ORIGINAL ARTICLE

Prevalence of Dental Fluorosis and Its Associated Factors After Cessation of Water Fluoridation

Faizah Abdul Karim[1,2], Zamros Yuzadi Mohd Yusof[2], Nor Azlida Mohd Nor[2]

[1] Oral Health Programme, Ministry of Health Malaysia, Level 5, Block E10, Parcel E, Precinct 1, Federal Government, Administrative Centre, 62590 Putrajaya, Malaysia
[2] Department of Community Oral Health and Clinical Prevention, Faculty of Dentistry, University of Malaya, 50603, Kuala Lumpur, Malaysia.

ABSTRACT

Introduction: Water fluoridation (WF) was ceased in the state of Pahang, Malaysia, in July 2012. Any changes in fluoride exposure during childhood can influence the development of fluorosis. Thus, this study aimed to compare the prevalence of fluorosis between WF-ceased and WF-continued areas in Malaysia, and its associated factors among 7 and 12-year-old children. Methods: A cross-sectional study was conducted among children in WF-ceased and WF areas (n=1211). The 12-year-olds in WF-ceased areas were exposed to WF between 4.5 to 6.5 years before cessation, and it was hypothesised they would have similar fluorosis prevalence as their counterparts who lived in WF areas. The 7-year olds were born during the cessation period. Examination of fluorosis was performed by a calibrated dentist using Dean's criteria and a parental questionnaire was utilised to collect information about demographics and exposure to fluoride. Results: Fluorosis prevalence (Dean's≥2) was significantly lower among children in WF-ceased areas than WF areas ($p< 0.001$). Intra-area comparison indicated that the 12-year olds have higher prevalence of fluorosis than the 7-year-olds, except for anterior fluorosis scores among those in WF areas. The majority of fluorosis cases were very mild in both areas. Simple logistic regression indicated that duration of exposure to WF, age, mother's income and education, age started toothbrushing with toothpaste, and frequency of toothbrushing were associated with fluorosis. Conclusions: Fluorosis prevalence decreased following water fluoridation cessation. The null hypothesis that 12-year-olds in both areas would have similar prevalence of fluorosis was rejected. Several factors were associated with fluorosis.

Keywords: Dental fluorosis, Dental public health, Water fluoridation cessation

Corresponding Author:
Nor Azlida Mohd Nor, PhD
Email: azlida@um.edu.my
Tel: +60379674805

INTRODUCTION

Decline in dental caries worldwide is attributed to the widespread use of fluoride (1). In many countries, the main public health policy to ensure wide coverage of fluoride exposure is through water fluoridation. It is defined as a controlled addition of fluoride compound into fluoride deficient public water supplies for the provision of caries prevention (2). The effectiveness of water fluoridation (WF) in caries prevention has been well documented and the only negative effect reported with this public health approach is dental fluorosis (3, 4). However, over time several countries have reviewed their fluoridation policy which led to total cessation in some areas such as Calgary, Canada, La Salud, Cuba, and Wick, Scotland (5, 6). There are also instances where water fluoridation was stopped temporarily due to technical problems such as in Durham, North Carolina, United States (7). The reason for cessation varies, which include political issues, anti-fluoridation movements, technical difficulties, lack of government funding, and increase in fluorosis prevalence (8). A similar scenario of WF cessation was observed in the state of Pahang, Malaysia. This occurred due to privatisation of public water treatment that led to discontinuation of state government funding for WF programmes (9). Water fluoridation in Malaysia was gazzeted as a national public health policy to control caries since 1972, and currently benefits almost 76% of the population (10). The optimal level of fluoride in the Malaysian water supply was set at 0.7 parts per million (ppm), however the level was adjusted downwards to 0.5 ppm in 2005 due to concerns of increased prevalence of fluorosis and exposure to other fluoride sources (11).

Fluorosis is a condition which occurs due to excessive exposure to fluoride during the critical period of tooth development at the pre-eruption stage. In terms of the critical period of tooth development, unerupted permanent teeth are more sensitive to fluorosis development between the first 2 to 3 years of life for

maxillary central incisors and the first 6 to 8 years of life for full dentition (12). As drinking water is one of the main fluoride sources, any changes to fluoride concentration in the water during this critical window will alter total daily fluoride intake. This will be an important factor which can influence fluorosis development.

Despite a good number of epidemiological studies assessing the impact of WF cessation, the reported outcomes focused mostly on dental caries (6). There is limited evidence reported about the consequences of stopping water fluoridation on fluorosis (13). Within the limited evidence in the literature, mixed findings were reported in relation to fluorosis prevalence following a temporary break or total cessation in WF. For instance, Clark and co-workers reported that the prevalence of fluorosis decreased significantly when fluoride was removed from the water supply in British Columbia, Canada (14). In contrast, a study conducted in Jau, Sao Paulo, Brazil analysed the impact of a seven year break in WF and reported that the prevalence of fluorosis increased among children born after WF ceased in comparison to children exposed to WF. The previous authors suggested the reason for this finding could be due to exposure to other sources of fluoride such as dentifrices and toothbrushing habits (15).

The situation in Malaysia offers a unique opportunity to assess the impact of WF cessation on fluorosis. The discontinuation of WF in the state of Pahang allows data comparison with a control community from other states that remained fluoridated (positive control). In an effort to fill the gap in the literature, this study aims to assess the prevalence and severity of dental fluorosis and its associated risk factors among Malaysian children aged 7 and 12 years old born during and after WF cessation. The null hypothesis was that the 12 year old children in WF-ceased areas who were exposed to WF for 4.5 to 6.5 years before cessation would have similar fluorosis prevalence with their counterparts who lived in WF areas.

MATERIALS AND METHODS

Ethical consideration

The protocol for the study was approved by the Medical Ethics Committee, Faculty of Dentistry, University of Malaya [DFCO1902/0002 (P)] and the National Medical Research and Ethics Committee of the Ministry of Health (MOH) Malaysia [NMRR-18-3309-44638]. Permission to conduct this study was obtained from the Malaysian Ministry of Health and Ministry of Education. Informed consent was obtained from parents/guardians of participating children and assented from 7 and 12 year old children.

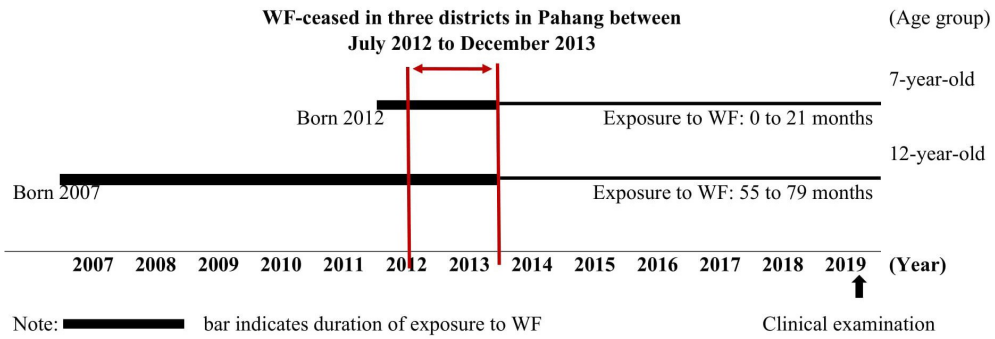
Study design and sample population

This is a cross-sectional study involving lifelong residents aged 7 and 12 years old who lived in WF (Perak) and WF-ceased (Pahang) areas conducted from January to May 2019. The selection of these two age groups is based on the susceptible risk period in developing fluorosis on permanent dentition. The method of comparing children from multiple birth cohorts that were exposed to different fluoride levels due to interruption during tooth development period was well discussed by Singhal et al., (2016) (16).

Children from both age groups living in fluoridated areas in Perak were exposed to WF at 0.5 ppm throughout their life. Children in three WF-ceased areas, namely Temerloh, Jerantut, and Kuala Lipis were partially exposed to WF before cessation took place in July 2012 (6 months before WF ceased). Following the cessation, fluoride content in the water diminished gradually depending on the availability of fluoride compound at each water plantation. Based on the fluoride level data obtained from the Ministry of Health Malaysia (17), the total cessation occurred in December 2013 in some residential districts. The exposure to WF was then calculated based on WF exposure and their date of birth. Of those in the WF-ceased state, the oldest 7 year old children (born 1.1.2012), had partial exposure to WF ranging from 7 to 21 months while the youngest children (born 31.12.2012) had a minimum of no exposure to a maximum of 12 months of WF exposure, according to time of cessation at the area of residence. For the 12 year old group, all children were partially exposed to WF ranging from 55 to 79 months prior to WF cessation (Figure 1).

Sample size calculation

Sample size was calculated based on the prevalence of mild fluorosis in 12 year old children in a fluoridated area



in Malaysia (11.2%) with a corresponding difference of 10.2% with a non-fluoridated area (18). The minimum estimated sample size required for the study to detect a clinically significant 10% difference (19) in fluorosis prevalence within the population with 90% power, a confidence interval of 95% and significance level of 0.05 was 116 participants for each age group per area. The estimated sample size was inflated by 20% to account for non-responding, 10% for non-consenting (13), and multiplied by a design effect of 3.0 (19) to accommodate the variance within the cluster. The final sample size was estimated at 452 subjects per group. In total, 1834 children aged 7 and 12 years old in both WF and WF-ceased areas were recruited for this study.

Selection of schools and selection of children within schools

A simple cluster random sampling using probability proportionate to size technique was adopted to sample the participants from each age group in WF and WF-ceased areas. The primary sampling unit was public primary schools in both states. In the first stage the schools were stratified into respective locations, i.e., urban and rural. The schools were further categorised by type, namely national and vernacular schools. The schools were randomly selected from rural and urban areas using simple random sampling proportionate to the number of school children in the respective location and types of school until the sample size was fulfilled. In total, 25 schools were invited and agreed to participate in this study. All lifelong residents (7 and 12 year olds) in the selected schools were included as study samples. Children who were not life-long residents of the selected areas, had medical contraindications to being dentally examined, non-consented parents and children, and absentees on the day of clinical examination were excluded.

Questionnaire distribution

A parental questionnaire was used to collect information on residential status, sociodemographic characteristics, exposure to fluoride varnish/gel, and previous oral hygiene practices of children before turning six years old (i.e., age started tooth brushing and use of fluoridated toothpaste, tooth brushing frequency, habits after tooth brushing, eating/licking toothpaste, tooth brushing supervision, amount of toothpaste and type of toothpaste). The questionnaire was adopted from a previous local study (18). The questionnaire was face and content validated by experts in the field. Minor amendments were made followed by pre-testing among 60 parents. The same parents were invited to answer the questionnaire for the second time after a one-week interval. Feedback from parents were obtained and minor modifications were made. Internal reliability was assessed using Cronbach's alpha coefficient ($\alpha > 0.70$) indicating substantial reliability. The final version of the questionnaire was distributed to the parents with help from representative school teachers.

Prior to data collection, informed consent from parents/guardians was obtained. Patient information sheet (PIS) together with the consent form and questionnaire were delivered to the parents through their children using a sealed envelope approximately two weeks before the scheduled visit to the schools. The class teacher helped in distributing the envelope to the children. Children were advised to deliver the envelope to their parents for completion and return it to their teachers. The signed consent forms and questionnaires were returned to the researcher through the head of each school. Assent was obtained from the children prior to clinical examination.

Clinical examination

Assessment of dental fluorosis was done after the parental questionnaire had been collected. It was assessed using Dean's Index (0 - normal, 1 - questionable, 2 - very mild, 3 - mild, 4 - moderate, 5 - severe). The teeth were examined for two outcome measures which were anterior fluorosis score (on fully erupted permanent maxillary central incisors) and full mouth score (all fully erupted permanent teeth). If fluorosis was present, scoring was based on the two most severely affected teeth. If the two teeth were not affected to the same degree, scoring was assigned based on the least affected tooth (20). The type of teeth affected with fluorosis was also recorded.

The children were asked to brush their teeth prior to oral examination. The fluorosis examination was conducted using a disposable mirror, a CPITN probe, and gauze for plaque removal (if any). Children were seated in an upright position facing the source of natural light (window or door) during examination. The teeth were examined in wet condition. The children were asked to moisten their teeth or if not possible, the teeth were moistened with a damp cotton wool. The physical appearance (diffuse, horizontal lines, irregular caps on cups), distribution (bilateral tendency) and frequency (involves several teeth) of fluorosis lesions were noted during examination. The CPITN probe was used to check the roughness of enamel surface when necessary. Partially erupted teeth and teeth with orthodontic brackets were excluded from the examination.

Clinical examination was done by a trained and calibrated examiner (FAK). The examiner underwent online training and calibration exercise on fluorosis examination using Dean's Index (21). An inter-examiner Kappa value of 0.7 was obtained. A second training session was done using 39 fluorosis images (22) at University of Malaya, Kuala Lumpur with an inter-examiner Kappa value of 0.97. In the field, 20 children were re-examined after two weeks to assess intra-examiner reliability of which the Kappa value was 0.88. Inter- and intra-examiner reliability indicated substantial to excellent agreement (23).

Data analysis

The prevalence of fluorosis in the present study was defined as very mild or greater (Dean's > 2). Descriptive

analysis was used to describe prevalence and severity of full mouth score fluorosis and anterior fluorosis. These two outcome measures were used in response to changes in fluoride level in the water during tooth development among those in WF-ceased areas (i.e., for the 7 year olds, changes in fluoride level occurred during development of maxillary central incisors; and for 12 year olds, changes occurred during development of central incisors and other permanent teeth). Fluorosis prevalence derived from full mouth scores was further analysed to explore distribution of the most affected teeth with such condition.

Chi-squared test was used to compare fluorosis prevalence among children of the same age group living in WF-ceased and WF areas (i.e., inter- and intra-area comparison) for both fluorosis outcomes (anterior and full mouth scores). Simple logistic regression was conducted to investigate the association between fluorosis and different durations of WF exposure. Association between other risk factors (sociodemographic characteristics, oral hygiene practices, and exposure to fluoride varnish/gel before the children turned six years old) and anterior fluorosis was further analysed using simple logistic regression for all participants. Data were weighted by the average probability of being selected from urban and rural areas in both states. All analyses were carried out using SPSS version 23 with complex sample analysis module (IBM Corp., Armonk, N.Y., USA).

RESULTS

An overall response rate of 83.6% was achieved with 1533 participants returning the questionnaire. Among those, 190 children were not life-long residents, two were non-consenting and four had medical contraindications, and they were excluded from this study. Questionnaires with incomplete responses of more than 20% were also excluded (n=50). Following clinical assessment, 843 and 1211 children were deemed eligible for analysis of anterior fluorosis and full mouth score fluorosis, respectively.

Figure 2 shows fluorosis prevalence among children who lived in WF and WF-ceased areas based on anterior and full mouth scores. Children who lived in WF-ceased areas had significantly lower prevalence of fluorosis based on anterior score [$\chi^2(4, N=843)=107.037$, $p<0.001$] and full mouth score [$\chi^2(4, N=1211)=145.725$ $p<0.001$]. Similar trends were observed for both age groups. When a comparison was made between children living in the same areas, the 12 year olds had higher prevalence of fluorosis than the 7 year olds, except for anterior fluorosis score among those in WF areas. However, the difference in anterior fluorosis score between these two age groups was not statistically significant.

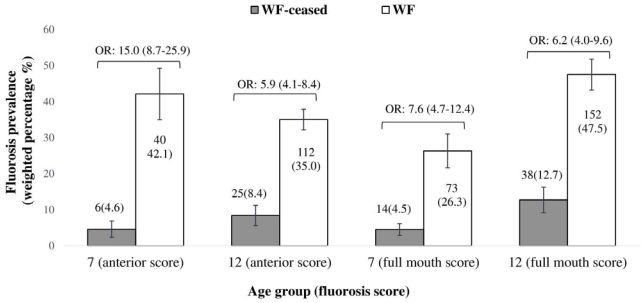


Figure 2: Fluorosis prevalence (Dean's≥2) between children living in WF-ceased and WF areas

Table I shows the distribution of the degree of fluorosis among the participants by age group and area of residence. The most common degree of fluorosis was very mild for both areas (WF-ceased: 3.7%, WF: 24.3%). A small proportion of children in WF areas had moderate (1.9%) to severe (0.2%) fluorosis. However, none of the children in WF-ceased areas has moderate to severe fluorosis. A similar trend of fluorosis distribution was observed when fluorosis was assessed by full mouth score.

Table 1: Distribution of the degree of fluorosis among study participants

Fluorosis Dean's Score	Anterior score		Full mouth score	
	WF-ceased	WF	WF-ceased	WF
	n (%)	n (%)	n (%)	n (%)
0 Normal	394 (92.1)	253 (61.0)	557 (90.9)	363 (60.7)
1 Question-able	3 (0.7)	10 (2.4)	4 (0.7)	10 (1.7)
2 Very mild	16 (3.7)	101 (24.3)	36 (5.9)	135 (22.6)
3 Mild	15 (3.5)	41 (9.9)	16 (2.6)	79 (13.2)
4 Moderate	0 (0)	9 (2.2)	0 (0)	9 (1.5)
5 Severe	0 (0)	1 (0.2)	0 (0)	2 (0.3)
Total examined	428 (100)	415 (100)	613 (100)	598 (100)
Total participants	667	656	667	656
Not recorded[a]	239 (35.8)	241 (36.7)	54 (8.1)	58 (8.8)

Note: n=unweighted sample count, % =weighted percentage, WF: water fluoridation
[a] Not recorded due to teeth that were unerupted or not fully erupted or excluded for other reasons

The teeth that were more commonly affected by fluorosis were the upper right first premolar, upper right central incisors, and lower right first premolars (Table II). The teeth in the upper arch were more commonly affected by fluorosis than mandibular teeth. The teeth least likely to be affected by dental fluorosis were lower incisors and lower left second premolars.

Simple logistic regression analysis between duration of WF exposure and fluorosis prevalence indicated that children with longer exposure to WF had higher odds of exhibiting dental fluorosis (Table III). Regardless of

Table II: Number of teeth affected with dental fluorosis based on full mouth scores

	Upper right quadrant						Upper left quadrant					
Fluorosis score	16 FM	15 SP	14 FP	13 C	12 LI	11 CI	21 CI	22 LI	23 C	24 FP	25 SP	26 FM
Questionable	3	6	9	6	9	4	2	1	1	3	1	1
Very mild	6	8	36	6	12	20	7	7	0	11	4	8
Mild	12	3	0	0	1	2	0	4	0	0	1	0
Moderate	0	2	2	0	0	2	1	0	1	0	0	0
Severe	0	1	0	0	0	2	0	0	0	0	0	0
Total	21	20	47	12	22	30	10	12	2	14	6	9
	Lower right quadrant						Lower left quadrant					
Fluorosis score	46 FM	45 SP	44 FP	43 C	42 LI	31 CI	31 CI	32 LI	33 C	34 FP	35 SP	36 FM
Questionable	2	4	6	1	0	0	0	0	0	1	0	1
Very mild	9	5	13	0	0	0	1	0	0	6	0	10
Mild	0	2	1	0	0	0	0	0	0	2	1	1
Moderate	0	1	0	0	0	0	0	0	0	0	0	0
Severe	0	0	1	0	0	0	0	0	0	0	0	0
Total	11	12	21	1	0	0	1	0	0	9	1	12

Note: The tooth numbering is according to FDI notation. FM: first molar, SP: second premolar, FP: first premolar, C: canine, LI: lateral incisor, CI: central incisor

which fluorosis measure was used, children who were exposed to WF for the first six years of life or longer had significantly higher odds of developing fluorosis than children who were exposed to WF for the first two years or less.

In terms of socio-demographic characteristics, gender, mother's income, and mother's education level were significantly associated with fluorosis prevalence (Table IV). In general, higher fluorosis prevalence was found to be associated with higher parental income (> MYR 4000) and education level (≥ secondary school). However, children whose mothers earned middle income (< MYR 4000) had significantly lower fluorosis scores compared to those children whose mothers earned no income (OR:0.78, 95% CI: 0.65-0.93).

Simple logistic regression between anterior fluorosis prevalence and oral hygiene practices when children were less than six years old and exposure to fluoride varnish/gel are presented in Table V. Only age when children started tooth brushing with toothpaste and frequency of tooth brushing were associated with fluorosis prevalence. Although most of the children started tooth brushing at an early age, the use of toothpaste during tooth brushing was reported after 24 months. Children who brushed with toothpaste after the age of 24 months (OR: 1.75, 95% CI: 1.57-1.96) and brushed twice or more a day (OR: 1.32, 95% CI: 1.13-1.53) were associated with higher prevalence of fluorosis.

Table IV: Simple logistic regression analysis of fluorosis prevalence by sociodemographic characteristics

	Anterior fluorosis (Dean's ≥2)				Unadjusted odds ratio (95% CI)	p-value
	Yes		No			
	n	%	n	%		
Gender						
Boys	83	19.9	334	80.1	Ref	
Girls	100	23.5	326	76.5	1.23 (1.06-1.44)	0.02*
Age (years)						
7	46	20.4	179	79.6	Ref	
12	137	22.2	481	77.8	1.11 (0.92-1.34)	0.20
Ethnicity						
Malay	143	20.6	550	79.4	Ref	
Chinese	28	29.2	68	70.8	1.58 (0.69-3.64)	0.20
Indian/others	12	22.2	42	77.8	1.10 (0.81-1.50)	0.45
Location						
Rural	134	20.7	513	79.3	Ref	
Urban	49	25.0	147	75.0	1.28 (0.46-3.52)	0.54
Father's monthly income						
No income	6	24.0	19	76.0	Ref	
<MYR 4000	97	22.4	336	77.6	0.91 (0.52-1.60)	0.68
>MYR 4000	30	24.4	93	75.6	1.02 (0.36-2.88)	0.96
Mother's monthly income						
No income	93	23.4	305	76.6	Ref	
<MYR 4000	56	19.4	233	80.6	0.78 (0.65-0.93)	0.02*
>MYR 4000	21	24.1	66	75.9	1.03 (0.51-2.07)	0.92
Father's highest education						
≤ Primary school	13	18.6	57	81.4	Ref	0.32
Secondary school	117	23.6	379	76.4	1.35 (0.64-2.84)	0.36
College/University	46	22.5	158	77.5	1.28 (0.66-2.46)	
Mother's highest education						
≤ Primary school	6	13.3	39	86.7	Ref	
Secondary school	110	22.8	372	77.2	1.95 (1.08-3.50)	0.03*
College/University	56	22.3	195	77.7	1.87 (0.67-5.21)	0.17

Note: n=unweighted sample count, % =weighted percentage, Ref: reference category.
*Statistically significant

DISCUSSION

WF has ceased in the state of Pahang, Malaysia for almost seven years. As expected, fluorosis prevalence is

Table III: Simple logistic regression analysis of fluorosis prevalence by duration of fluoride exposure from water

Exposure to fluoride in the water supply	Fluorosis prevalence (anterior score) (Dean's ≥2)				Unadjusted Odds ratio (95% CI)	p-value	Fluorosis prevalence (full mouth score) (Dean's ≥2)				Unadjusted Odds ratio (95% CI)	p-value
	Yes		No				Yes		No			
	n	%	n	%			n	%	n	%		
1st 2 years of life	6	4.7	122	95.3	Ref		13	4.2	297	95.8	Ref	
1st 6 years of life	22	7.7	262	92.3	1.71 (1.02-2.87)	0.05*	36	12.2	259	87.8	2.94 (2.03-4.26)	0.001*
1st 8 years of life	43	39.4	66	60.6	13.25 (6.78-25.90)	<0.001*	75	26.5	208	73.5	7.62 (4.70-12.38)	<0.001*
Life-long	111	34.5	211	65.5	10.84 (6.19-19.00)	<0.001*	142	44.0	181	56.0	18.79 (11.40-31.00)	<0.001*

Note: n=unweighted sample count, % =weighted percentage.
*Statistically significant, Ref: reference category

Table V: Simple logistic regression analysis of fluorosis prevalence by oral hygiene practices and fluoride varnish/gel exposure prior to turning six years old

	Anterior fluorosis (Dean's ≥2)				Unadjusted odds ratio (95% CI)	p-value
	Yes		No			
	n	%	n	%		
Age started tooth brushing						
Before 24 months	108	21.3	400	78.7	Ref	
After 24 months	72	23.5	234	76.5	1.14 (0.98-1.32)	0.07
Age started tooth brushing with toothpaste						
Before 24 months	40	15.7	214	84.3	Ref	
After 24 months	140	24.7	427	75.3	1.75 (1.57-1.96)	<0.001*
Frequency of tooth brushing						
≤ Once/day	67	19.1	283	80.9	Ref	
≥ Twice/day	112	23.8	359	76.2	1.32 (1.13-1.53)	0.01*
Supervised tooth brushing						
Yes	166	21.8	597	78.2	Ref	
Never	3	14.3	18	85.7	0.60 (0.20-1.76)	0.26
Type of toothpaste used						
Non-fluoridated	18	19.8	73	80.2	Ref	
Fluoridated	158	22.1	556	77.9	1.15 (0.84-1.57)	0.27
Habits after tooth brushing						
Spat	171	21.4	613	78.2	Ref	
Swallow	1	6.7	14	93.3	0.26 (0.03-2.55)	0.18
Eating/licking toothpaste						
Never	104	23.9	332	76.1	Ref	
Yes	74	19.7	301	80.3	0.79 (0.59-1.05)	0.08
Amount of toothpaste used						
Smear/pea sized	75	22.6	257	77.4	Ref	
Medium to large	104	21.5	379	78.5	0.94 (0.72-1.22)	0.548
Exposure to fluoride varnish/gel						
No	115	23.6	373	76.4	Ref	
Yes	32	24.1	101	75.9	1.03 (0.69-1.54)	0.861

Note: n=unweighted sample count, % =weighted percentage, Ref: reference category
*Statistically significant

lower among children in WF-ceased areas than those in WF areas. Results from the present research confirmed existing findings by studies conducted in Canada (14) and Guangzhou, China (24) that assessed the impact of WF cessation on fluorosis. There is a consensus in the literature that fluorosis has a linear association with fluoride level and duration of exposure (3, 4). Similar findings were observed in this study, where the longer children were exposed to fluoride in the water, the higher the odds of developing fluorosis. In terms of severity, the majority of the cases were very mild in both areas and are unlikely to have a negative effect on aesthetic appearance.

Higher fluorosis prevalence was observed with full-mouth scoring compared to partial scoring in both areas. This finding suggests that partial scoring of fluorosis limited to central incisors underestimates the true prevalence of fluorosis in comparison to full-mouth scoring. Having said that, it is known that the choice of partial or full mouth scoring is often based on the primary objective of the study. For instance, the use of anterior teeth fluorosis scoring is commonly reported to measure fluorosis prevalence when changes occur in fluoride exposure either in water (7, 25) or dentifrice (26) during the critical period of central incisors development. A similar justification was used when reporting anterior fluorosis scoring for the present study as well as it allows data comparison with other studies. Apart from that, partial scoring of fluorosis limited to central incisors (aesthetically important teeth) is often reported in studies that use photographic assessment for blinding of examiners (25, 27). As such, inclusion of posterior teeth in the photographic method poses additional complexities and is technique sensitive.

As far as this research is concerned, it was hypothesised that the 12 year old children in WF-ceased areas would show similar fluorosis prevalence as their counterparts in WF areas. This is because they had 4.5 to 6.5 years of exposure to WF during the critical window of tooth development before cessation. During this critical period, most teeth like central incisors, molars, and premolars are susceptibility to fluorosis if excessive fluoride ingestion occurs. However, this hypothesis could not be supported. Regardless of which outcome measure was used, fluorosis prevalence among 12 year old children in WF-ceased areas was significantly lower than their counterparts with lifetime exposure to WF. A potential explanation for this unexpected finding could be due to non-compliance of fluoride levels in the water supply in some residential districts in Pahang prior to cessation. In terms of exposure to other fluoride sources, most children in both areas reported using fluoridated toothpaste after 24 months. However, there is a possibility of under reporting of post-brushing habits such as licking or accidentally swallowing toothpaste during childhood.

When a comparison was made between children living in the same areas, the 12 year olds had higher fluorosis prevalence than the 7 year olds, except for the anterior fluorosis score among those in WF areas. It was expected that those who had longer exposure to WF were more likely to develop fluorosis. This indicated that fluorosis decreased following WF cessation. However, the contradicting finding of lower anterior fluorosis scores among the 12 year olds in WF areas could be caused by the high number of 7 year old children (n=241) excluded from anterior fluorosis analysis due to unerupted or partially erupted maxillary central incisors. This may have resulted in an overestimation of fluorosis prevalence among them. Hence, this finding must be interpreted with caution and not to be inferred as an increase in fluorosis prevalence among the younger age group. In addition, there is a possibility that those in WF area used a water filtration system at home which may reduce the fluoride concentration in the water supply

(28). Evidence suggests that the use of certain types of water filters such as reverse osmosis reduces the fluoride concentration in the water (28), which may reduce the total fluoride intake among the study population. This could potentially explain the lower scores in full mouth fluorosis in WF area observed in the present study than the data from the national survey (77%) (19).

In terms of other risk factors, a frequency of tooth brushing more than 2 times a day before the age of six was significantly associated with higher fluorosis prevalence. This finding is supported by other studies conducted in Brazil (29), Australia (27), Mexico (30), and Belgium (31). Results of the present study indicated that children who started tooth brushing with toothpaste after 24 months of age were associated with higher fluorosis prevalence in comparison to those who started brushing earlier. Although this result is counterintuitive, it may suggest the possibility of a combined effect of water fluoridation and fluoridated toothpaste that contribute to the development of anterior fluorosis. The complexity of assessing the impact of water fluoridation in the presence of other confounders such as exposure to fluoride from other sources has been acknowledged in the literature (4).

Higher fluorosis prevalence was found to be associated with higher parental income (> MYR 4000) and education level. Possibly parents with higher education had more knowledge on good oral hygiene practices and had access to fluoride toothpaste and fluoride varnish compared to those of lower education level (32). However in terms of income, children with mothers who earned middle income (< MYR 4000) were significantly less likely to have fluorosis compared to those whose mothers earned no income. Similar findings were reported by studies in Mexico (33), Pakistan (34), and Brazil (30) that reported an inverse relationship between socioeconomic status and fluorosis. Results across studies have been mixed and do not fully establish the relationship between fluorosis and socio-economic status. With regards to gender, girls were more likely to have fluorosis than boys. This could be due to a higher tendency for girls to frequently brush their teeth compared to boys due to aesthetic concerns (35).

Data from the present study also indicates that fluorosis was common in premolars in both upper and lower arches. Similarly, Bhagavatula et al., reported that the most prevalent teeth affected with fluorosis were first premolars (36). Another study in Mexico reported that fluorosis more likely affects posterior than anterior teeth (37). In relation to fluoride exposure, longitudinal data indicates that high intake of fluoride in the first three years of life strongly determines the likelihood of having fluorosis in maxillary central incisors and first molars (38), while continuous elevated intake of fluoride from the age of two to seven significantly increases fluorosis occurrence in canines and premolars (36). Thus, future research should consider the number and type of teeth examined when partial fluorosis scoring is used to minimise possible data loss.

The present study is cross-sectional in nature and has common limitations such as parents' self-reported behaviour for their child and recall bias. However, this method of data collection is commonly reported in the literature because of its practicality, time and cost saving. In addition, data comparison of fluorosis prevalence between other published studies is often difficult because of differences in indices and clinical examination methods. When discussing literature on fluorosis data, the comparisons across studies were made with this limitation in mind.

One may argue the potential misclassification of fluorosis with other types of enamel defect or discoloration such as molar incisor hypomineralisation (MIH) and amelogenesis imperfecta. If misclassification had occurred, it would have resulted in more fluorosis cases in WF-ceased areas as well which was not seen in the results. Efforts had been made to reduce the risk of classification bias by examining the patterns of bilateral distribution and tactile examination when assessing fluorosis.

It is acknowledged that the examiner was not blinded to the fluoridation status of the children when fluorosis examination was carried out on site. Although a photography technique by means of taking the children's photos and assessing the fluorosis away from the study areas can be done to reduce bias, this method was not employed in this study due to cost and time limitations. In addition, this technique has technical complexities in capturing clear images of posterior teeth for full mouth fluorosis score.

CONCLUSION

Fluorosis prevalence decreased following water fluoridation cessation. The null hypothesis that 12 year old children in WF-ceased areas who were exposed to WF for 4.5 to 6.5 years before cessation would have similar fluorosis prevalence with their counterparts who lived in WF areas was rejected. Children who lived in WF-ceased areas and received partial exposure to fluoride in the water had significantly lower prevalence of fluorosis than those in WF areas who received lifetime exposure. The majority of fluorosis cases were very mild and mild. Duration of exposure to WF, age, mother's income and education, age started tooth brushing with toothpaste and frequency of tooth brushing were associated with fluorosis in simple logistic regression analysis.

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REFERENCES

1. Whelton HP, Spencer AJ, Do LG, Rugg-Gunn AJ. Fluoride Revolution and Dental Caries: Evolution of Policies for Global Use. J Dent Res. 2019;98(8):837-46.
2. O'Mullane DM, Baez RJ, Jones S, Lennon MA, Petersen PE, Rugg-Gunn AJ, et al. Fluoride and Oral Health. Community Dent Health. 2016;33(2):69-99.
3. McDonagh MS, Whiting PF, Wilson PM, Sutton AJ, Chestnutt I, Cooper J, et al. Systematic review of water fluoridation. BMJ. 2000;321(7265):855-9.
4. Iheozor-Ejiofor Z, Worthington HV, Walsh T, O'Malley L, Clarkson JE, Macey R, et al. Water fluoridation for the prevention of dental caries. Cochrane Database Syst Rev. 2015(6):Cd010856.
5. McLaren L, Patterson S, Thawer S, Faris P, McNeil D, Potestio M, et al. Measuring the short-term impact of fluoridation cessation on dental caries in Grade 2 children using tooth surface indices. Community Dent Oral Epidemiol. 2016;44(3):274-82.
6. McLaren L, Singhal S. Does cessation of community water fluoridation lead to an increase in tooth decay? A systematic review of published studies. J Epidemiol Community Health. 2016;70(9):934-40.
7. Burt BA, Keels MA, Heller KE. The effects of a break in water fluoridation on the development of dental caries and fluorosis. J Dent Res. 2000;79(2):761-9.
8. McLaren L, Singhal S. Does cessation of community water fluoridation lead to an increase in tooth decay? A systematic review of published studies. J Epidemiol Community Health. 2016:jech-2015-206502.
9. Ministry of Health Malaysia. Annual Report 2016: Oral Health Programme, Ministry of Health Malaysia. Putrajaya: Oral Health Programme, Ministry of Health, Malaysia; 2018:p. 28.
10. Ministry of Health Malaysia. Annual Report 2017: Oral Health Programme, Ministry of Health Malaysia. Putrajaya: Oral Health Programme, Ministry of Health, Malaysia; 2019:p. 38.
11. Ministry of Health Malaysia. Implementation of Water Fluoridation Programme in Malaysia: Oral Health Division, Ministry of Health Malaysia Putrajaya: Oral Health Division, Ministry of Health, Malaysia; 2006:p. 10-6.
12. Buzalaf MAR, Levy SM. Fluoride intake of children: considerations for dental caries and dental fluorosis. Monographs in Oral Science. 2011;22:1-19.
13. Mohd Nor NA. The Impact of Downward Adjustment of Fluoride Concentration in The Malaysia Public Water Supply On Dental Caries and Fluorosis [PhD thesis]. Cardiff, UK: Cardiff University; 2017.
14. Clark DC, Shulman JD, Maupome G, Levy SM. Changes in dental fluorosis following the cessation of water fluoridation. Community Dent Oral Epidemiol. 2006;34(3):197-204.
15. Buzalaf MA, de Almeida BS, Olympio KP, da SCVE, de CSPSH. Enamel fluorosis prevalence after a 7-year interruption in water fluoridation in Jau, Sao Paulo, Brazil. J Public Health Dent. 2004;64(4):205-8.
16. Singhal S, Farmer J, McLaren L. Methodological considerations for designing a community water fluoridation cessation study. Community Dent Oral Epidemiol. 2017;45(3):193-200.
17. Ministry of Health Malaysia. Health management information system: monthly district and state report of fluoride feeder status PKP 204 for year 2008 to 2019. Putrajaya, Malaysia: Health Informatics Centre; 2020.
18. Mohd Nor NA, Chadwick BL, Farnell DJ, Chestnutt IG. The impact of a reduction in fluoride concentration in the Malaysian water supply on the prevalence of fluorosis and dental caries. Community Dent Oral Epidemiol. 2018;46(5):492-9.
19. Abd Muttalib K, Abdullah N, Tan E, Che Salleh N, Ismail N, Jaafa N, et al. Fluoride enamel opacities in 16-year-old schoolchildren in Malaysia 2013: Oral Health Division, Ministry of Health. Putrajaya: Oral Health Division, Ministry of Health, Malaysia; 2018:p. 13.
20. Dean HT, McKay FS. Production of Mottled Enamel Halted by a Change in Common Water Supply. Am J Public Health Nations Health. 1939;29(6):590-6.
21. Whelton H, Browne D, Felicia P, Whelton J. E-training for Dean's Index Version 2 Ireland: Oral Health Services Research Centre, University College Cork, Ireland. Website: http://www.fluorosisindex.com/. Accessed January 10, 2020.
22. Mohd Nor NA, Chestnuttl IG, Chadwick BL. Examiner reliability in fluorosis scoring: a comparison of photographic and clinical methods. Community Dent Health. 2016;33(2):145-50.
23. Landis JR, Koch GG. The measurement of observer agreement for categorical data. Biometrics. 1977;33(1):159-74.
24. Wei Z-d, Wei Y. Fluoridation in China: a clouded future. Fluoride. 2002;35(1):1-4.
25. Wong HM, McGrath C, King NM. Diffuse opacities in 12-year-old Hong Kong children–four cross-sectional surveys. Community Dent Oral Epidemiol. 2014;42(1):61-9.
26. Do LG, Spencer AJ. Decline in the prevalence of dental fluorosis among South Australian children. Community Dent Oral Epidemiol. 2007;35(4):282-91.
27. Pretty IA, Boothman N, Morris J, MacKay L, Liu Z, McGrady M, et al. Prevalence and severity of dental fluorosis in four English cities. Community Dent Health. 2016;33(4):292-6.
28. Mohd Kamil NAF, Bahari M, Akhbar NA, Mizad M. Evaluation of fluoride concentration in water filter system for households. Int J Integr Eng. 2018;10(2).

Retrieved from https://publisher.uthm.edu.my/ojs/index.php/ijie/article/view/2639.
29. Azevedo MS, Goettems ML, Torriani DD, Demarco FF. Factors associated with dental fluorosis in school children in southern Brazil: a cross-sectional study. Braz Oral Res. 2014;28.
30. Perez-Perez N, Irigoyen-Camacho ME, Boges-Yanez AS. Factors affecting dental fluorosis in low socioeconomic status children in Mexico. Community Dent Health. 2017;34(2):66-71.
31. Bottenberg P, Declerck D, Ghidey W, Bogaerts K, Vanobbergen J, Martens L. Prevalence and determinants of enamel fluorosis in Flemish schoolchildren. Caries Res. 2004;38(1):20-8.
32. Gurunathan D, Moses J, Arunachalam SK. Knowledge, Attitude, and Practice of Mothers regarding Oral Hygiene of Primary School children in Chennai, Tamil Nadu, India. Int J Clin Pediatr Dent. 2018;11(4):338-43.
33. Pontigo-Loyola AP, Medina-Solis CE, Lara-Carrillo E, Patino-Marin N, Escoffie-Ramirez M, Mendoza-Rodriguez M, et al. Impact of socio-demographic, socioeconomic, and water variables on dental fluorosis in adolescents growing up during the implementation of a fluoridated domestic salt program. Odontology. 2014;102(1):105-15.
34. Sami E, Vichayanrat T, Satitvipawee P. Dental fluorosis and its relation to socioeconomic status, parents' knowledge and awareness among 12-year-old school children in Quetta, Pakistan. Southeast Asian J Trop Med Public Health. 2015;46(2):360-8.
35. Mamai-Homata E, Koletsi-Kounari H, Margaritis V. Gender differences in oral health status and behavior of Greek dental students: A meta-analysis of 1981, 2000, and 2010 data. J Int Soc Prev Community Dent. 2016;6(1):60-8.
36. Bhagavatula P, Levy SM, Broffitt B, Weber-Gasparoni K, Warren JJ. Timing of fluoride intake and dental fluorosis on late-erupting permanent teeth. Community Dent Oral Epidemiol. 2016;44(1):32-45.
37. Medina-Solis CE, Pontigo-Loyola AP, Maupome G, Lamadrid-Figueroa H, Loyola-Rodrнguez JP, Hernбndez-Romano J, et al. Dental fluorosis prevalence and severity using Dean's index based on six teeth and on 28 teeth. Clin Oral Investig. 2008;12(3):197-202.
38. Hong L, Levy SM, Warren JJ, Broffitt B, Cavanaugh J. Fluoride intake levels in relation to fluorosis development in permanent maxillary central incisors and first molars. Caries Res. 2006;40(6):494-500.
39. Lygidakis NA, Wong F, Jљlevik B, Vierrou AM, Alaluusua S, Espelid I. Best Clinical Practice Guidance for clinicians dealing with children presenting with Molar-Incisor-Hypomineralisation (MIH): An EAPD Policy Document. Eur Arch Paediatr Dent. 2010;11(2):75-81.