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Journal of Nanjing Medical University, 2009, 23(1):33–39
Research paper

JNMU
www.elsevier.com/locate/jnmu

Relationships between daily total fluoride intake and dental fluorosis and dental caries☆

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Received 2 November 2008

Abstract

Objective: To explore the relationships between the daily total intake of fluoride, dental fluorosis and dental caries. **Methods:** An epidemiological method was used to investigate the daily total intake of fluoride, dental fluorosis, and dental caries among 236 and 290 children aged 8-13 years in a severe endemic area and in a non-fluorosis control area, respectively. The children were divided into eight subgroups according to each child's estimated daily total intake of fluoride. The prevalence of dental fluorosis and dental caries in each group was calculated. **Results:** As expected, elevated levels of fluoride intake were significantly associated with a higher prevalence of dental fluorosis and an increasing amount of more severe defect dental fluorosis. When the daily total F intake was 2.78 mg/child/day, the prevalence of dental fluorosis was nearly 100%, with the prevalence of defect dental fluorosis increasing with increasing fluoride intake. There was also a significant negative(inverse) dose-response relationship between the daily total intake of fluoride and the overall prevalence of dental caries, the prevalence of which decreased when the daily total intake of fluoride increased up to 3.32 mg/child/day. However, at higher levels of daily total intake of fluoride the prevalence of dental caries increased, giving rise to a U-shaped dose-response relationship curve. **Conclusion:** It is important to monitor total fluoride exposure and protect children from excessive fluoride intake, especially during the years of tooth development.

Key words: Fluoride; Daily total fluoride intake; Dental fluorosis; Dental caries

INTRODUCTION

A 2002 World Health Organization review of dental surveys conducted in the United States over 60 years ago by H Trendley Dean and co-workers of the US Public Health Service concluded that the "optimum" level of fluoride(F) in drinking water associated with maximum protection against dental caries(DC) and a minimum amount of dental fluorosis(DF) was approximately 1 mg F ion/L[1]. Ever since these surveys, drinking water with 1 mg F/L has been promoted as an effective way to reduce DC, especially among children. During Dean's studies, the primary source of F intake was considered to be drinking water[2]. In the past 30-40 years, however, there has been a considerable increase in the prevalence of DF among populations consuming non-fluoridated as well as fluoridated drinking water[1,3-5]. Although greater numbers of people are now living in communities with "optimally" fluoridated(0.7-1.3 mg F/L) drinking water, much of this increased prevalence of DF has been attributed to the widespread intake of F from sources other than drinking water, such as foodstuffs, toothpaste, fluoridated milk, dietary supplements, fluoridated salt, mouth rinses, and brick tea[6-8].

Unfortunately, there is limited information to characterize the dose-response relationships among the different health effects of F. In particular, there is only

☆ This work was supported by Jiangsu Provincial Foundation of Prevention Medicine(Y200621).
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limited information on total F exposure in relation to dental health[1]. In this study, the daily total F intake, DF and DC in children were investigated in two rural villages of China, and the relationships between the daily total intake of F and the prevalence of DF and DC were analyzed.

MATERIALS AND METHODS

As in our recent studies[9], the present investigation was conducted between September 2002 and June 2004 in two villages(Wamiao and Xinhuai), 64 km apart, in Sihong County, Jiangsu Province, People's Republic of China, and during this time the investigation of age, gender, DF, and DC was conducted from Feb. 2003 to June 2003. Wamiao village(drinking water F mean, standard deviation and range in mg/L: 2.45 ± 0.80, 0.57-4.50), in northeast Sihong County, about 32 km northeast of Sihong is located in a severe endemic fluorosis area, whereas Xinhuai village(drinking water F mean, standard deviation and range in mg/L: 0.36± 0.15, 0.18-0.76), in the southwest part of Sihong County, about 32 km southwest of Sihong, is in a non-endemic area. Neither village has F pollution from burning coal or other industrial F sources. None of the residents drink brick tea.

All the children, 8-13 years old, were examined in the primary schools of both villages. A questionnaire, completed with the assistance of the parents, was used to collect information on personal characteristics, F exposure history, medical history, family socioeconomic status, and lifestyle. Children who had been absent from either village for two years or longer, or who had a history of immigration were excluded. In Wamiao 99.16% of the children(236 out of 238) were included, while in Xinhuai 95.08% were included(290 out of 305).

The teeth were examined with a mouth mirror and an explorer under natural light by a dentist and a specialist in endemic fluorosis control and prevention. Dean's classification of six grades of DF[10] and the Chinese Clinical Diagnostic Standard for dental fluorosis(WS/T208-2001)[11] were used to diagnose dental fluorosis. In the WS/T 208-2001 Chinese Standard, DF is divided into eight grades. Grades 5 (including a small amount pitting on the enamel surface) and above are considered "defect dental fluorosis" (DDF). Statistical analysis of the prevalence of DF was made according to the percentage of DF by Dean's classification and the percentage of DDF by the WS/T208-2001 scheme. DDF included some "moderate" DF (grade 3) and all "severe" DF(grade 4) diagnosed using the Dean scale.

Diagnosis of DC was conducted according to the criteria of Fan[12] (superficial caries, moderate caries, and deep caries). Because caries in the children was most common on the coronal surface[13], we used coronal decay for measuring caries activity.

$$\text{Prevalence of dental caries (DC\%)} = \frac{\text{No. of children with coronal DC}}{\text{No. of children examined}} \times 100$$

Water samples(100 mL) were collected from each child's household shallow well, and the F content in the water was measured with a F ion selective electrode according to the Chinese National Standard Method for determination of fluoride in drinking water of endemic fluorosis areas(WS/T 106-1999)[14].

Samples of staple food (rice, bread, etc., over 50 g) and vegetables(over 100 g) were collected from various families selected at random by diet investigators. The samples were kept in clean plastic bags that were immediately sent to the laboratory for analysis. Fluoride in food was determined according to the Chinese National Standard Method for determination of fluoride in foods(GB/T 5009.18-1996)[15], again with a F ion selective electrode.

The investigation of F intake was conducted according to the Chinese National Standard of Hygienic Standard for daily total intake fluoride(WS/T 87-1996)[16]. The methods of inquiry and weighing were used to investigate the daily total intake of various foods, water, boiled water, tea, etc. of each person in the family. The questionnaire was designed according to the Investigation of the Status of Nutrition and Health of Chinese Residents, 2002[17]. The families in Wamiao village were divided into five groups according to the F concentration in the drinking water of the household well: < 1.00 mg/L, ~1.00 mg/L, ~2.00 mg/L, ~3.00 mg/L, and = 4.00 mg/L. In each group, 6 families were randomly sampled for the dietary investigation. In Xinhuai, 6 families were also randomly sampled for the dietary investigation. The dietary investigation was performed by trained and certified investigators.

Five families were also selected in each village according to the location, number of people, and construction style of the house to check for indoor F air pollution. Each village was divided into three portions along the east-west direction, and the air samples were collected at the east, middle, and west sites for assaying outdoor air F levels. Flouride in air samples was measured as described in the Chinese Stationary Source Emission-Determination of fluoride-ion selective electrode method(HJ/T 67-2001)[18]. The air was sampled at noon each day for 20 min, and the sampling lasted for 5 days.

The data collected were analyzed by SPSS, Excel, and Curve Experts Software.

Before the investigation, an informed consent had to be signed by each child's parents. We have complied with all requirements of international regulations for human investigation.

RESULTS

The mean F concentration in the household shallow well water in Wamiao village was 2.45 ± 0.80 mg/L (236 samples; range 0.57-4.50 mg/L), and in Xinhuai village, it was 0.36 ± 0.11 mg/L(290 samples; range 0.18-0.76 mg/L). There was a significant difference between two villages in the drinking water F(t = 45.13, P < 0.001), as reported earlier[9]. The F in indoor and outdoor air was too low to measure.

For examination of the effect of boiling the water on F concentration, a total of 50 water samples in Wamiao and 11 in Xinhuai were collected from household shallow wells at random at the same time that they were sampled to determine the F in the drinking water. Each sample was divided into two parts, one being un-treated and the other being boiled. As shown in ***Table 1***, the F concentration of boiled water from typical household shallow wells was lower than that in the crude water. In both villages, iron pots were used to heat the water, and the mean values of total hardness of shallow groundwater in the household wells of the two villages were over 300 mg/L(the total hardness was measured by the GB750-85 using the EDTA method, which mainly tests the total calcium and magnesium ion contents, and the result is expressed as the equivalent quantity of calcium carbonate).

There were significant differences in the rice F and bread F levels between the two villages. As seen in Table 1, F concentrations in rice and bread in Wamiao were higher than those in Xinhuai. On the other hand, vegetable F concentrations in the two villages were not significantly different(***Table 2***).

Table 1 **F concentration in crude and boiled water from typical household shallow wells, rice, and bread in Wamiao and Xinhuai(M±SD)**

Village	Crude water		Boiled water		Rice		Bread	
	No. of samples	Fluoride (mg/L)	No. of samples	Fluoride (mg/L)	No. of samples	Fluoride (mg/kg)	No. of samples	Fluoride (mg/kg)
Wamiao	50	2.40 ± 0.76	50	1.87 ± 0.79	40	1.17 ± 0.43	36	1.49 ± 0.33
Xinhuai	11	0.41 ± 0.08*	11	0.33 ± 0.006†	9	0.78 ± 0.19**	9	1.09 ± 0.15††

Compared with Wamiao village, *t=8.51, P < 0.001; †t=6.43, P < 0.001. **t=2.70, P < 0.01;††t=3.44, P < 0.01.

Table 2 **F concentrations(mg/kg) in vegetables in Xinhuai and Wamiao**

Village	Crude water		Boiled water		Rice		Bread	
	No. of samples	Fluoride (M ± SD)	No. of samples	Fluoride (M ± SD)	No. of samples	Fluoride (M ± SD)	No. of samples	Fluoride (M ± SD)
Wamiao	6	0.19 ± 0.06	6	0.19 ± 0.02	6	0.21 ± 0.07	6	0.45 ± 0.09
Xinhuai	6	0.20 ± 0.07	6	0.15 ± 0.03	6	0.20 ± 0.06	6	0.44 ± 0.04

The results of the dietary investigation indicated that the total daily average food intake of 8-13 year-old children was 644 g in the two villages, including 395.5 g of staple food(rice and bread, accounting for 61.32% of the total), 171.0 g of various vegetables(26.51%), 77.5 g of other foods (meat, fish, egg, and fruit etc., 12.01%). As shown in ***Table 3***, the estimated F intake per child per day from food was 0.609 mg/day in Wamiao and 0.424 mg/day in Xinhuai.

Results of analyses indicated that the children's average daily total intake of water in the two villages was 156 mL crude water and 1085 mL boiled water. Tea intake was included in the boiled water. For children in the 2 villages, the average tea intake per-day was only 2 mL.

The F intake was calculated according to Appendix A of the Hygienic Standard for Daily Total Fluoride Intake(WS/T 87-1996): daily total intake of F(mg/person/day)=intake of F from diet + intake of F from drinking water + intake of F from air. In this way, the estimated average F intake of per day per child in Wamiao was 3.05 ± 0.99 mg(range: 0.73-5.57 mg), and 0.78 ± 0.13 mg (range: 0.57-1.27 mg) in Xinhuai. The estimated F intake in the children in Wamiao village was significantly higher than that in Xinhuai village and was 0.64 mg/child/day above the allowable value of National Standard(WS/T 87-1996)(which is 2.41 mg/person/day for children below 15 years of age in the

Table 3 **Daily total intake of food and F from food per child in Wamiao and Xinhuai**

Food	Daily total intake of food (g/day)	F intake(mg/day)	
		Wamiao	Xinhuai
Bean	83.0	0.016	0.017
Capsicum	31.0	0.006	0.005
Aubergine	24.5	0.005	0.005
Potato	32.5	0.015	0.014
Rice	274.0	0.321	0.216
Bread	121.5	0.181	0.122
Others*	77.5	0.065	0.045
Total	644.0	0.609	0.424

*Others food included meal, fish, egg, fruit, and so on.

high drinking water fluoride endemic fluorosis areas).

The prevalence of DF, DDF, and DC were 88.56% (209/236), 38.56%(91/236), and 37.71%(89/236), respectively in Wamiao village; and were 4.48%(13/290), 0.00%(0/290), 56.90(165/290) in Xinhuai village. The prevalence of DF and DDF in the children in Wamiao was significantly higher than in Xinhuai(DF: $c^2 = 9.0$, $P < 0.01$. DDF: $c^2 = 135.2$, $P < 0.001$), but the overall prevalence of DC among children in Xinhuai was significantly higher than that in Wamiao($c^2=19.18$, $P < 0.001$).

For further analysis of the data, the 526 children in the two villages were considered together and divided into eight subgroups according to their estimated daily total intake of F: < 1.00 mg/child(group A), ~1.00 mg/child(group B), ~1.50 mg/child(group C), ~ 2.00 mg/child(group D), ~2.50 mg/child(group E), ~3.00 mg/child(group F), ~3.50 mg/child(group G), ⩾ 4.00 mg/child(group H). The prevalence of DF, DDF, and DC were then calculated for each subgroup.

As the F intake increased, the prevalence of DF and DDF also increased(see ***Table 4*** and ***Fig. 1,2***), which indicated a positive relationship between the F intake and the prevalence of DF and DDF. When the F intake reached 2.78 mg/child/day, the prevalence of DF was nearly 100%. The prevalence of DDF increased continuously as the F intake increased to 4.50 mg/child/day. ***Fig. 1***, ***2*** represented the best-fit curves between the F intake and the prevalence of DF and DDF by logistic and linear fit models. The regression coefficients were 0.99 and 0.92, respectively, and the fitting equations were $y = 93.05/(1+149.68e^{-3.64x})$, and $y = -14.44 + 17.04x$, respectively.

The prevalence of DC decreased as the F intake

Table 4 **F intake/child and the prevalence of DF and DDF in Wamiao and Xinhuai children combined**

Group	No. of samples	F intake (M ± SD) [mg/child]	No.with DF	No.with DDF	Prevalence of DF(%)	Prevalence of DDF(%)
A	268	0.75 ± 0.09	11	0	4.10	0.00
B	41	1.15 ± 0.12	13	2	31.71	4.87
C	19	1.80 ± 0.11	14	0	73.68	0.00
D	27	2.31 ± 0.15	25	11	92.59	40.74
E	50	2.78 ± 0.16	48	18	96.00	36.00
F	46	3.32 ± 0.13	43	21	93.48	45.65
G	38	3.76 ± 0.13	33	16	86.84	42.11
H	37	4.50 ± 0.42	35	23	94.59	62.16

increased up to 3.32 mg/child/day. However, above this level of intake, the prevalence of DC increased. As seen in ***Table 5*** and in ***Fig. 3***, the relationships between F intake and the prevalence of DC thus produced polynomial U-shape curves(regression coefficient 0.90).

When the children in each village were divided into subgroups according to age, the prevalences of DF and DDF in each age group in Wamiao were significant higher than those in Xinhuai, which corresponded with the estimated F intake(***Tables 6***,***7***).

As seen in ***Table 6***, the prevalence of DF and DDF in each age group in Wamiao village showed slight variation. However, there were no significant differences between the different age groups. The prevalence of caries in the 8-year-old group was the highest, and was significantly different from the 11- and 12-year-old groups. Overall, there was no significant association between DC prevalence and age(Pearson Correlation = −0.698, $P = 0.123$).

In Xinhuai village, there were no significant differences in terms of the prevalence of DF and DDF in each age group. As age increased, the prevalence of DC decreased, and there was a significant negative relationship between age and the prevalence of DC (Pearson Correlation = −0.944, $P = 0.05$, ***Table 7***).

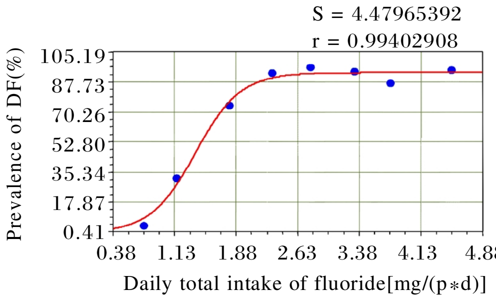

Fig. 1 Logistic curve fit relationship between F intake /child and the prevalence of DF

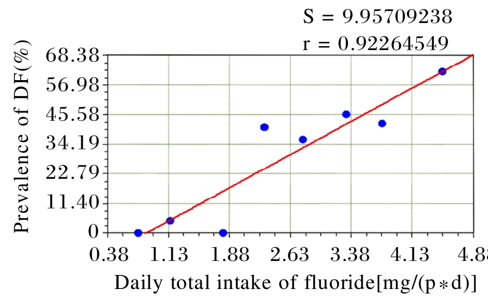

Fig. 2 Linear fit relationship between F intake/child and the prevalence of DDF

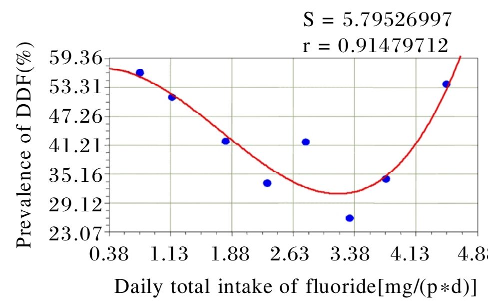

Fig. 3 Polynomial curve of the relationship between F intake/child and the prevalence of DC in Wamiao and Xinhuai ildren combined

Table 5 **F intake/child/day and the prevalence of DC in Wamiao and Xinhuai children combined**

Group	No. of samples	F intake(M ± SD) [mg/child]	No.with DC	Prevalence of DC(%)
A	268	0.75 ± 0.09	151	56.34
B	41	1.15 ± 0.12	20	48.78
C	19	1.80 ± 0.11	8	42.11
D	27	2.31 ± 0.15	9	33.33
E	50	2.78 ± 0.16	21	42.00
F	46	3.32 ± 0.13	12	26.09
G	38	3.76 ± 0.13	13	34.21
H	37	4.50 ± 0.42	20	54.05

Except for the 13-year-old group, the prevalences of DC in Xinhuai were higher than those in Wamiao, but the difference was significant only in the 9-year-old group($\chi^2 = 4.82$, $P = 0.028$).

DISCUSSION

Wamiao and Xinhuai are located in isolated low-income areas of Jiangsu Province of China with very little economic development and a relative lack of communication with the outside world[19]. The diet of the villagers is relatively simple and lacking in refined food. In this study, the results indicated that in the two villages, for children aged 8-13 years of age, the staple food (rice and bread) accounted for 61.32%, vegetables 26.51%, and the others(meat, fish, egg, fruit, etc.) only 12.01% of the total diet. Therefore, it was relatively easy to investigate and estimate the F intake, and to analyze the relationship between the F intake and dental health effects. In our previous report, the fluoride in drinking water in these two villages was considered the main source of F intake, and the findings of F levels in the drinking water and serum therefore probably reflected the F exposure of the children during infancy and early childhood[9].

Table 6 **F intake /child(mg/child) and the prevalence of DF, DDF, and DC by age in Wamiao**

Age	F intake (M ± SD)	No. of children	No.with DF	No. with DDF	No. with caries	Prevalence of DF(%)	Prevalence of DDF(%)	Prevalence of DC(%)
8	3.27 ± 1.24	20	18	5	12	90.00	25.00	60.00
9	2.92 ± 0.83	16	13	4	7	81.25	25.00	73.75
10	3.16 ± 0.88	19	18	7	9	94.74	36.84	47.37
11	3.21 ± 0.75	39	35	18	10	89.74	46.15	25.64
12	2.97 ± 1.03	69	60	23	21	86.96	33.33	30.43
13	2.67 ± 1.04	73	65	34	30	89.04	46.58	41.10
Total	3.05 ± 0.99	236	209	91	90	88.56	38.56	38.14

Table 7 **F intake/child(mg/child) and the prevalence of DF, DDF, and DC by age in Xinhuai**

Age	F intake (M ± SD)	No. of children	No.with DF	No. with DDF	No. with caries	Prevalence of DF(%)	Prevalence of DDF(%)	Prevalence of DC(%)
8	0.78 ± 0.13	39	1	0	30	2.56	0.00	76.92
9	0.80 ± 0.12	46	1	0	34	2.17	0.00	73.91
10	0.75 ± 0.10	31	1	0	23	3.23	0.00	74.19
11	0.77 ± 0.13	60	5	0	32	8.33	0.00	53.33
12	0.75 ± 0.10	61	4	0	25	6.56	0.00	40.98
13	0.77 ± 0.13	53	1	0	21	1.89	0.00	39.62
Total	0.78 ± 0.13	290	13	0	165	4.48	0.00	56.90

In both villages, the residents always use an iron pot to cook food and boil water. The mean values of the total hardness of shallow groundwater in the household wells in two villages were over 300 mg/L. Whether the iron pot and the total hardness of the drinking water cause the decrease in F in the boiled water or not requires further investigation, since Wilkister *et al.* reported that the boiling had no effect on the levels of fluoride in the water[20].

Established literature indicates that the intake of F has a positive relationship with the prevalence of DF and a negative relationships with the DC[13, 21-24]. In our study, the relationship between the estimated F intake and the prevalence of DC revealed a U-shape relationship, as was the case in a previous study[25]. However, the work of Ermis *et al*[26] indicated that increasing water F levels were associated with higher prevalence and severity of dental fluorosis and had no influence on the caries experience of children with poor oral hygiene. According to Meyer-Lueckel *et al*[27], ingestion of naturally fluoridated water(1.3 mg F/L) appeared to have a negligible effect on caries prevalence, but clearly resulted in a higher prevalence of dental fluorosis. The results of the study conducted by Nohno et al. indicated that the mean DMFT(diseased, missing and filled teeth) score in the moderate fluoride area (the total daily fluoride intakes were 0.0252-0.0254 mg F/kg/day) was significantly lower than that in the low fluoride area(0.0126-0.0144 mg F/kg/day) in the children aged 13-15 years[28]. Fejerskov *et al*[29] also argued that in severe DF or DDF, damage to enamel and exposure of the dentine can allow an increase in the growth of caries-causing micro-organisms and therefore an increase in DC, as found here. In this study, the subjects were from 8 to 13 years old so that the prevalence of DC included milk teeth and permanent teeth. The prevalence of DC was highest in the 8 years old group in both villages. Considering both permanent teeth and milk teeth together may have affected the research results. Perhaps the best way to determine the exact relationship between fluoride intake and caries would be to calculate the prevalence of DC in permanent teeth and milk teeth separately.

Studies on the F intake in relation to the prevalence of DC are not reported often, especially those that include dose-response relationships[1]. Fluoride ingestion can result in delayed tooth eruption, and the preeruptive incorporation of fluoride into teeth and dental enamel during their formation and development is not believed to be the mechanism by which fluoride helps to reduce tooth decay. Permanent teeth with more fluoride in their enamel from such exposure develop caries as easily as teeth with less fluoride in their enamel. Consequently, ingestion(swallowing) of fluoride is not essential to help prevent tooth decay, at least for permanent teeth[30]. In this study, except for the group of 9-year-old children, there were no significant differences in the prevalence of DC between the other age groups in the two villages, but the overall prevalence of caries between the two villages was significantly different. The eruption time of teeth, nutrition(especially saliva levels of calcium and phosphorus), age, education, dental care, socio-economic status, sample size, and so on, may affect the prevalence of caries. Clearly, the relationship between F and caries needs further investigation. Our study, however, indicates that as DF and DDF increase with excessive F intake, and the DC rates also increase with excessive F intake. Thus, it is important to monitor total fluoride exposure and protect children from excessive F intake, especially during the years of tooth development.

After their eruption, milk teeth can be affected by caries; the prevalence of DC will become highest in these teeth among 6-8 year-old children. After this, milk teeth are shed, and permanent teeth gradually replace them. Therefore, the prevalence of DC and DMFT will gradually decrease. Among 12 year-old children, after the establishment of permanent dentition, the prevalence of DC and DMFT scores generally begins to increase and become stable by age 25[31]. This pattern is basically consistent with the results of the present study. The U-shape relationship between the estimated daily total intake of F and the prevalence of DC was possibly due to the presence of both milk teeth and permanent teeth at age 8-13 in relation to the eruption time of teeth. The exact mechanism, however, requires further study.

The World Health Organization data on dental decay trends in 12-year-old children in 24 countries do not support fluoridation as being a reason for the decline in dental decay that has been occurring in recent decades[32]. The report of Duxbury and colleagues indicated that the use of fluoride varnish should not be recommended as a public health measure for reducing caries in this population[33]. As our study has shown, when the total intake of F was excessive, the prevalence of DC increased. Consequently, the value fluoridation of water, milk, toothpaste, salt, etc. as measures to prevent DC needs further critical assessment.

In the study of Han and his colleagues, when the daily total intake of F was 1.23, 1.51, 3.03, and 4.57 [mg/(p • d), the prevalence of DF was 0, 6.7, 63.7, and 100%, respectively, with the regression equation $y = 27.742x-27.3698(R^2 = 0.95)$[34]. Liang's report indicated that when the daily total intake of F was 2.34, 9.67, and 10.03 mg/p/d, the prevalence of severe dental fluorosis(DDF) was 0.00, 42.00, 72.55%, respectively[35]. In this study, the curve fitting equation was $y = 93.05/(1+149.68e^{-3.64x})$, and $y =-14.44+17.04x$, respectively, between the estimated daily total intake of F and the prevalence of DF and DDF. In these studies the patterns of total F exposure and DF were similar to those reported here.

Acknowledgements

We thank Prof BH Chen (School of Public Health, Fudan University) and Prof MS Zhou(Sihong County Center for Disease Control and Prevention) for their valuable suggestions.

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