widely studied and the literature indicates that 95% of bioavailable fluoride comes from water (Whitford 1996).

Studies by Erdal & Buchanan (2005) on the risk of fluorosis in the USA, identified children as the most affected segment of the population in that country. They state that young children are highly susceptible to skeletal and non-skeletal manifestations of fluorosis due to the low calcium strength in early bone development in infants as well as excess exposure to fluoride in toothpaste and food/water. This is reinforced by Chakma *et al.* (2000) in Mandla, Madhya Pradesh, India, who reported children as young as six years to be suffering from skeletal fluorosis. The clinical manifestations of skeletal fluorosis in children are knock knee or bow leg, while adults present with stiffness of the neck and back muscles.

This paper, therefore, explores methods of quantifying the health risk from excess exposure to fluoride in water and food, including disaggregated analysis to account for the risk for children. Based on the generic approach outlined in the third edition of the Guidelines for Drinking-water Quality (WHO 2004), this paper applies risk assessment principles for establishing health-based targets. While health targets and outcomes are local or national in character, they can be informed by risk analysis, which provides a means to support the development of nationally relevant targets, adapted to specific local conditions.

Work undertaken by Fewtrell *et al.* (2006) attempted to estimate the disease burden associated from fluorosis. This paper explores the application of quantitative chemical risk assessment (QCRA) to setting health-based targets.

METHODS

The study was undertaken in six villages and nineteen schools of the tribal districts of Dhar and Jhabua of the central Indian State of Madhya Pradesh. These tribal schools and communities were selected as they represent the communities at highest risk of fluorosis. The major source of water in these communities is groundwater. Drinking water availability through these sources is less than 40 l per person per day in the dry months between January and June.

To investigate the prevalence of fluorosis, communities and schools were randomly selected for study in two districts. A baseline survey was undertaken in 2005. There were repeat visits to the same group (i.e. a quasi-longitudinal study) in 2007. Households and individuals were selected in these six communities using the statistical *t*-test based on two comparable means from two variables: existing levels of fluoride contamination (mg/l) and population served. Using the *t*-test, the number of households required for the QCRA with 95% confidence level of estimating average levels of contamination within 2% of the true value was calculated using Equations (1) and (2):

$$n_0 = \frac{\{tS\}^2}{rY^2} \quad (1)$$

where n_0 first approximation of sample size, t confidence probability (t statistics); this value is 1.96 for confidence probability of 95%, S population standard deviation, r relative error, Y population mean.

$$n = \frac{n_0}{(1 + (n_0/N))} \quad (2)$$

where n sample size, N population size, n_0 first approximation sample size (see Equation (1)) (Table 1).

The QCRA approach taken was based on WHO guidelines and comprised: hazard identification, dose-response assessment, exposure assessment and risk characterization (WHO 2004; NEERI 2007). The severity of skeletal fluorosis was determined separately for children (8–18 years) and adults (>18 years). For adults (>18 years), the calibrated prescribed physical exercise indicated in Figure 1 was undertaken (Susheela 2001).

Susheela (2001) notes that the above exercises determine muscle stiffness in the back, neck, shoulder, calf and thighs of an individual. Susheela (2001, 2005) also notes that in endemic areas persons with stiff muscles are not able to perform muscular exercises and are considered to be suffering from skeletal fluorosis. These exercises were also used by Chakma *et al*. (2000) as a tool while identifying fluorosis in another district of Madhya Pradesh and validated with

Table 1 | Families and schools selected by the *t*-test

No	Name of school (Dhar district)	No. of students	No	Name of school (Jhabua district)	No. of students
1	G.A. Mandu	187	1	Dhekalbadi	115
2	G.A. Teetipura	40	2	Kalidevi	342
3	G.A. Malipura	51	3	Rama	67
4	G.A. Nalcha	177	4	Gadwada	211
5	G.A. Sodhpur	99	5	Ban	110
6	N.G.A. Kakalpura	180	6	Jhakela	20
7	Sikarpura	43	7	Senalia	24
8	Kheri	40	8	Rajla	19
9	Kanya Ashram Ukala	53	9	Kokawat	92
10	Ganganagar	130		Total	1,000
	Total	1,000			

Village/school	Fluoride level range (mg/l)	Population (families)	*t*-test families
Jhabua			
Tikai Jorgie	1.7–4.16	1,209	26
Charolipada	2.03–8.83	1,001	21
Pithanpur	2.72–9.44	2,036	39
Dhar			
Aadvi	0.6–2.6	530	30
Shikarpura	2.4–5.07	730	35
Jamanpati	4.01–8.46	768	30

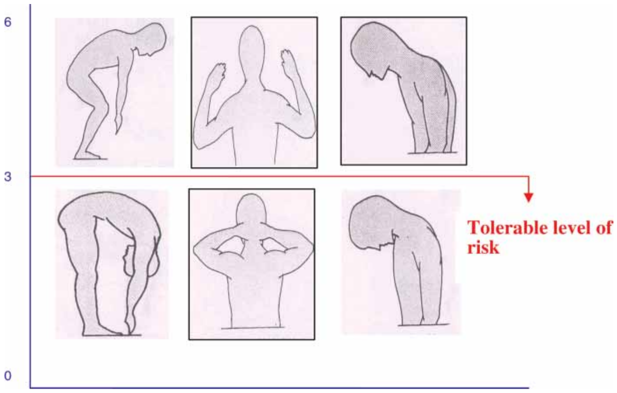

Figure 1 | Muscle exercise for determination of skeletal fluorosis among adults.

radiological and biochemical parameters. The same method was used for children under 5 years of age.

Fluorotic lesions on teeth are usually bilaterally symmetrical and tend to show striated patterns across the tooth. The recording of dental fluorosis was done according to Dean's dental fluorosis index. The codes and criteria used are shown in Table 2.

Table 2 | Dean's classification (excluded e.g. a crowned tooth and not recorded)

No	Category	Description
0	Normal	The enamel surface is smooth, glossy and usually a pale, creamy-white colour.
1	Questionable	The enamel shows slight aberrations from the translucency of the enamel, which may range from few white flecks to occasional spots.
2	Very mild	Small, opaque, paper-white areas scattered irregularly over the tooth but involving less than 25% of the labial tooth surface.
3	Mild	The white opacity of the enamel of the teeth is more extensive than for code 2, but covers less than 50% of the tooth surface.
4	Moderate	The enamel surface of the teeth shows marked wear and brown stain is frequently a disfiguring feature.
5	Severe	The enamel surfaces are badly affected and hypoplasia is so marked that the general form of the tooth may be affected. There are pitted or worn areas and brown stains are widespread; the teeth often have a corroded appearance.

Fluoride exposure

Fluoride exposure can be through food, water, soil and air. In the study region, uptake of fluoride through air pollution (both indoor and outdoor) is negligible. The study was undertaken in rural areas of devoid of any industrial activities. As there are no industrial sources in the study villages, it is safe to presume that fluoride intake through air pollution is negligible.

Water exposure

For water exposure, the National Environmental Engineering Research Institute, Nagpur, India collected water samples following methods described in AWWA & APHA (1998). One litre of water was collected from the water source that was reported to be the primary drinking water source in the village in plastic containers to avoid possible contamination from the borosilicate present in glass bottles. Samples were analysed using the ion selective electrode (ISE) method. First, the electrode slope of the ion meter (54–60 mV/decade) was checked as per the specifications of the ORION 5*™. For each standard (1.0 mg/l, 10.0 mg/l and 100.0 mg/l fluoride), 10 ml were added to 1 ml TISAB III to calibrate the instrument. After calibration, 10ml of the sample was added to the beaker and stirred with a magnetic stirrer with 1 ml of TISAB III. The electrode is then rinsed with distilled water after completion of each sample analysis and the reading is taken.

Food exposure

For food exposure a dietary survey was conducted to determine dietary practices. The survey involved a household survey which included a questionnaire on the types and quantities of foodstuffs consumed on a daily basis. A further biometric test of the age and weight of each household member was then undertaken. Based on household interviews, foodstuffs consumed at the household level were collected in 50 g samples in marked plastic bags. The samples were dried at 163–165 °C in a hot air oven in a borosilicate evaporating dish. After cooling, the dried material was powdered and passed through a 40-mesh sieve. One gram of this material was diluted with 5 ml concentrated hydrochloric acid in a 150 ml plastic beaker and mixed for 1 hr; 100 ml of buffer solution was added and a sample extract was then filtered and stirred for 3 min by a magnetic stirrer. The millivolt reading is taken using an ion selective electrode to measure the fluoride concentration.

Total daily intake

An estimated total daily intake (TDI) was calculated using Equation (3):

$$\mathrm{TDI} = \frac{(\mathrm{NOAEL\ or\ LOAEL})}{\mathrm{UF}} A \tag{3}$$

where NOAEL and LOAEL are no-observed-adverse-effect level and lowest-observed-adverse-effect level, UF stands for uncertainty factor.

In this study the LOAEL value was determined by referring to the minimum total daily intake value at which there is an observed effect of fluorosis. An uncertainty factor of 1 was considered for determining TDI.

To determine the most susceptible age group, two age groups i.e. <18, and >18 were selected. For each a guideline value was calculated using Equation (4):

$$\mathrm{GV} = \frac{(\mathrm{TDI} \times \mathrm{BW} \times P)}{C} \tag{4}$$

where GV, Guideline value; TDI, Total daily intake; BW, Body weight; P, Fraction of TDI fraction allocated to drinking water; C, Daily drinking-water consumption.

Risk characterization

In this study, DALY (disability adjusted life years) were calculated based on methods outlined in Havelaar & Melse (2003) with background data from Murray & Lopez (1996). The following equation was used to determine DALY due to dental and skeletal fluorosis:

$$\mathrm{DALY} = \mathrm{YLL} + \mathrm{YLD} \tag{5}$$

where YLL, Years of life lost due to premature mortality, YLD, Years lived with disability.

As noted by Fewtrell *et al.* (2006), fluorosis is not considered as fatal; therefore the DALY calculation is based on YLD (years lived with disability) only and estimated on a population basis as suggested by Havelaar & Melse (2003). In this case, the number of disability cases is multiplied by the average duration of the disease and a weight factor by using the following basic formula (without applying social preferences) for one disabling event is:

$$\mathrm{YLD} = I \times \mathrm{DW} \times L \tag{6}$$

where I, Number of incident cases; DW, Disability weight, L, Average duration of disability (years).

The following assumptions are made to estimate DALY due to skeletal and dental fluorosis:

1. Onset of dental fluorosis is considered to be 6–8 years. Dean's Classification was used to classify dental fluorosis; Class 3 and above is considered to be indicate cases of dental fluorosis.
2. Onset of skeletal fluorosis is considered to be 3 years from the date of continuous consumption. Chakma *et al.* (2000) reported that children 3 years and above also developed skeletal fluorosis in Mandla District of Madhya Pradesh, India.
3. The severity of dental fluorosis is considered to be low (0.003) and constant with age as suggested by Fewtrell *et al.* (2006).
4. The severity of skeletal fluorosis is relatively high and increases with age (0.24 to 0.50) (Fewtrell *et al.* 2006). The severity weight of 0.24 is assumed to apply up to the age of 18, while the weight of 0.5 is assumed to apply to those aged 18 and above (Murray & Lopez 1996) considering average life expectancy of 55 years in Madhya Pradesh, India (Government of India 2001).

Integrated fluorosis mitigation programme

Water intervention

The dilution of groundwater contaminated with fluoride using collected rainwater was carried out so as to ensure the level of fluoride was below the threshold limit. Rooftop rainwater harvesting may not be sufficient to supply potable water for 365 days in a year, but provides a low-fluoride source with which to dilute other higher-fluoride water sources. A dilution technique is applied to mix rainwater collected through rainwater harvesting and groundwater containing fluoride to ensure availability of safe water for drinking and cooking purposes. A schematic of dilution technology is shown in Figure 2.

This may include a 1:2 dilution to reduce the concentration of fluoride of 3 to 1 mg/l. To dilute the contaminated groundwater, the groundwater and rainwater are blended in two overhead tanks.

Nutrition intervention

The calcium-rich crop *chakoda bhaji* (*Cassia tora*) was promoted. This vegetable is commonly found in rural areas of India and has a high calcium content (3,200 mg/100 g of dry leaves). Consumption of this crop has the following advantages:

a) High content of calcium, iron, vitamin C and other nutritional elements
b) Commonly grown in rural areas
c) Can be used for consumption throughout the year after drying.

Data analysis

Data was processed and analysed using EPI-info software programme. The results indicate the impact of the Integrated Fluorosis Mitigation Programme on the level of dental fluorosis in children in the cohort. The results are divided into Dhar and Jhabua and presented in the Results and Discussion section below.

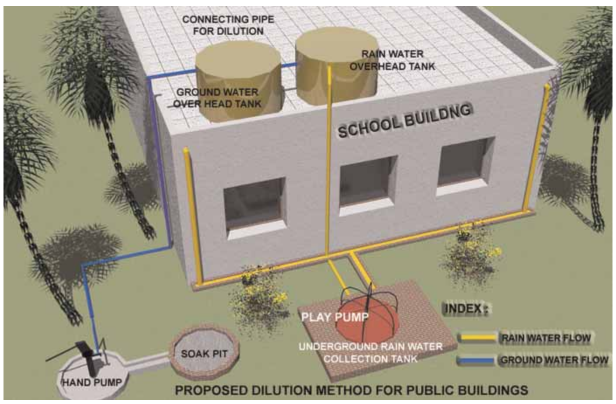

Figure 2 | Dilution technology.

RESULTS AND DISCUSSION

To estimate the exposure of the population to the hazard (fluoride in water and food), the size and the nature of the population exposed and the route, amount and duration of exposure were calculated.

Hazard identification

Firstly, the hazard identification undertaken in 2005 indicated that the concentration of fluoride in water ranged from 0.3 to 1.5 mg/l in the schools and communities selected. Similarly, fluoride in food samples ranged from 0.005 to 1.1 mg/kg dry weight. Further analyses of fluoride content in food and water undertaken in 2007 indicated that fluoride in water varies between 0.5 and 3.7 mg/l. and in food between 3.3 and 7.8 mg/kg.

The bioavailability of fluoride in food is dependent on whether the food is consumed raw or cooked. Studies from Ethiopia by Malde *et al*. (2004) note that a considerable amount of fluoride is retained in food prepared in high fluoride cooking water. This therefore emphasises the need to include food contaminated with fluoride when considering the total exposure. Figure 3 indicates that rice has a high content of fluoride due to: (a) content of rice per se since this increased between 2005 and 2007 it was attributed to high uptake of fluoride by rice paddy fields when irrigated with fluoride-contaminated irrigation water and (b) uptake of fluoride when rice is boiled in fluoride-rich water.

Exposure assessment and dose response

Firstly, the exposure assessment indicated that 49.9% persons are affected with dental fluorosis and 11.8% persons were affected with skeletal fluorosis. Dental fluorosis was common among children (8–18 years), whereas adults (above 20 years) contribute majority of the total skeletal fluorosis cases.

Secondly, the exposure assessment is required to determine the TDI for different age groups, from food and water.

The TDI value for all exposure routes was observed to be in the range 0.37 to 0.42 mg/kg of body weight.

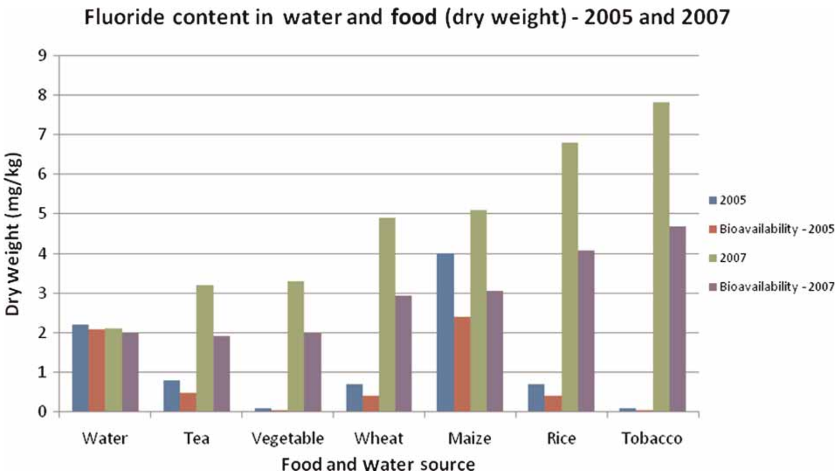

Figure 3 | Fluoride content in water and food (dry weight) (2005 and 2007).

The weight of students (<18 years) varied from 16 to 48 kg and water intake varied between 3 and 5 litres per day per child, whereas weight of adults (>18 years) varied from 34 to 65 kg and water intake varied between 3 and 8 litres per day per person. The calculated guideline values for age group <18 years and >18 years are 1.7 and 1.9 mg/l respectively.

Risk characterization

Data indicated that 49.9% persons among studied population are affected with dental fluorosis, whereas 9.3% and 2.5% persons are affected with skeletal fluorosis below and above 40 years respectively. DALYs due to dental fluorosis, skeletal fluorosis (<40 years) and skeletal fluorosis (>40 years) are estimated to be 8, 112 and 62 per 1,000 persons.

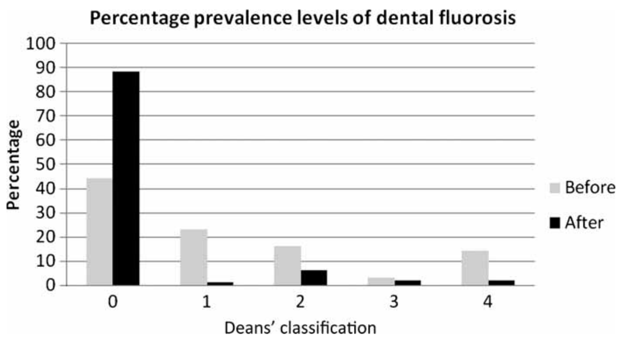

Figure 4 | Percentage reduction in levels of dental fluorosis.

Fewtrell *et al.* (2006) estimated an average DALY of 15 per 1,000 persons-year in India based on estimate of 1.8% of dental fluorosis and 0.72% skeletal fluorosis cases. The DALYs for fluorosis in Madhya Pradesh, India are higher than those reported by Fewtrell *et al.* (2006) due to the fact that the study in Madhya Pradesh is undertaken in a fluorosis-endemic area having a higher proportion of the fluorosis-affected population.

Integrated fluorosis mitigation programme

Figure 4 shows that there is a reduction in the prevalence of grade I fluorosis of 22%, for grade II there is reduction by 10%, grade III 1% and grade 4 12%. Overall there is a significant reduction in the 0 prevalence of dental fluorosis from 44% to 88% which is attributable to the dilution of water and change in nutritional status.

The study was undertaken in a limited area and it is recommended that it is repeated in other geographic areas based on a conservative screening approach that considers the hydrogeology and recorded cases of fluorosis.

CONCLUSIONS

This study concludes that to control fluorosis total daily consumption from all exposure routes (water and food) must be considered, quantifying chemical risk is required for appropriate risk management strategies for reducing fluorosis and that reduction of dental fluorosis is possible through the application of appropriate mitigation measures.

REFERENCES

AWWA & APHA 1998 *Standard Methods for the Examination of Water and Wastewater*. American Water Works Association, McGraw-Hill, New York.

Cao, J. 2003 Brick tea fluoride as a main source of adult fluorosis. *Food Chem. Toxicol.* **41**, 535–542.

Cao, J., Zhao, Z. & Liu, J. W. 1997 Brick tea consumption as the cause of dental fluorosis among children from Mongol, Kazak and Yugu Populations in China. *Food Chem. Toxicol.* **35**, 827–833.

Cao, J., Zhao, Y., Li, Y., Deng, H. J., Yi, J. & Liu, J. W. 2006 Fluoride levels in various black tea commodities: measurement and safety evaluation. *Food Chem. Toxicol.* **44**, 1131–1137.

Chakma, T., Rao, P. V., Singh, S. B. & Tiwary, R. S. 2000 Endemic genuvalgum and other bone deformities in two villages of Mandla District in Central India. *Fluoride* **33**, 187–195.

Dean, H. T. 1934 Classification of mottled enamel diagnosis. *J. Am. Dent. Assoc.* **21**, 1421–1426.

Driscoll, F. 1986 *Groundwater and Wells*. 2nd edition, Reynolds Guyar Designs, USA.

Erdal, S. & Buchanan, S. N. 2005 A quantitative look at fluorosis, fluoride exposure, and intake in children using a health risk assessment approach. *Environ. Health Perspect.* **113**, 111–117.

Fawell, J., Bailey, K., Chilton, J., Dahi, E., Fewtrell, L. & Magara, Y. 2006 *Fluoride in Drinking-water*. WHO/IWA, UK

Fewtrell, L., Smith, S., Kay, D. & Bartram, J. 2006 An attempt to estimate the global burden of disease due to fluoride in drinking water. *J. Water Health* **4**, 533–542.

Government of India 2001 National Census. Government of India, New Delhi.

Havelaar, A. & Melse, J. 2003 *Quantifying public health risk in the WHO Guidelines for Drinking Water Quality – A burden of disease approach*. RIVM Report 734301022.

IPCS 2002 *Fluorides, Environmental Health Criteria 227*. International Programme on Chemical Safety, World Health Organization, Geneva, Switzerland.

Lian-Feng, W. & Jian-Zhong, H. 1995 Outline of control practices of endemic fluorosis in China. *Soc. Sci. Med.* **41**, 1191–1195.

Malde, M. K., Zeribon, L., Julsham, K. & Bjovatin, K. 2004 Fluoride, calcium and magnesium intake of children living in a high fluoride area in Ethiopia: intake through food. *Int. J. Pediatr. Dent.* **14**, 167–174.

Michael, P., Whyte, M. D., Kevan Essmyer, M. S., Francis, H., Gannon, M. D., William, R. & Reinus, M. D. 2005 Skeletal fluorosis and instant tea. *Am. J. Med.* **118**, 78–82.

Mullins, M., Warden, C. & Barnum, D. 1998 Pediatric death and fluoride-containing wheel cleaner [Letter]. *Ann. Emerg. Med.* **31**, 523–525.

Murray, C. & Lopez, A. 1996 *The Global Burden of Disease*. WHO/World Bank, Harvard School of Public Health, Cambridge, Massachusetts.

Murray, M. M. & Wilson, D. C. 1948 Fluorosis and nutrition in Morocco: dental studies in relation to environment. *Br. Dent. J.* **84**, 97–100.

Mwaniki, D., Courtney, M. & Gaylor, D. 1994 Endemic fluorosis: and analysis of needs and possibilities based on case studies in Kenya. *Soc. Sci. Med.* **39**, 807–813.

NEERI 2007 *Integrated Fluorosis Mitigation – Guidance Manual*. National Environmental Engineering Research Institute, Nagpur, India.

Rao, G. S. 1984 Dietary intake and bioavilibity of fluoride. *Ann. Rev. Nutr.* **4**, 115–136.

Sha, J. Q. & Zheng, D. X. 1994 Study on the fluorine content in fresh leaves of tea plant planted in Fujian Province. *J. Tea Sci.* **14**, 37–42.

Singh, V., Gupta, M. K., Rajwanthy, P., Mishra, S., Srivastava, S., Srivastava, R., Srivastava, M., Prakash, S. & Dass, S. 1995 Plant uptake of fluoride in irrigation water by ladies finger (*Abelmorchus esculentus*). *Food Chem. Toxicol.* **33**, 399–402.

Slooff, W. 1988 *Basisdocument fluoriden*. Bilthoven, Netherlands, National Institute of Public Health and Environmental Protection (Report No. 758474005).

Susheela, A. K. 2001 *A Treatise on Fluorosis*. 2nd edition, Fluorosis Research and Rural Development Foundation, New Delhi, India Publication.

Susheela, A. K. 2005 *Fluorosis – an easily preventable disease through practice of interventions – safety & diet supplementation*. Ministry of Health and Family Welfare, Government of India, WHO India Country Office, New Delhi, India.

Toth, K. I. & Sugar, E. 1978 Fluorine content of foods and estimated daily intake from foods. *Acta Physiol. Acad. Sci. Hung.* **51**, 361–369.

Trautner, K. & Einwag, J. 1987 Factors influencing the bioavailability of fluoride from calcium-rich, health-food products and CaF2 in man. *Arch. Oral. Biol.* **32**, 401–406.

WHO 2004 *Guideline for Drinking Water Quality*. 3rd edition. World Health Organization, Geneva.

Whitford, G. M. 1996 *Metabolism and Toxicity of Fluoride*. Karger, Basel, Vol. 1, pp. 12–15, 46–48.

First received 11 January 2011; accepted in revised form 26 May 2011