Dental Fluorosis and Altitude: a Preliminary Study

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Purpose: Previous studies have reported higher dental fluorosis prevalence in high-altitude communities than in low-altitude communities. This investigation determined and compared dental fluorosis prevalence in populations of children living at high and low altitudes in Mexico.

Materials and Methods: Fluorosis prevalence was determined in 7 to 10-year-old children: 67 in Mexico City (2,240 m) and 71 in Veracruz (sea level). Previous fluoride exposure of those children was surveyed by retrospective, questionnaire data. The fluoride content of water and salt from those communities was also documented.

Results: Fluorosis prevalence in Mexico City (53.0%) was significantly higher than in Veracruz (24.3%) ($p < 0.0001$). While there were statistical differences in one of the fluorosis risk factors between the two communities, the observed difference in fluorosis prevalence was still significant when data were analyzed after adjusting for the reported differences in that factor.

Conclusion: Our results led to the conclusion that the difference in fluorosis prevalence in Mexico City and Veracruz could not be explained by differences in fluoride content of the salt or water samples, self-reported exposure to fluorosis risk factors or estimated fluoride intake.

Key words: fluoride, fluorosis, altitude

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Results of several investigations have established that there is a direct relationship between the amount of fluoride ingested during periods of susceptibility in tooth development and subsequent presentation of dental fluorosis (Evans and Stamm, 1991; Evans and Darvell, 1995). However, there are other factors that have been reported to influence the prevalence of dental fluorosis in geographical areas or population groups where it cannot be explained merely on the basis of the fluoride consumed through the diet. One important factor that has been reported, based mainly on circumstantial evidence, is living at a high altitude.

Several epidemiological reports have indicated that a relationship may exist between higher prevalence and severity of dental fluorosis and residence at high altitude, even when sub-optimal concentrations of fluoride are present in the drinking water (Leatherwood et al, 1965; Angmar-Månsson et al, 1984; Manji et al, 1986; Angmar-Månsson and Whitford, 1990; Yoder, 1997; Yoder et al, 1998). Leatherwood et al (1965) reported that 50% of the

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residents of Thailand who lived at higher elevations presented some form of dental fluorosis regardless of the fluoride concentration in the water, which ranged from negligible to 8 ppm. Results of a similar study in Kenya (Manjii et al, 1986) showed that 100% of the children living at or above 2,400 m above sea level exhibited dental fluorosis in their anterior teeth, even though the water fluoride concentration ranged from only 0.2 to 0.4 ppm. In their Tanzanian study, Yoder et al (1998) reported that 98% of the participants living at high altitude had at least 50% of their teeth affected by dental fluorosis. Water fluoride concentration at the high-altitude site was low (0.18 ± 0.32 ppm) and was not consistent with the observed severity of dental fluorosis.

Some Mexican investigators have also conducted studies to establish possible associations between dental fluorosis and altitude. Mexico is an optimal site for studying the possible effects of altitude on fluorosis development since more than 60% of the Mexican population lives at altitudes higher than 1,000 m above sea level. Furthermore, there are six large urban communities and many rural communities located at least 2,000 m above sea level. More than 30% of the Mexican population (approx. 28 million people) lives at these altitudes. Luengas-Aguirre et al (1991) found a fluorosis prevalence of 97.3% in children living at high altitudes in Mexico. The prevalence and severity of dental fluorosis of the Mexican children examined in that study were higher than the one that had been found in several US communities with comparable (2.8 ppm) or higher concentrations of fluoride in the water. The authors proposed that this strikingly high prevalence and severity of fluorosis could be due to factors that may affect fluoride metabolism, such as altitude, malnutrition, and climate. However, none of these confounding factors were assessed in this study. The same group of investigators (Irigoyen et al, 1995, 1997) followed their 1991 study with a second survey that included communities located at similar altitudes but with low fluoride concentration in their water. They reported 90 to 100% prevalence of fluorosis. Again, no confounding factors, which may have played a role in fluorosis development, were studied. Results of this second study demonstrated that even in one community where water fluoride content was below optimal (0.60 ppm), the investigators encountered a high prevalence and severity of fluorosis. Living at an altitude of 2,000 m was again proposed as the main explanation for the extremely high prevalence of fluorosis (Irigoyen et al, 1995, 1997).

Results of at least one study by Malbelya et al (1992) have also pointed out the importance of considering other factors when studying the effect of altitude on dental fluorosis. Results of the investigation conducted by this group of collaborators showed that the higher prevalence of fluorosis found in the higher altitude community in their study was most likely explained by differences in dietary fluoride intake, more than by altitude.

In light of the reports that have suggested a possible relationship between residence at high altitude and the development of dental fluorosis, and the fact that Mexico provides favorable geography to study these effects, the objective of this pilot investigation was to determine the dental fluorosis prevalence and exposure to fluorosis risk factors in cohorts of Mexican children residing at two different altitudes. Results of this study were used to explore the possible effect of altitude on the development of dental fluorosis in these cohorts of children.

MATERIALS AND METHODS

Panelist Recruitment

Prior to initiation of the study, approval was obtained from the Scientific and the Ethics and Safety Committees of the Escuela de Odontología de la Universidad Intercontinental (UIC). Fluorosis prevalence was determined in 7 to 10-year-old children, living in orphanages. These orphanages also served as foster homes where children lived during the weekdays and then spent weekends with their parents. Children who are admitted into this type of foster care are classified according to socio-economic status, and only those who are considered to be in severe need of these services are admitted. Two study communities were selected: Veracruz, located at sea level, and Mexico City, situated at the high-altitude level of 2,240 m. Children ranging from 7 to 10-years of age were selected because their permanent central maxillary incisors and first molars would have erupted and could be examined for fluorosis. Panelists accepted for the study had to be between 7 and 10-years of age; be a lifetime resident at their study site; have a health history free from conditions that might have affected their diet during their first 4 years of life; have no medical conditions that would contraindicate their receiving a dental exam; and report no consumption of fluoride supplements. The parent or guardian of each child signed an Informed Letter of Consent before the children participated in the study.

Clinical Examinations

Prior to initiation of the study, the researchers who performed the clinical evaluations (AESR and EAMM) were subjected to a process of training, and both inter- and intra-examiner calibrations were performed until reliable kappa values were obtained. In order to blind dental examinations as much as possible, some children who did not meet the criteria for panelists' acceptance were included. Due to the nature of the investigation, cross infection guidelines (SSA/OFEDO, 1994; Centers for Disease Control and prevention, 1993) were strictly followed. The tooth surface index of fluorosis (TSIF) developed by Horowitz et al (1984) was used to score dental fluorosis. In order to differentiate fluorosis from non-fluorosis opacities, a flowchart for differential diagnosis, adapted from the one developed by Cutress and Suckling (1990), was used. A participant was classified as having fluorosis if his or her maximum score was higher than or equal to one.

Previous Fluoride Exposure Questionnaire

Retrospective information regarding the children's previous fluoride exposure was obtained by a self-administered questionnaire answered by the parent or caregiver. This questionnaire was translated into Spanish from a previously used and validated questionnaire (Levy and Zarei, 1991). Questions inquired about previous exposure to known fluorosis risk factors such as toothbrushing frequency, the amount and type of toothpaste used when brushing, use of fluoride rinses, participation in school fluoride programs, use of bottled water, professional topical fluoride applications and/or visits to the dentist. Answers to this questionnaire were used to determine if there had been differences in exposure to fluorosis risk factors between the children living in Mexico City and in Veracruz. Ten percent of the medical personnel and parents at the orphanages, who returned the questionnaire, were asked to complete a duplicate questionnaire at a different time, in order to assess the reliability of the instrument.

Collection of Water and Salt Samples

In both communities, water samples from the panelists' usual water source were collected in 15 ml plastic vials at 3 different times (different days, different times of day) at each orphanage. In addition, because of answers reported on the questionnaires, samples of bottled water were purchased in both communities. Approximately 40 g of salt were also collected from salt samples actually used to prepare meals in the orphanages. Since results of studies have shown that most Mexican children derive their dietary fluoride from water and salt (Martinez-Mier et al, 2003), results of these analyses were used to determine current differences in fluoride levels between communities, as a possible indicators of past differences.

Statistical Analysis

Intra- and inter-examiner agreement was assessed by means of the kappa coefficient. Using a linear weighting scheme, a kappa coefficient and a 95% confidence interval for the kappa coefficient were calculated. Intra- and inter-examiner agreement was assessed on classifying a participant as having fluorosis and on a surface-to-surface basis. Age and gender distribution of the panelists were analyzed in order to determine if there were any statistically significant differences in these parameters between the two study sites. A Student's t-test was used for age, and a chi-square test was used for gender. To determine if there was a difference in fluorosis prevalence of these two populations living at different altitudes in Mexico, comparisons were made between the dental fluorosis scores of the groups from both communities. The prevalence of fluorosis between the communities was compared using a chi-square test. Water and salt fluoride content values were compared by means of Student's t tests. Answers obtained from the questionnaires regarding previous fluoride exposure were dichotomized and compared, excluding responses of "can't remember" and "don't know". Fisher's two-tail exact tests were used to determine associations between fluorosis risk factors and fluorosis prevalence. Cochran-Mantel-Haenszel analysis was used to adjust for the effect of risk factors when comparing differences in fluorosis prevalence between the two study sites.

Table 1 Demographic data for the 7 to 10-year-old cohort

	N Participants and Gender	Community	Mean age in years (± sd)
Orphanage 1	7 males 5 females	Veracruz	8.8 ± 2.3
Orphanage 2	22 males 15 females	Veracruz	8.9 ± 2.1
Orphanage 3	10 males 12 females	Veracruz	9.0 ± 1.9
Orphanage 4	32 males	Mexico City	8.5 ± 2.6
Orphanage 5	35 females	Mexico City	8.7 ± 2.3

RESULTS

One hundred and thirty-eight, 7 to 10-year-old children were recruited for this study. Seventy-one of these children were examined for dental fluorosis in Veracruz and 67 in Mexico City. Demographic data are presented in Table 1. There were no statistically significant differences in the mean age, weight or gender distributions between children recruited at the two sites. There were also no significant differences in socio-economic status due to recruitment practices in the orphanages.

Fluorosis Prevalence

Agreement, as calculated by kappa values, was good to excellent for clinical examinations. During the study, both examiners AESR and EAMM scored twenty participants, while EAMM examined seven participants twice, and three participants were examined twice by AESR. The kappa value for examinations repeated by EAMM was 0.92 (1.0 kappa value for prevalence), the kappa value for AESR's repeated examinations was 0.97 (1.0 kappa value for prevalence), and the kappa value for examinations conducted by both AESR and EAMM was 0.83.

The prevalence of fluorosis within a community was determined by the number of participants with fluorosis divided by the number of participants examined multiplied by 100. Severity was reported on the basis of maximum score assigned to an individual. Results from the fluorosis exams using the TSIF showed that the prevalence in Mexico City (53.0%) was significantly higher than in Veracruz (24.3%) ($p < 0.0001$). These data are presented in Fig 1. Of the 24.3% of the Veracruz children, who presented with fluorosis upon examination, 21.5% had a maximum score of 1 and only 2.8% of the children had a maximum score of 2. Of the 53.0% of children in Mexico City who had fluorosis, a maximum score of 1 was assigned to 30.5% of the children; 7.5% were assigned a maximum score of 2; 1.5% had a maximum score of 3 and 13.5% were assigned the highest score of 5. Differences in severity were also statistically significant ($p < 0.0001$).

Table 2 shows the distribution of TSIF scores on all graded surfaces within each city. The severity of fluorosis observed on most tooth surfaces in the two study sites ranged from 'very mild' to 'mild' (TSIF scores: 1 – 2). In Veracruz, no pitting was observed in any of the examined surfaces and there were no surfaces assigned scores higher than 2. In Mexico City, however, 13 of the examined surfaces showed pits, which are only present in 'moderate' to 'severe' cases (TSIF score: 5).

Exposure to Fluorosis Risks Factors

One hundred and twenty three parents or medical personnel at the orphanages completed and returned the questionnaire regarding previous fluoride exposure: (81.7% in Veracruz and 97.9% in Mexico City). Responses to the duplicate set of questionnaires were similar to responses on the first questionnaires in 93.0% of the cases, indicating a reliability coefficient of 0.87. Responses to the questionnaires were analyzed after eliminating

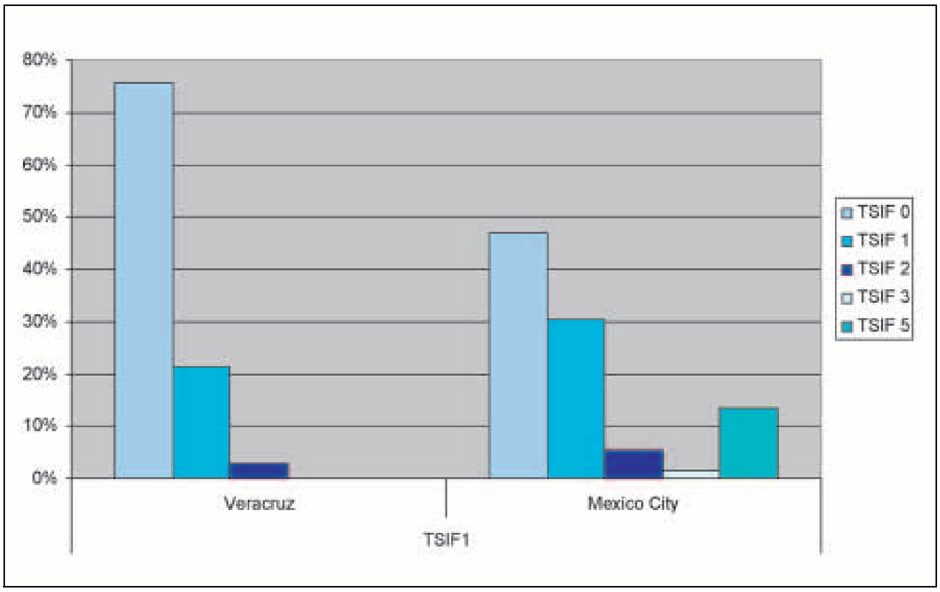

Fig 1 Maximum Tooth Surface Index of Fluorosis (TSIF) score assigned to children in Veracruz and Mexico City expressed as percentages.

Table 2 Distribution of Tooth Surface Index of Fluorosis (TSIF) scores over all surfaces graded

	TSIF 0		TSIF 1		TSIF 2		TSIF 3		TSIF 4		TSIF 5		TSIF 6 and 7		Total
City	n	%	n	%	n	%	n	%	n	%	n	%	n	%	n
Veracruz	1642	90.7	151	8.3	17	0.9	0	0.0	0	0.0	0	0.0	0	0.0	1810
Mexico City	1197	79.8	242	16.1	32	2.1	16	1.1	0	0.0	13	0.9	0	0.0	1500

non-respondents and those reporting “don’t remember” or “don’t know”. Some questions regarding early feeding habits were excluded from the analyses because they had particularly high (> 50%) non-response rates. Answers to the questionnaire are presented in Table 3. One fluorosis risk factor was reported differently between both sites. In Veracruz, 69.6% of respondents reported they began brushing their children’s teeth before age 3. In Mexico City, parents or legal guardians of 44.4% of those children reported they began brushing before age 3. A two-tail Fisher’s exact test found this difference between sites was significant with more children in Veracruz having their teeth brushed before age 3 ($p = 0.026$). Data showed that the observed difference in fluorosis prevalence between the children in Veracruz and Mexico City was still significant when analyzed using Cochran-Mantel-Haenszel tests in order to adjust for the reported exposure to all fluorosis risk factors ($p < 0.05$ in all cases).

When data were analyzed on the basis of presence or absence of fluorosis regardless of altitude of residence, an association between fluorosis presence and the consumption of bottled water, alone or in combination with tap water, was found. Also associated with fluorosis presence was lack of visits to the dentist before age 3. No statistically significant differences in exposure to these risk factors were found between the children in Veracruz and Mexico City. The percentage of children who reported using bottled water in Veracruz (24.1%) and Mexico City (21.3%) was similar. The percentages of parents who reported their children had visited the dentist at early ages were also very similar for both communities. In Veracruz, 42.0% of the children had visited the dentist before age 3. In Mexico City, 33.9% of the respondents reported their chil-

Table 3 Participants' exposure to risk factors for the development of fluorosis

Risk factor	P value for prevalence of fluorosis after adjusting for difference in risk factor (Cochran-Mantel-Haenzel)	Percentage of children who were exposed to risk factor	
		Veracruz % (n/N)	Mexico City % (n/N)
Use of bottled water before age 3[A]	P = 0.001	24.1 (14/58)	21.3 (14/64)
Visits to the dentist before age 3[A]	P = 0.008	42.0 (21/50)	34.0 (16/47)
Professional fluoride applications before age 6	P = 0.0001	42.2 (19/45)	58.7 (27/46)
Toothbrushing before age 3[B]	P = 0.039	69.6 (32/46)	44.4 (16/36)
Toothbrushing twice a day or more before age 3	P = 0.001	56.4 (31/55)	51.2 (22/43)
Use of fluoridated toothpaste before age 3	P = 0.049	75.6 (34/45)	51.9 (14/25)
Use of toothpaste amount larger than 'pea size' before age 3	P = 0.03	91.9 (34/37)	77.3 (17/22)
Participation in fluoride rinse program at school before age 6	P = 0.001	74.5 (41/55)	74.4 (29/39)

[a] Statistically significant differences were found in the exposure to these risk factors between children with and without fluorosis (p = 0.01)
[b] Statistically significant differences were found in the exposure to this risk factor between sites (p = 0.026)

dren went to a dentist before age 3. No statistically significant difference was found between these data from the two study sites (p = 0.531). Toothbrushing habits, brushing frequency and use of toothpaste, were similar for both communities, with no significant differences found.

Water and Salt Fluoride Content

In order to document the concentration of fluoride in water, and to ensure that it was similar in both study sites, a Student's t-test was used to compare the fluoride concentration in tap, bottled, filtered and boiled water samples obtained from the two communities. The average fluoride concentration for all water from Veracruz was 0.04 ± 0.04 ppm; in Mexico City it was 0.07 ± 0.02 ppm (p = 0.0382). Comparisons were made of tap and bottled water from both sites and of boiled water from Veracruz and tap water from Mexico City. Tap water from Veracruz had a mean fluoride content of 0.01 ± 0.04 ppm while the mean fluoride content of tap water from Mexico City was 0.06 ± 0.02 ppm. These data were found to be significantly different (p = 0.0451). The mean fluoride content of bottled water from Veracruz was 0.11 ± 0.21 ppm while bottled water samples from Mexico City contained 0.23 ± 0.05 ppm fluoride. Boiled water from Veracruz contained 0.06 ± 0.01 ppm fluoride but did not differ statistically from the tap water in Mexico City (p = 0.1702). There was no significant difference in fluoride concentration of salt obtained from the two communities. The mean fluoride content of salt was 210.5 ± 150.0 ppm in Veracruz, while it was 227.1 ± 64.8 ppm in Mexico City. No statistically significant difference was observed in water consumption, as determined by the amount (ml) of beverages ingested per day by the children (626.9 ± 158.7 ml in Veracruz and 701.4 ± 212.8 ml in Mexico City).

DISCUSSION

The prevalence and severity of fluorosis was significantly higher in Mexico City than in Veracruz. Our finding of a larger number of cases in Mexico City where pitting was present (TSIF 5) is a good indication of the severity of the condition being higher in Mexico City. There were few more cases diagnosed as TSIF 5 than cases diagnosed as TSIF 3. This finding is in agreement to previous reports from other investigators (Luengas-Aguirre et al, 1991; Horowitz et al, 1984). Since the TSIF is not an ordinal measure it is not uncommon to have a decreasing number of cases from TSIF 1 to TSIF 3, with a larger number of cases diagnosed as TSIF 4 or 5.

The fact that the prevalence and severity of fluorosis was significantly higher in Mexico City than in Veracruz supports the findings of several epidemiological reports that have indicated a relationship may exist between higher prevalence and severity of fluorosis and high altitude (Leatherwood et al, 1965; Manjii et al, 1986; Angmar-Månsson and Whitford, 1990; Yoder, 1997; Yoder et al, 1998). The intra- and inter-examiner calibration in the present study was good to excellent, as shown by kappa values. Based on the overall kappa, the reported differences in fluorosis prevalence most likely reflected a true difference in fluorosis, and not differences in the examiners' perceptions of dental fluorosis.

There have been a limited number of studies conducted in Mexico to which our results can be compared. The fluorosis prevalence results of the present study are much lower than those reported by Luengas-Aguirre et al (1991); although a direct comparison of prevalence is not possible due to differences in the fluoride level in water and salt between communities in the study by Luengas-Aguirre et al (1991) and the current investigation, our findings seem to support their hypothesis proposing altitude as one of the possible explanations for the extremely high fluorosis prevalence they found.

When results from the present study were compared to studies conducted in the state of Hidalgo in 1995 and 1997 (Irigoyen et al, 1995, 1997) the prevalence of fluorosis found in our study was also lower. As in the earlier investigation conducted by this group (Luengas-Aguirre et al, 1991), all communities studied were located at high altitudes. However, in the studies conducted in 1995 and 1997, the researchers included some communities where water fluoride was below optimal (0.55 ppm) in order to determine if higher than expected fluorosis prevalence would also be found in high-altitude communities where water fluoride was not a problem. In those studies fluorosis prevalence was not related to water fluoride content, since higher than expected prevalence of fluorosis was found in high-altitude communities with above- and below-optimal water fluoride. Again, our findings seem to support their hypothesis proposing altitude as one of the possible explanations for the extremely high fluorosis prevalence they found.

The severity of fluorosis reported in the studies conducted in other Mexican communities located at high altitudes is also higher (Luengas-Aguirre et al, 1991; Irigoyen et al, 1995, 1997) than that reported in the present study. In our study, only a few moderate to severe cases were reported, while the researchers from Hidalgo reported severe cases of fluorosis where staining and severe and confluent pitting were present. The 7 to 10-year olds who participated in our study were slightly younger than the 6 to 14-years old cohort in those earlier studies and this may partially explain the fact that no staining or confluent pitting was found in the children examined in this study, even though some pitting was reported in Mexico City. Confluent pitting is believed to be the result of individual pits united by enamel lost due to normal wear so this phenomenon would not have been observed in the newly erupted teeth of the children in the present study.

Even when our results seem to support the conclusions presented by other epidemiological studies conducted in communities located at high altitude in Mexico, drawing conclusions by comparison of those results with the results of the present study must be done with caution. Methodological differences in the conduct of the studies are very probable. Different fluorosis indices were used. Participants in the studies were likely to have been recruited in different ways, resulting in studies of populations with different demographic and socio-economic characteristics. These differences could have had an impact on the panelists' exposure to fluorosis risk factors, and on their nutritional status, which in turn would have affected their reported fluorosis prevalence.

The fact that a socio-economic study is conducted prior to each child's admittance to the orphanages that participated in this investigation and that the results of these studies are used to admit only those children who belong to the lowest socio-economic strata, ensured that socio-economic factors did not differ for the cohorts of children in this study. Results of our retrospective assessment of exposure to fluorosis risk factors support the assumption that there were no differences between the two groups of children, other than residence at different altitudes. Only one known fluorosis risk factor, toothbrushing at an early age, was reported differently between the participants in Mexico City and Veracruz; however, when adjusting for the difference in this risk factor, the reported difference in fluorosis prevalence between sites remained significant. In Veracruz, children began brushing their teeth at a younger age and brushed more often than children in Mexico City, so this would not explain the fact that more fluorosis was present in Mexico City. As a consequence, this difference in reported exposure to early toothbrushing could not account for the difference found in fluorosis prevalence.

A concurrent study conducted by our group (Martínez-Mier, 2003), using the duplicate plate technique (Rojas-Sánchez et al, 1999) obtained direct measurements of total fluoride intake in 15 to 36-month-old children who lived in the same orphanages from which the children who participated in the present study were recruited. The data obtained showed that fluoride intake for two cohorts of younger children from the same communities was similar and enough to cause the developing of dental fluorosis: a mean (± sd) of 0.20 ± 0.08 and 0.18 ± 0.07 mg fluoride/kg/day were ingested by children in Mexico City and Veracruz, respectively. No statistically significant difference was found between the dietary fluoride intake by children in these two cities. Documentation was obtained regarding the dietary guidelines followed at the orphanages, which showed that menus for the 15 to 36-month-old children in the concurrent study (Martínez-Mier, 2003) were prepared using the same dietary guidelines used in previous years for the 7 to 10-year olds. Results of that study, therefore, support the assumption that the dietary fluoride intake of the 7 to 10-year-old children in this pilot study was similar when they were 15 to 36 months old and at risk for developing dental fluorosis. This information is relevant to discard other possible factors that may be responsible for the association of higher prevalence of fluorosis to higher altitude, as suggested by the results of Mabelya et al (1992).

Results of our fluoride analyses of water and salt, which were not different between communities, support the assumption that previous water and salt fluoride levels were similar since there have been no changes in water sources in either of the two communities since the 1990s and there has been no change in the regulations relative to salt fluoride content since their introduction in the 1980s. Therefore, they also support the assumption that the previous fluoride intake in the two cohorts of children was probably similar and should not have played a role in the observed, significant differences in fluorosis prevalence and severity.

The reported prevalence of fluorosis for both communities in this study was higher than would be expected if, as proposed by Nath et al (1992), salt fluoridation is an appropriate vehicle for fluoride distribution and children who consume salt fluoridated at 250 ppm (as distributed in Mexico) are not considered to be at risk for the development of dental fluorosis (Bergmann and Bergmann, 1995). From the retrospective data obtained through the questionnaire, which showed that children younger than six years old were covering at least one fourth of their toothbrush with a thick strip of toothpaste, it was clear that the children for whom data were obtained, were using excessive amounts of fluoridated toothpaste. It should be pointed out that none of these children used child-sized toothbrushes. From answers to the questionnaires it could also be concluded that the children had started using this large amount of toothpaste at an early age when they may have swallowed most of what is placed on the toothbrush, and that may have increased their risk of fluorosis.

The association found between fluorosis presence and the consumption of bottled water alone or in combination with tap water, was in agreement with the findings of a previous Mexican investigation (Cervantes-González et al, 1998). Results of the analyses of bottled water showed that some samples from both communities had above optimal fluoride concentrations (1.83 to 2.07 ppm). This is of particular concern because in some areas of Mexico tap water is not suitable for human consumption and bottled water is the only water of choice. In addition, in recent years in Mexico, as well as in the rest of the world, there has been a shift towards healthier lifestyles that has increased the consumption of bottled water by segments of the population that have access to potable tap water. Salt fluoridation was introduced under the assumption that all water consumed had negligible amounts of fluoride, so if this is not true for those individuals who prefer bottled water, the use of bottled water could be placing them at an increased risk of developing dental fluorosis. Although no significant differences were reported between the communities in the proportion of children who used bottled water, alone or in combination with tap water, differences in the fluoride concentration of the bottled water used by the panelists at a time when they were susceptible to developing dental fluorosis and not analyzed by our study may have played a role in the differences observed in fluorosis prevalence in our study.

In an attempt to explain why those children who had not visited the dentist before the age of three had less fluorosis, reports on the utilization of dental services in Mexico were reviewed. Early dental visits have been reported as a fluorosis risk factor because during those visits children may be exposed to professionally-applied fluoridated products at an age where they are still at risk of developing dental fluorosis. In addition, the frequency of early visits to a dentist is a direct reflection of the parents' awareness of the importance of oral health. This awareness may also increase the exposure of children to fluoridated dental products. Studies on utilization of dental services in Mexico (López Cámara, 1992; López Cámara et al, 1983, 1990) have shown than less that 10% of the time spent in dental offices is dedicated to preventive procedures, and that more than one third of the patients only seek dental care to relieve pain. The percentage of time spent on preventive procedures tends to decrease as the socio-economic status of the patients in a dental practice decreases. It could be hypothesized that due to the particular characteristics of the panelists in this study, differences in parents' awareness of the need for dental care probably did not play a role in the children's exposure to fluoridated products, because parents were not the principal caregivers for children in the study cohorts. And, in light of the low socio-economic level of the panelists, exposure to fluoridated products through preventive dental procedures practiced as a result of dental visits, was not very likely. However, no definite conclusions can be drawn.

When drawing conclusions from our questionnaire results, it must be taken into account that the reliability of retrospective data obtained through the questionnaire in this pilot study was not optimal. In this particular sample of children whose parents or early caretakers were absent, a considerable proportion of some of the questions were left unanswered because of lack of information. For this reason, it was necessary to exclude questions regarding certain risk factors, such as infant feeding habits, which had particularly high non-response rates. Answers to those questions could have further aided in comparing the fluorosis risk factors of the two compared cohorts of children in this study and would have further helped to clarify whether or not those risk factors had a significant effect on the prevalence of dental fluorosis observed in this study.

There are conflicting results regarding the effect of altitude on fluoride metabolism. Although some laboratory reports on rats placed at altitudes higher than 1,800 m have not detected any metabolic changes that could explain the increase of the prevalence in dental fluorosis observed at high altitudes (Elia et al, 1985), there are a number of epidemiological studies where results have shown a definite association between dental fluorosis and altitude. There is also a laboratory report (Zhang et al, 1995), which demonstrated that altitude increases toxic effects of fluoride, such as dental fluorosis. Because our study found a significantly higher prevalence of dental fluorosis in Mexico City than in Veracruz, which could not be explained based upon comparisons of the risk factors for which we were able to obtain adequate data, the results from this pilot study suggest that altitude may have had an effect on the significant difference in fluorosis prevalence and severity between Mexico City and Veracruz. In our study, no data were collected to determine metabolic factors, which may have contributed to the increased prevalence of dental fluorosis observed in Mexico City. Results of the current pilot study indicate the need for future studies in this area. The aim of those studies should be to determine total fluoride intake and to measure metabolic factors in two comparable cohorts of young children residing at different altitudes. Those same cohorts of children should then later be examined for fluorosis presence upon eruption of their permanent teeth.

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