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Contents

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Former President's message

Dear Members

Shin, Seung-Chul
The former President of AAPD

Happy New Year!

I would like send a deep appreciation for your supporting on 8th conference of AAPD, Jeju, in 2008.

I ll put some photographs of the AAPD on the homepage: www.aapdasia.com , and please enjoy yourself with them.

I have a plan to open English journal as International Journal of Clinical Preventive Dentistry which has been published 3times per year, periodically, since 2005, to everyone in Asia who want to publish ones academic works, except the AAPD journal as IJOH which was published one or two years interval.

IJCPD has been managed by mainly Korean members (Korean Academy of Clinical Preventive Dentistry, Chief Editor: Prof. Cho, Ja-Won. Dankook Univ.)

and sponsored by Korean authors and companies, for several years, and we are trying the journal for SCI level.

So, we need many articles, papers and clinical case reports, moreover we need many professors for appraisal and judgement of the papers, from several countries in Asia, in order to be in international.

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Thank you.

Jan. 1. 2009

Original Article

Intraoral Residual Fluoride Following Tooth Brushing with a Weak Fluoride Solution

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Abstract

We measured intraoral residual fluoride following tooth brushing with a weak fluoride solution (100 ppm F, as NaF) to confirm and secure the safety of this fluoride modality for infants.
Recovered amounts of fluoride after tooth brushing in an ordinary method by the guardians of the experimental subjects were calculated. The mean residual fluoride amount was 63.8 g F (21.3%; correspond to the fluoride amount prepared in the solution), and equivalent to 4.9 g F/kg b.w.. There was significant difference in intraoral fluoride by groups (the eruption completed group > the eruption developing group) and guardians (guardian b,c > a). It was shown that tooth brushing with a weak fluoride solution was a highly safe home care modality of fluoride application for infants. However, it was also shown that the production of a manual to standardize the amount of weak fluoride solution that was applied to tooth brush by guardians was necessary.

Keywords: Fluoride residue, Tooth brushing, Weak fluoride solution, Caries prevention, Deciduous teeth

Int J Oral Health 2009;5:3-6

Introduction

In Japan, there are no appropriate modalities of fluoride application that can be used in 1-to 3- year-old infants in whom deciduous teeth erupt. Reasons for the lack are as follows.

Systemic fluoride application, the typical one of which is communal water fluoridation, has not been implemented. Professionally applied topical fluoride has limited effectiveness, because it is difficult to secure moisture exclusion and /or paint in younger children. Fluoride moth-rinsing cannot be used, as young children are unable to rinse moth well and may swallow. Paste type fluoride dentifrices are difficult to apply, as tooth brushing is done by a guardian, while young children, who are reclining, may swallow.

Accordingly, tooth brushing with a weak fluoride solution (hereinafter referred to as TWFS) is promoted as a regional health care in Kanagawa Prefecture (Public Health and Welfare Dept. 1998), by referring to previous reports (Arakawa, 1985, Arakawa *et al.* 1997) and good results in Sendai City (Taura *et al.* 1995), where TWFS is implemented as a public health measure for prevention of deciduous teeth caries. However, no study results of intraoral residual fluoride have been reported following the use of weak fluoride solution (WFS). Currently, TWFS is implemented on the assumption that safety is secured, even if the entire amount of a one-time does of fluoride contained in WFS (100ppm F, 3mL) is swallowed. It is necessary to offer safer and more reliable information. Thus, intraoral residual fluoride was measured after TWFS. If the present study can prove that TWFS is a safer modality of fluoride application for young children, the spread of TWFS and reduction of caries of deciduous teeth can be expected.

Materials and Methods

The present study was reviewed and approved by the ethical committee of Kanagawa Dental College (No. 53. June 2007)

Subjects

Subjects were eight infants with deciduous dentition who received daily tooth brushing by guardians while lying down and the head being placed on the knee of guardians who sit down on the

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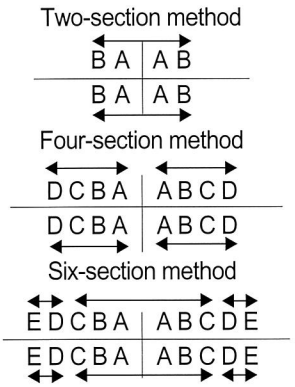
Table 1. *Demographic characteristics and grouping of study subjects*

Group	Subject Code	Gender	Age	Guardian code
Eruption completed	A	F	4	a
	B	F	4	b
	C	M	3	c
Eruption developing	D	M	2	a
	E	F	2	d
	F	M	2	b
	G	M	1	e
	H	M	1	f

Subjects in whom all deciduous teeth were erupted belonged to the eruption completed group. Subjects in whom the second deciduous molar had not erupted belonged to the eruption developing group.



A: deciduous central incisor, B: deciduous lateral incisor,
C: deciduous canine, D: deciduous first molar, E: deciduous second molar.

According to the state of eruption of deciduous teeth, sections shown in the figure were set up. One section denoted 10 back-and-forth scrubbing by a tooth brush with WFS. In the present study, the eruption completed group adopted a six section method and the eruption developing group a four section method.

Figure 1. *A usual method of tooth brushing with a weak fluoride solution*

floor (hereinafter referred to as lying down brushing). None of the subjects had caries or other oral diseases or abnormalities. Table 1 shows the grouping of subjects by their basic attributes and the sate of eruption of deciduous teeth. As there were two couples consisting of two siblings each (subject codes A and D, and B and F), there were six guardians for eight subjects. For the purpose of analysis of results, subjects were divided into two groups based on the stated of eruption of deciduous teeth; one is eruption completed and the other eruption developing.

Experimental procedure

WFS used in this study was a sodium fluoride solution at 100 ppm fluoride concentration prepared by dissolving sodium fluoride powder (special grade, Wako Pure Chemical Industries Co., Ltd.) in deionized distilled water (DDW).

One of authors (YA) visited each subject s home bringing sample bottles containing 3 mL WFS, tooth brushes appropriate for age (the eruption completed group: Dent EX kodomo 13M, the eruption developing group: Dent EX kodomo 14M, Lion Dental Products Co., Ltd.), and a container of DDW for recovery of samples. Guardians performed TWFS in each subject in an ordinary method (Figure 1). At the completion of TWFS, sample bottles with remaining WFS and tooth brushes with WFS and saliva were put into the above-mentioned container. YA carried back samples to the laboratory and measured fluoride concentrations with the fluoride ion specific electrode (Model 96-09, Orion Research, Inc). The above-described operation was repeated three times on separate day.

Recovered amounts of fluoride were calculated using measured fluoride concentrations. By subtracting the recovered amount of fluoride from the amount of fluoride in prepared WFS, the amount of intraoral residual fluoride was obtained (Arakawa, 1994).

Table 2. *Rates and amount of intraoral residual fluoride after tooth brushing using a weak fluoride solution*

Subject (guardian)code	Rate of intraoral residual fluoride (%)	Amount of intraoral residual fluoride (g)	Amount of intraoral residual fluoride per body weight (g/kg b.w.)
A(a)	17.7	53.1	3.5
B(b)	32.0	96.1	6.9
C(c)	26.8	80.4	5.4
D(a)	10.2	30.5	2.5
E(d)	20.9	62.6	5.2
F(b)	23.3	69.9	5.0
G(e)	23.9	71.7	6.0
H(f)	15.3	46.0	4.6
Total	21.3	63.8	4.9

Values shown are the mean of three measurements in each subject

Statistical analysis

As experimental data compiled with normal distribution, parametric test was used. The difference of means between two groups was compared suing the Student s t test. For comparisons of the difference of means among three guardians, analysis of variance (ANOVA) and multiple comparisons (Tukey s HSD test) were used.

Results

Table 2 shows the results of measurement. There was no difference by gender of subject. Whereas the mean residual rate of intraoral fluoride was 21.3%, results of three measurements in each subjects ranged widely between 7.1% and 34.0%. The mean intraoral residual fluoride was 63.8 g F (range: 21.3 ~ 102.0 g F). The mean intraoral residual fluoride was converted into 4.9 g F (range: 1.8 g F ~ 7.5 g F) per 1 kg body weight of subjects.

Figure 2 shows intraoral residual fluoride by groups and by guardians in the eruption completed group. The intraoral residual fluoride in the eruption completed group was significantly higher than that in the eruption developing group ($p=0.0402$). One way ANOVA with a factor of guardians (a, b and c) in the eruption completed group revealed significance ($p=0.0048$). In multiple comparisons, guardian a was significantly lower than guardian b and c. There was no significant difference in intraoral residual fluoride by guardians in the eruption developing group.

Discussion

Safety

The dose of fluoride that manifests acute toxicity (digestive organ symptom) is referred to be 3,000 g F/kg b.w. or more (Japan Poison Information Center. 2008). The mean intraoral residual fluoride after TWFS in this study was 4.9 g F/kg b.w. or equivalent to about 1/600 of the above dose, demonstrating that TWFS was a highly safe modality of fluoride application.

The mean body weight of Japanese 1-year-ole children, the youngest subject of this study, is about 10kg (Health and Welfare Statistics Association. 2007), and the fluoride dose manifesting acute toxicity (digestive organ symptom) for these children is 30,000 g F. Accordingly, even if the whole of one time dose of fluoride, WFS 3mL (300 g F) used in TWFS is swallowed accidently, acute toxicity will not occur.

Furthermore, as Dunipace *et al.* (1998) proposed that the maximal permissible intake of fluoride that will not cause dental fluoosis in 1-to 3-year-old children is 1,300 g F/day, the occurrence of chronic toxicity is hardly possible.

Manipulation of TWFS

There was a significant difference in intraoral residual fluoride between two groups divided according to the state of eruption of deciduous teeth. The difference was caused by the difference of the amount of fluoride applied to tooth brush, that is, lying-down-brushing of the eruption completed group was implemented in six sections, whereas that of the eruption developing group was implemented in four sections. There also was a significance by a factor of guardians even when the same tooth brush was used, suggesting that the amount of WFS used in one brushing differed by guardians.

Future prospect

The present study demonstrated that TWFS was a highly safe home care modality of fluoride application in 1 - to 3-year-old infants. However, it is necessary to produce a manual to standardize the

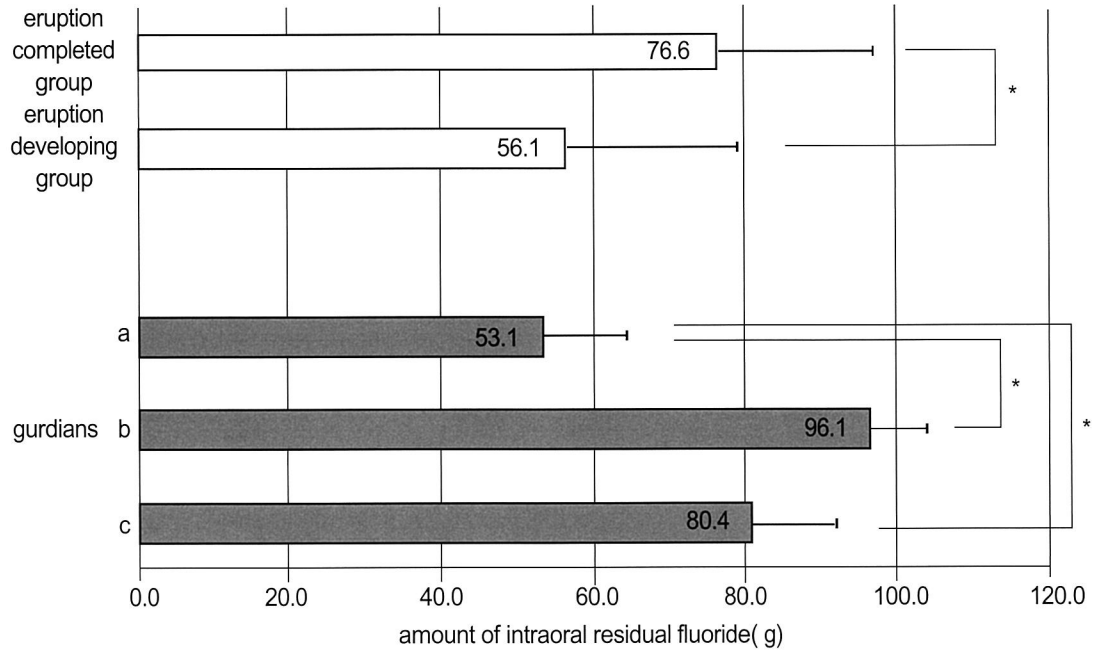


Figure 2. *Intraoral residual fluoride by groups and by guardian in the eruption completed group(*:p<0.05)*

amount of WFS that is applied to tooth brush by guardians in order to secure further safety. In addition, it is necessary to find scientific bases for establishing an appropriate time-frame during a day when TWFS is implemented.

Upon solving these problems, the next task is to create an environment where the application of TWFS at home can be spread widely and maintained.

Acknowledgements

We would like to express our thanks to the subjects and their guardians who cooperated to this study. This research was presented in part at the 42th general meeting of Kanagawa Shigaku held in Yokosuka, December 8, 2007.

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Original Article

Fluoride Intake from Tea in Japanese Infants

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Abstract

We surveyed the state of tea intake in Japanese infants by means of questionnaire, and estimated the amount of fluoride intake from tea. Guardians of day nursery children through 21 day nurseries in six prefectures (designated major tea producing prefectures into the tea-growing district group and other prefectures into the non tea-growing district group) answered the questionnaire. We also estimated the amount of daily fluoride intake in each child.

It was suggested that intakes of teas with high fluoride content such as *sencha*, oolong tea and black tea were somewhat low in infants younger than 1 year and 6 months in the regions with non-tea-growing districts. Children in the tea-growing district group started drinking Japanese and oolong teas at younger age than did children in the non-tea-growing district group. The estimated amount of daily fluoride intake from tea was significantly higher in the tea-growing district group than in the non-tea-growing district group.

The findings obtained from this study indicate that Japanese takes teas which contain high fluoride from relatively young age, suggesting that fluoride from tea affects systemically. It is also demonstrated that children in tea-growing districts started drinking green tea at young age, and the estimated amount of daily fluoride intake is a substantial portion of the total fluoride intake. Therefore, the optimum fluoride concentration in tap water should be determined by taking into consideration the fluoride intake from tea in children.

Keywords: Fluoride intake, Tea, Japanese infant, Systemic effect

Int J Oral Health 2009;5:7-15

Introduction

The prevalence of dental caries has greatly decreased world-wide as people implement fluoride application for prevention of dental caries by following the recommendation made by many professional organizations such as WHO and FDI (Rugg-Gunn. 2001). In Japan, the market share of fluoride dentifrice, which was 10% or so in the first half of 1986, increased dramatically to 89% in 2006 (The Japan Association for Dental Hygienists Education. 2007). The number of children who participated in school-based fluoride mouth-rinsing was approximately 100,000 in 1983 (Kimoto *et al.* 2005) and increased to 491,334 in 2006 (NPO-JPUF. 2008). Of children age between 1 and 14 years old, 11% of them had an experience of professionally applied topical fluoride in 1975, which increased to 59% in 2005(The Statistical Analysis Committee on the Survey of Dental Diseases. 2005). As described above, topical fluoride application in Japan has become popular. However, systemic fluoride application such as communal water fluoridation has not been implemented.

Fluoride also exists in the nature such as sea water and foods and is one of the nutrients necessary for promotion of lifelong health. Whereas, in the Western countries, the adequate intakes of daily fluoride as well as tolerable upper intake levels are set up for age groups (The Fluoride and Fluoridation Research in Japan. 2007), there are no fluoride reference intakes in Japan.

Tea, one of the favorite drinks of Japanese, has relatively high fluoride content. It is possible that fluoride originating from tea has a systemic effect in individuals who drink tea daily. Therefore, it is necessary to take into consideration the fluoride intake from tea, when the fluoride concentration in tap water is to be determined from communal water fluoridation in Japan.

In the present study, the state of tea intake is

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Table 1. *Number and demographic characteristics in study subjects*

Region	Total	Male	Female	Age
Aomori	95	47	48	4.42 – 1.52 *
Gunma	199	94	105	3.83 – 1.28
Tokyo	59	31	28	3.44 – 1.58
Kanagawa	85	35	50	3.82 – 1.34
Shizuoka	802	398	404	4.25 – 1.53
Kagoshima	90	46	44	4.40 – 1.13
Total	1330	651	679	4.14 – 1.48

* Mean –S.D

Table 2. *Number of subjects by age classes in each group*

Age class	Tea-growing district	(%)	Non tea-growing district	(%)
<1-year and 6-month	45	(3.4)	25	(1.9)
1-year and 6-month to < 3-year	138	(10.4)	88	(6.6)
3-year to < 4-year and 6-month	264	(19.9)	162	(12.2)
‡ 4-year and 6-month	445	(33.5)	163	(12.3)
Total	892	(67.1)	438	(32.9)

surveyed and investigated in infants who tend to have systemic effects of fluoride because their teeth are in the stage of formation. The amount of fluoride intake from tea was estimated and factors affecting the estimated amount of fluoride intake were analyzed.

Materials and Methods

Study subjects

Unregistered questionnaires were delivered to guardians of nursery children through 21 day nurseries, which were in cooperation with the study, in Aomori, Gunma, Tokyo, Kanagawa, Shizuoka and Kagoshima prefectures. After inappropriate answers were excluded, analyses were conducted in 651 male children and 673 female children, totaling 1,330 children (mean age: 4.1–1.5 years old, 0 to 6 years old). The recovery rate of the questionnaires was 79.4%. Table 1 lists the number of subjects by regions and demographic characteristics.

It was conjectured that as the amount of tea production would differ by regions, the tea intake also would differ by regions. Accordingly, based on statistics of the amount of pre-refining tea production compiled by the Ministry of Agriculture, Forestry and Fisheries (The Ministry of Agriculture, Forestry and Fisheries of Japan. 2008), major tea producing prefectures of Kagoshima (the amount of tea production in 2002: 18, 400t) and Shizuoka (36,900t) were designated into the tea-growing district group, and prefectures of Aomori (0t), Gunma (10t), Toyo (72t) and Kanagawa (216t) were the non tea-growing district group. In addition, as it was conjectured that the state of tea intake would differ by age, subjects were divided into 4 age classes (Table 2).

Questionnaire on intakes of tea and other drinks

Figure 1 shows the contents of the questionnaire used in this study. Guardians were asked to answer all of 11 questions including if children liked or disliked Japanese teas and other drinks, age when child started drinking tea, the type of tea and amount of daily drinks tea and how Japanese teas were brewed.

Estimation of the amount of daily fluoride intake from tea

Referring to a report by Uematsu (2001), the amount of daily fluoride intake was estimated by multiplying the mean concentration of extracted fluoride (ppm) calculated using time (sec) and temperature (℃) needed to brew teas listed in question IV and V of Figure 1 by the amount of tea drunk (cc) recorded in question IV.

Factors affecting the estimated amount of fluoride intake

For the purpose of elucidating factors affecting the estimated amount of daily fluoride intake from tea, multiple logistic regression analysis was performed using the estimated amount of fluoride intake from tea as the dependent variable. Because the median of estimated fluoride intake of subjects was 91.0 g,

I. Age
II. Gender
III. 1. Does your child like to drink Japanese tea (*sencha*, coarse tea, roasted tea, etc.)?
1. like 2. undecided 3. has never drunk Japanese tea 4. does not like
2. Does your child like to drink oolong tea?
1. like 2. undecided 3. has never drunk ooiong tea 4. does not like
3. Does your child like to drink barley tea?
1. like 2. undecided 3. has never drunk barley tea 4. does not like
4. Which type of tea does your child drink frequently? (multiple answer can be chosen)
1. *sencha* 2. coarse tea 3. roasted tea 4. oolong tea 5. black tea
6. barley tea 7. others (name:) 8. no tea drinking
5. What was the age when your child started drinking tea?
Japanese tea: about year and month (1-year or younger/older than 1-year)
Oolong tea: about year and month (1-year or younger/older than 1-year)
6. What kind of drink does you child (multiple answers can be chosen)?
1. Fruit beverage 2. dairy drink 3. lactic acid beverage
4. carbonated beverage 5. mineral water 6. vegetable beverage
7. sports drink 8. nourishing & functional drink 9. others

IV. When, how much and what type of tea does you child drink daily?

Time-frame	Before breakfast	After breakfast	In the morning	At lunch time	In the afternoon	After dinner	At night
Types of tea*							
Amount of drink (about 000 cc)							

* Answer the number written for teas in Question III-4.

V. How do you prepare daily Japanese tea (*sencha*, coarse tea and roasted tea) for your child?
A. How do you pour boiled water onto tea leaves?
1. Boiled water (near 100℃) is poured directly onto tea leaves (including the tea is drunk after cooled naturally).
2. Boiled water is poured directly onto tea leaves and water (1. a little 2. about 1/3 to 1/2 of tea 3. more than 1/2 of tea) is added to cool.
3. Boiled water is cooled a little by pouring first into tea cups and then collecting in a teapot and then poured onto tea leaves.
4. Other ways of pouring onto tea leaves(please describe)
B. How long do you brew tea leaves?
1. Less than 10sec. 2. 10 to 29 sec. 3. 30 to 59 sec. 4. longer than 1 min.

The mark / presents the boundary between levels in logistic regression analysis.

Figure 1. *Contents of questionnaire used in the study*

two levels of low fluoride intake (90 g or less) and high fluoride intake (higher than 90 g) were designated as the dependent variable. Eight items of tea-growing district or non-tea-growing district , age classes , like or dislike of Japanese tea , like or dislike of oolong tea , like or dislike of barley tea . age when your child started drinking Japanese tea , and age when your child started drinking oolong tea were designated as independent as independent variable. With each independent variable, tea-growing district or non-tea-growing district was in two levels and age class was in four levels. With three variables of like or dislike of Japanese tea , like or dislike of oolong tea and like or dislike of barley tea , the like of any tea was designated to one level, and three choices of undecided , never had drunk and dislike were unified as others and designated to one level (total came to two levels). Variables of age when your child started drinking Japanese tea and age when your child started drinking oolong tea were in 2 levels of 1 year old or younger and older than 1 year old . The boundaries of levels in each independent variable are presented with the / in Figure 1.

Statistical analysis

Because there was no difference in tea intake

Table 3. *Results of investigation by questionnaires (%)*

Item	Choices	TGD*	NTGD*	Total	P-value
1. Preference of Japanese tea	Like	62.5	52.9	59.4	.0015
	Undecided	32.5	39.8	34.9	
	Has never drunk	1.1	3.4	1.9	
	Does not like	3.8	3.9	3.8	
2.Preference of oolong tea	Like	39.0	35.5	37.9	N.S
	Undecided	41.5	41.2	41.4	
	Has never drunk	12.1	16.0	13.4	
	Does not like	7.3	7.3	7.3	
3. Preference of barley tea	Like	70.6	73.3	71.5	N.S
	Undecided	25.8	24.0	25.2	
	Has never drunk	1.9	1.1	1.7	
	Does not like	1.7	1.6	1.7	
4. Type of tea to drink frequently (multiple answer can be chosen)	*Sencha*	65.9	37.9	56.7	<.0001
	Coarse tea	16.1	15.5	15.9	N.S
	Roasted tea	5.4	23.1	11.2	<.0001
	Oolong tea	28.7	30.1	29.2	N.S
	Black tea	15.2	21.7	17.4	.0036
	Barley tea	61.3	78.3	66.9	<.0001
	Others	5.9	7.8	6.5	N.S
	No tea drinking	2.9	6.2	4.0	.0044
5.Age started drinking Japanese tea	<1-year and 6-month	88.3	69.1	82.4	<.0001
	1-year and 6-month to < 3-year	9.0	21.6	12.9	
	3-year to < 4-year and 6-month	2.3	7.9	4.0	
	‡ 4-year and 6-month	0.5	1.4	0.8	
6. Age started drinking oolong tea	<1-year and 6-month	63.8	55.4	61.2	N.S
	1-year and 6-month to < 3-year	24.1	28.2	25.4	
	3-year to < 4-year and 6-month	11.6	15.7	12.9	
	‡ 4-year and 6-month	0.5	0.7	0.6	
7. Kinds of drink to take other than tea (multiple answer can be chosen)	Fruit beverage	56.4	60.7	57.8	N.S
	Dairy drink	65.6	67.4	66.2	N.S
	Lactic acid beverage	41.9	40.0	41.3	N.S
	Carbonated beverage	15.1	24.7	18.3	<.0001
	Mineral water	10.8	11.4	11.0	N.S
	Vegetable beverage	23.1	28.3	24.8	.0048
	Sports drink	35.8	45.0	38.8	.0016
	Nourishing & functional Drink	2.9	4.6	3.5	N.S
	Others	2.5	3.0	2.6	N.S

Each numerical value represent percentages after unclear answers were excluded.
Statistical tests were performed for results of 4×2 contingency table for questions 1,2,3,5 and 6. Questions 4 and 7 are results of 2×2 contingency table for drink frequently or drink none and tea-growing district or non-tea grewing district .
P values of over 0.05 was considered to indicate not significant(NS).
*TGD: Tea-growing district, NTGD: Non-tea-growing district

between genders analyses were performed without dividing subjects into two groups of gender. Cross tabulation and proportions of categorical data obtained from questionnaires were examined using x^2 test. For test of mean difference of numerical data, Student s t test was used for two samples and analysis of variance (ANOVA) and then, multiple comparisons (Tukey-Kramer s HSD test) for three or more samples.

Hideyoshi 2003 (Wass Home 3.0, Social Survey Research Information Co., Ltd.) was used for summarization of questionnaires and JMP 7.0.1J (SAS Institute Japan) and SPSS Ver 11.0J (SPSS Japan Inc.) for statistical analysis.

Results

Investigation using questionnaire

Results of a cross tabulation of responses to questionnaire are listed in Table 3. Regarding Japanese tea, there was a significant difference that subjects who liked Japanese tea were 52.9% of those in the non-tea-growing district group and 62.5% of

Table 4. *Age when child started drinking tea in each region*

	Analyzed number	Age started drinking						
		Total	Aomori	Gunma	Tokyo	Kanagawa	Shizuoka	Kagoshima
Tea	11.56	0.96	1.71	1.15	1.03	1.15	0.83	0.96
			NTGD*:1.24				TGD*:0.85	
						p<.0001		
Oolong tea	901	1.42	1.92	1.46	1.26	1.59	1.36	1.46
			NTGD*:1.54				TGD*:1.37	
						p= .012		

*NTGD: Non-tea-growing district, TGD: Tea-growing district

Table 5. *The estimated mean amount of fluoride intake from tea per capita in each region (g/day)*

Region	Estimated mean amount of fluoride intake		P-value
Aomori	65.4		
Gunma	154.4	116.1	
Tokyo	82.6		
Kanagawa	106.2	Non-tea-growing district	<.0001
Shizuoka	163.7	161.9	
Kagoshima	147.2	Tea-growing district	
Total	146.8		

those in the non-tea-growing district group. As for oolong tea, whereas there were similar tendencies to Japanese tea, there was no significant difference. As for barley tea, there was no difference in tendencies between the tea-growing district group and non-tea-growing district group.

Types of tea that were frequently drunk were *sencha* (65.9%) followed by barley tea (61.3%) in the tea-growing district group and barley tea (78.3%) followed by *sencha* (37.9%) in the non-tea-growing district group. *Sencha* was drunk significantly more in the tea-growing district group (65.9%) than the non-tea-growing district group (37.9%). Roasted tea (*houjicha*) and black tea were drunk significantly more in the non-tea-growing district group (23.1% and 21.7% respectively) than in the tea-growing district group (5.4% and 15.2% respectively). Subjects who did not drink tea were significantly more in the non-tea-growing district group (6.2%) than in the tea-growing district group (2.9%). The age when child started drinking Japanese tea shifted toward significantly younger age classes in the tea-growing district group than in the non-tea-growing district group.

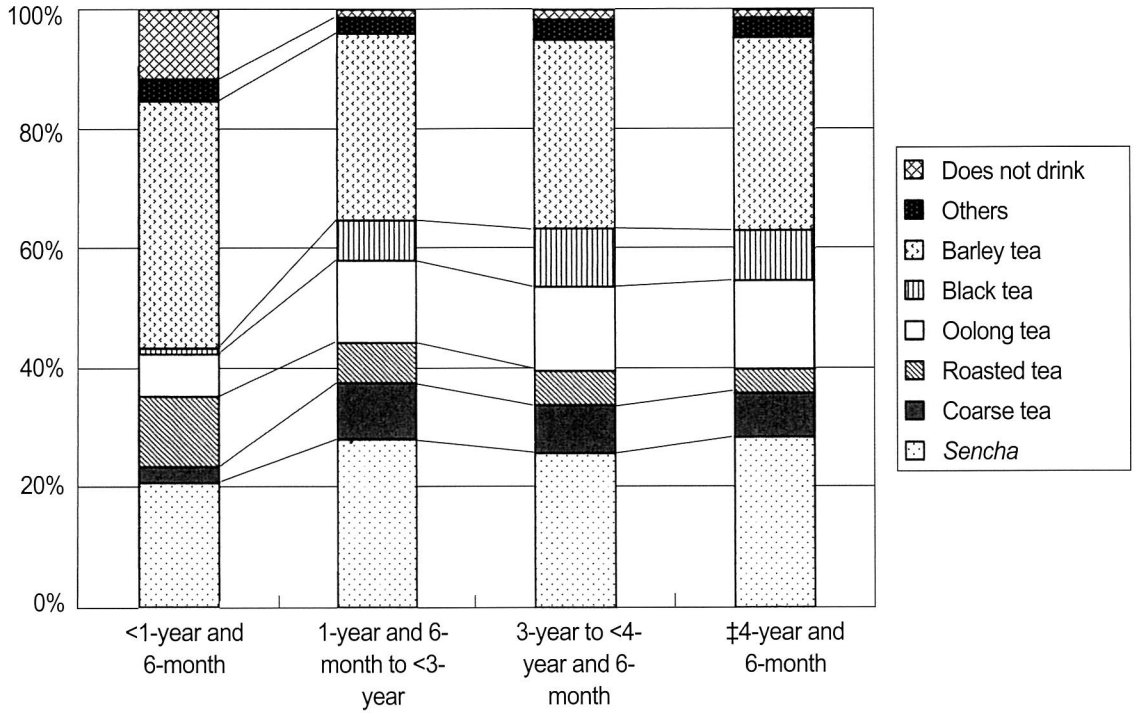
As for the age when child started drinking oolong tea, there was a similar tendency of shifting toward younger age classes, although it was not significant. As for drinks other than teas, carbonated beverages , vegetable beverages and sports drinks are drunk significantly more in the non-tea-growing district group than in the tea-growing district group.

Figure 2 shows changes in types of tea that were frequently drunk according to age classes. Teas that were drunk frequently in all age classes were barley tea , *sencha* and oolong tea. The *sencha* , coarse tea(*sencha*) , oolong tea , black tea , roasted tea (*houjicha*) and no tea drinking changed significantly according to age classes. Of said teas, roasted tea and no tea drinking seemed to be more prevalent in children younger than 1 year and 6 months than in other age classes, whereas other teas showed reverse tendencies.

Table 4 lists mean ages when the child started drinking teas. When compared between the tea-growing district group and the non-tea-growing district group, the age when the child started drinking teas was 0.85 – 0.66 years old in the tea-growing district group and 1.24 – 0.95 years old in the non-tea-growing district group. There was a significant difference between the two groups. The age when the child started drinking oolong tea was 1.37 – 0.88 years old in tea-growing district group and 1.54 – 0.97 years old in the non-tea-growing district group. There also was a significant difference between the two groups.

Estimation of the amount of daily fluoride intake from tea

Table 5 lists the estimated mean amount of fluoride intake from tea per capita calculated in each region. In order of greater intake, the first was



Type of answer in this question was multi-answer. The total number of answers (drink frequently) was defined as 100 and each item was itemized. Age classes and like or dislike of each tea were cross summarized and tested.
Sencha, coarse tea, oolong tea and black tea appeared to be drunk significantly less in children younger than 1-year and 6-month (P<.001, P<.01, P<.01 and P<.001 respectively).
Roasted tea appeared to be drunk significantly more in children younger than 1-year and 6-month (P<.001). No tea-drinking seemed significantly more in children younger than 1-year and 6-month (P<.001). There were no significant differences in Other and Barley tea .

Figure 2. *Type of tea drunk frequently in each age class*

Shizuoka Prefecture (163.7 g), followed by Gunma (154.4 g) Kagoshima (147.2 g), Kanagawa (106.2 g), Tokyo (82.6 g) and Aomori (65.4 g). There was a significant difference in the estimated mean amount of fluoride intake per capita between the tea-growing district group and the non-tea-growing district group.

Table 6 lists the estimated mean amount of fluoride intake per capita calculated in each age class. The two-way ANOVA revealed that both factors of age classes (P<0.001) and districts of tea-growing or non-tea-growing (P<0.001) produced significant differences, and there was no interaction. Multiple comparisons demonstrated that the mean estimated amount of fluoride intake in the age class of younger than 1 year old was significantly lower than that all other age classes.

Factors affecting the estimated amount of fluoride intake

Results of multiple logistic regression analyses are shown in Table 7. Items significantly affecting the estimated amount of fluoride intake based on multivariate odds ratios were districts of tea-growing or non-tea-growing , age classes and like or dislike of Japan tea .

The estimated amount of fluoride intake was compared between subjects who drank frequently any one of several drinks (Fig. 1, III.6) other than green tea, that is, carbonated beverages, lactic acid beverages and sport drinks, and who drank none. There was no significant difference between the two groups.

Discussion

Tea intake is said to not only be useful for systemic health promotion such as prevention of cardiovascular disease and cancer (Trevisanato *et al.* 2000) but also contribute to the prevention of dental caries because of relatively high content of fluoride. It has been reported that daily drinking of coarse tea (*bancha*) means the daily intake of additional 0.22 to 0.35 mg fluoride, which decreased pit and fissure caries by 22.1% and proximal surface caries by 26.1% (Ohnishi. 1985). However, another study reported that fluoride intake by daily drinking of tea, which accounted for 32% (Zohouri *et al.* 2000), or a substantial portion of the total fluoride intake, would be a risk factor of dental fluorosis (Mann *et al.* 1985). Mann *et al.* (1985) reported that the more

Table 6. *The estimated mean amount of fluoride intake from tea per capita in each age class (g/day)*

Age class	Estimated mean amount of fluoride intake	Tea-growing district	Non-tea-growing diatrict	Multiple comparison
<1-year and 6-month	43.4	46.7	37.4	a*
1-year and 6-month to < 3-year	136.3	149.2	116.1	b
3-year to < 4-year and 6-month	143.6	160.7	115.7	b
‡ 4-year and 6-month	165.0	178.3	128.6	b
Total	146.8			

*There are significant differences among different alphabets.

Table 7. *Logistic regression analysis with the estimated fluoride intake as the dependent variable*

Item	Crude odds radio	Multiple adjusted odds radio	95.0% CI for Multiple adjusted odds radio (P-value)
Tea-or non-tea-growing districts	1.84	1.19	1.16 - 2.15 (.004)
Preference of Japanese tea	2.61	2.26	1.62 - 3.14 (<.001)
Preference of oolong tea	0.96	0.88	0.65 - 1.16 (.340)
Preference of barley tea	1.43	0.74	0.52 - 1.07 (.107)
Age class	1.27	1.58	1.01 - 1.41 (.043)
Age started drinking Japanese tea	1.49	1.14	0.75 - 1.73 (.538)
Age started drinking oolong tea	1.05	0.95	0.65 - 1.16 (.340)

frequently children drank teas, the significantly lower the incidence of dental caries of deciduous and permanent teeth was and the higher the incidence of dental fluorosis was. Therefore, the amount of fluoride ingested from teas should be examined from both beneficial aspect and deleterious one. The present study investigates the state of fluoride intake in infants who are susceptible to fluoride because they have the formative teach.

Uematsu (2001) investigated by questionnaires the likings of tea and tea-drinking behavior of students of a dental college whose home towns were divided into tea-producing districts (yearly production 2,500 tons or more) and non-tea-producing districts. The study found that whereas there was no difference in the likings of teas between the two districts, how to brew teas differed and the tea-producing districts had a specific way of brewing. Accordingly, for analyses of data in the present study, Shizuoka and Kagoshima prefectures, which are the first and second prefectures in the production of unrefined tea, were classified as the tea-growing district group and others, Kanagawa, Tokyo, Gunma and Aomori Prefectures were classified in the non-tea-growing district group. Subjects who liked Japanese tea were 62.5% of those in the tea-growing district group and 52.9% in the non-tea-growing district group, the former exceeding the latter by 9.6% points (P=0.0015). Of types of teas drunk frequently, *sencha* was drunk by 65.9% of subjects in the tea-growing district group and 37.9% in the non-tea-growing district group, the former exceeding the latter by 28 points (P<0.0001). On the other hand, the percentages of subjects who drank roasted tea , black tea . barley tea and no tea drinking were higher in the non-tea-growing district group than in the tea-growing district group by 17.7 points (P<0.0001), 6.5 points (P=0.0036), 17.0 points (P<0.001) and 3.3 points (P=0.0044) respectively. The above results demonstrated that whereas children in regions with high production of green teas liked and started drinking *sencha* at younger age, children in regions with low production of green tea shifted toward drinking roasted tea , black tea , barley tea and no tea drinking . In addition, percentages of subjects who drank carbonated beverages , vegetable beverages and sports drinks were higher in the non-tea-growing district group than in the tea-growing district group by 9.6 points (P<0.0001), 5.2% (P=0.0048) and 9.2 points (P=0.0016) respectively, indicating the shift toward these drinks in the non-tea-growing district group. Many infants younger than 1 year and 6 months in the non-tea-growing district group drank roasted tea or no tea. Although there was no significant difference, the barley tea was also drunk by many infants. These results suggested that intakes of teas with high fluoride content such as *sencha* , oolong tea and black tea were somewhat low in infants younger than 1 year and 6 months, particularly younger ones, in the regions with low tea production (non-tea-growing districts). Thus, it was thought that systemic effects of fluoride from tea were small.

Children in the tea-growing district group started drinking Japanese and oolong teas at younger age than did children in the non-tea-growing district group ($P<0.0001$ and $P=0.012$ respectively) The results suggested that as children in the tea-growing district group took fluoride from tea during the dental calcification stage, it was possible that fluoride affected systemically.

The amount of fluoride intake from tea was estimated by referring to the report by Uematsu (2001). The amount of intake was high in subjects in Shizuoka, Gunma and Kagoshima prefectures, suggesting that intake of fluoride from tea was significantly higher in the tea-growing district group than in the non-tea-growing district group ($P<0.0001$).

The estimated amount of fluoride intake also differed by age classes. The estimated amounts of fluoride intake were significantly higher in all of other age classes than younger than 1 year and 6 months. In particular, it was suggested that fluoride intake from tea affected systemically in the tea-growing district group. The daily fluoride intake in subjects older 4 year and 6 months was 178 g in the tea-growing district group and 129 g in the non-tea-growing district group, the former exceeding 134 g reported by Zohouri et at (2000). Multiple logistic analyses revealed that factors affecting the estimated amount of fluoride intake were the tea-growing district or the non-tea-growing district , like or dislike Japanese tea and age classes , substantiating the above-described tendencies.

The above findings indicates that Japanese takes teas which contains high fluoride from relatively young age, suggesting that fluoride from tea affects systemically, particularly so in the regions with high tea production. Accordingly, the prevalence of dental caries of deciduous and permanent teeth was investigated in subject children in each prefecture.

At the 1-year and 6-month old children s dental health examination conducted in 2005 (NPO-JPUF. 2008), the prevalence of dental caries was 4.8% in Aomori Prefecture, 3.9% in Gunma, 2.5% in Tokyo, 2.5% in Kanagawa, 2.3% in shizuoka and 5.5% in Kagoshima, the lowest being Shizuoka Prefecture. At the 3-year old children s dental health examination in the same year 2005 (NPO-JPUF.2008), the prevalence of deciduous dental caries was 44% in Aomori Prefecture, 28.7% in Gunma, 19.4% in Tokyo, 21.4% in Kanagawa, 20.9% in Shizuoka and 35.3% in Kagoshima, Shizuoka Prefecture being at the second lowest. The prevalence of deciduous teeth caries appears to be higher in the Tohoku and Kyushu districts, which are remote from the center of Tokyo. Kagoshima, one of subject districts, was the third lowest following Fukuoka and Saga among seven prefectures in Kyushu.

The DMFT index in 12-tear old children (in the survey of school health statistics in 2006) was 1.3 in Tokyo Metropolis, 1.4 in Kanagawa Prefecture, 1.4 in Shizuoka, 1.7 in Gunma, 2.2 in Aomori and 2.3 in Kagoshima, indicating that the prevalence of dental caries in Shizuoka was the third lowest and that in Kagoshima was the fifth lowest among seven Kyushu prefectures. The present study found that shizuoka Prefecture, one of the tea-growing districts, has the highest estimated amount of fluoride intake from tea and the low prevalence of dental caries in deciduous and permanent teeth. However Kagishima Prefecture also a tea-growing district, had the lower estimated amount of fluoride intake from tea than Gunma Prefecture, a non-tea-growing district, suggesting the cause of high prevalence of dental caries.

At any rate, the present study demonstrated that children in tea-growing districts started drinking green tea at young age and the estimated amount of daily fluoride intake was 164 g, which was a substantial portion of the total fluoride intake. Although communal water fluoridation has not been implemented in Japan, the optimum fluoride concentration in tap water has been inferred (Tsusui.2003). However, the intake of fluoride from tea accounts for 60% or more of the total daily intake of fluoride, and it has been reported that individual intake fluctuates widely between 0.04 and 2.7 g (Duckworth *et al*. 1978). Therefore, the optimum fluoride concentration in tap water should be determined by taking into consideration the fluoride intake from tea in children.

Future studies include effects of fluoride intake during the period of tooth formation in children on dental caries and fluorosis.

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Original Article

Reduction of Gingival Bleeding by Professional Toothbrushing Compared to One-stage Full-mouth Disinfection

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Abstract

A foundation for effective periodontal treatment is the removal of subgingival plaque, and this may be carried out using the one-stage full-mouth disinfection method. In addition, the host response should be considered in periodontitis. In this study, gingival bleeding by professional toothbrushing on moderate chronic periodontitis were compared with those of one-stage full-mouth disinfection. Twelve periodontitis patients were randomly assigned to professional toothbrushing or one-stage full-mouth disinfection and received the assigned treatment daily from Day 0 to 7, and once a week from Day 7 to 28. On Day 0, 7, 14 and 28, subgingival plaque samples were obtained and clinical parameters were recorded on 4 teeth per patient. Bacterial DNAs from subgingival plaque of the teeth were analyzed using polymerase chain reaction- denaturing gradient gel electrophoresis and real-time polymerase chain reaction. Professional toothbrushing resulted in a decrease in the number of periodontal pathogens that followed the decrease in bleeding on probing. Moreover, professional toothbrushing improved the clinical parameters as effectively as one-stage full-mouth disinfection. The treatment time for professional toothbrushing was about 1/4 of that for one-stage full-mouth disinfection. These findings highlight the merit of professional toothbrushing in improving gingival bleeding and reducing the proportion of periodontal pathogens in the subgingival plaque.

Key words: Periodontitis, professional toothbrushing, one-stage full-mouth disinfection, real-time polymerase chain reaction, mechanical stimulation

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Introduction

Some 40 years ago, it was demonstrated that cessation of toothbrushing resulted in deposition of plaque followed by gingivitis; with resumption of toothbrushing, plaque was removed and inflammation resolved (L e *et al.* 1965). Results of this experimental gingivitis study confirmed the central etiological role of supragingival plaque in gingivitis. In the case of periodontal disease, subgingival plaque is considered to be the principal source of pathogenic organisms, and treatment of periodontal disease is centered around its removal, most effectively performed using the one-stage full-mouth disinfection method (Quirynen *et al.* 1995). Toothbrushing is also believed to be an effective means of helping in the treatment, as well as prevention, of periodontal disease, even though access to subgingival plaque is limited.

A growing body of evidence suggests that

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mechanical stimulation of the gingiva by toothbrushing promotes local host defense mechanisms. Toothbrushing is more effective than plaque removal with a curette in reducing inflammatory cell infiltration and increasing the number of collagen-synthesizing gingival fibroblasts (Horiuchi *et al.* 2002). Brushing twice a day reduces gingival inflammation and favorable clinical effects appear earlier than with brushing once a day (Yamamoto *et al.* 2004). Toothbrushing every other day maintains gingival health, although the amount of accumulated plaque is theoretically sufficient enough to induce gingivitis (Lang *et al.* 1973). These results suggest that the effect of toothbrushing on gingivitis is not only due to plaque removal but also stimulation of the host defense mechanism.

Toothbrushing should cover all the gingiva around a tooth, as the effect of mechanical stimulation on gingival cell activation is limited to the brushed area (Sakamoto *et al.* 2003). Self toothbrushing may not be technically enough to stimulate all the gingiva, and professional toothbrushing is recommended in periodontal diseases.

The effects of professionally delivered meticulous supragingival plaque control on clinical symptoms of periodontitis and composition of subgingival microbiota have been shown (Smulow *et al.* 1983, Dahlén *et al.* 1992, Katsanoulas *et al.* 1992, McNabb *et al.* 1992, Al-Yahfoufi *et al.* 1995, Hellstr m *et al.* 1996). In these studies, supragingival scaling and polishing were performed. No information is available regarding the clinical and microbiological effects of professional toothbrushing. Here, we performed a single-blind randomized clinical study to compare the effects of professional toothbrushing and one-stage full-mouth disinfection on the clinical parameters and subgingival microbiota in patients with moderate chronic periodontitis.

Materials and methods

Patient selection and experimental design

Twelve periodontitis patients (mean age 56 years, range 41-74 years) with at least 20 teeth and at least 4 teeth with a probing pocket depth of 5 mm or more were randomly assigned to the toothbrushing or one-stage full-mouth disinfection group. Randomization was performed using a table of random numbers. Individuals were excluded if they were pregnant, had received periodontal therapy, antibiotics or an antimicrobial product in the previous 3 months, or if they had any systemic condition which may affect the progression or treatment of periodontitis. The study protocol was approved by the Ethical Committee of the Okayama University Dental School, and subjects provided written informed consent for study participation.

Treatment in the toothbrushing group consisted of professional toothbrushing using the Toothpick Method (Morita *et al.* 1998) once daily from Days 0 to 7 and on Days 14 and 21. No oral hygiene instruction was provided. In the one-stage full-mouth disinfection group, removal of supra- and subgingival plaque and calculus was performed within the first 24 hrs (Quirynen *et al.* 1995), and supra- and subgingival plaque removal was subsequently performed from Days 3 to 7 and on Days 14 and 21. Subgingival irrigation with 0.04% chlorhexidine, brushing of the dorsum of the tongue and rinsing with 0.04% chlorhexidine were also performed in the one-stage full-mouth disinfection group. Both treatment groups maintained self-performed oral hygiene measures between visits.

Sample collection and DNA extraction

In each patient, 4 teeth with a probing pocket depth of 5 mm or more at baseline were selected as the representative teeth. Microbiological sampling was performed on Days 0, 7, 14, and 28. After removal of supragingival plaque, the sampling sites of the representative teeth were isolated with sterile cotton roll, and then air-dried. The subgingival plaque sample was removed from each pocket using a sterile curette (Paster *et al.* 2001). The sample was immersed in 1.0 ml phosphate-buffered saline (pH 7.2) (Invitrogen Co., Grand Island, NY, USA), mixed for 30 sec, and centrifuged at 12,000 g for 20 min to remove the supernatant. The bacterial DNAs were extracted from cultivated strains and clinical plaque samples using a kit (InstaGene¤ Matrix, Bio-Rad Lab., Hercules, CA, USA) and according to the manufacturer s instructions. The extracted DNAs were used in polymerase chain reaction (PCR) - denaturing gradient gel electrophoresis (DGGE) and real-time PCR analyses.

PCR-DGGE analysis

An approximately 585-bp 16S rDNA fragment corresponding to nucleotide positions from 341 to 926 in the sequence of *Escherichia coli* was amplified with two conserved primers. A forward primer, with a 40-bp GC-rich sequence added to the 5 -end (5 -CGCCCGCCGCGCCCCGCGCCCGT C C C G C C G C C C C C G C C C G - C C T A CGGGAGGCAGCAG-3), and reverse primer (5 -

Table 1. *Primers used for real-time PCR*

Targets	Forward Reverse	Sequence of the primer (5 -3)
Aggregatibacter actinomycetemcomitans[a]	F	CTTACCTACTCTTGACATCCGAA
	R	ATGCAGCACCTGTCTCAAAGC
Porphyromonas gingivalis[a]	F	CTTGACTTCAGTGGCGGCAG
	R	AGGGAAGACGGTTTTCACCA
Prevotella intermedia[a]	F	AATACCCGATGTTGTCCACA
	R	TTAGCCGGTCCTTATTCGAA
Tannerella forsythia[b]	F	GCGTATGTAACCTGCCCGCA
	R	TGCTTCAGTGTCAGTTATACCT
Treponema denticola[b]	F	TAATACCGAATGTGCTCATTTACAT
	R	CTGCCATATCTCTATGTCATTGCTCTT
Total bacteria[a]	F	GTGSTGCAYGGYTGTCGTCA
	R	ACGTCRTCCMCACCTTCCTC

[a] Designed according to published methods (Maeda *et al.* 2003).
[b] Designed originally for this study.

CCGTCAATTCCTTTRAGTTT-3) were prepared for the PCR. Amplification was performed using a kit (EX-Taq¤, TaKaRa Shuzo Ltd., Otsu, Japan) in 50- l reactions. The thermocycling program was performed as previously described (Fujimoto *et al.* 2003). Prior to DGGE analysis, the PCR products were visualized by electrophoresis in a 2% agarose gel to confirm the 16S rDNA amplification.

A DCode Universal Mutation Detection System (Bio-Rad Lab.) was used for sequence-specific separation of the PCR products (Muyzer & Smalla 1998). The PCR products were separated on a 6% (w/v) polyacrylamide gel containing a linear gradient ranging from 20 to 50% denaturant. The gel was run for 5.5 hrs at 200V in 0.5 x Tris-acetate-EDTA (TAE) buffer (1 x TAE buffer contains to 0.04 mol/l Tris base, 0.02 mol/l acetic acid and 1.0 mmol/l EDTA, pH 8.5) at a constant temperature of 60ßC. After staining with ethidium bromide, the gel was viewed under UV transillumination.

Partial 16S rDNA gene sequencing of excised denaturing gradient gel bands

The dominant 16S rDNA bands on the denaturing gradient gel were excised, purified, and re-amplified using PCR with the same conditions described above. The re-amplified DNA fragments were cloned into a vector (TOPO TA Cloning kit¤, Invitrogen, Carlsbad, CA, USA), and the insert DNAs were sequenced using a kit (ABI Prism BigDye¤ terminator v1.1 cycle sequencing ready reaction kit, Perkin Elmer, Foster City, CA, USA) and an automated DNA sequencer (ABI PRISM 3100, Perkin Elmer). The sequence data were subjected to the BLAST sequence homology search program at GenBank and analyzed. The bacterial species were identified at similarity values above 99%.

Bacterial strains and primers for real-time PCR

The bacterial strains used for real-time PCR were *Aggregatibacter actinomycetemcomitans* ATCC 43718 (Y4), *E. coli* K-12, *Porphyromonas gingivalis* ATCC 33277^T, *Prevotella intermedia* ATCC 25611^T, *Tannerella forsythia* ATCC 43037^T, and *Treponema denticola* ATCC 35405^T. Table 1 shows the sequences of primers and probes, designed using software (Primer Express, version 1.0, PE Applied Biosystems, Foster City, CA, USA; OLIGO, version 4.0, Molecular Biology Insights, Cascade, CO, USA) (Maeda *et al.* 2003). For the identification of bacterial species, primers were designed from the species-specific region on the 16S rDNA, whereas the conserved sequences were used for amplification of all bacteria present.

Real-time PCR analysis

The real-time PCR reactions were performed using the GeneAmp¤ 5700 Sequence Detection System (PE Applied Biosystems). The reaction mixture (25 l) contained 13 1 2 x double-stranded DNA-binding reporter fluorescent dye solution (SYBR¤ Green PCR Master Mix, PE Applied Biosystems), 1 l of forward and reverse primer, 2.5 l of extracted DNA and sterile distilled molecular biology grade water to adjust the volume and brought to thermocycling program (Maeda *et al.* 2003). The specificity of the amplification products was assayed with dissociation curves, constructed in the range of 60℃ to 90℃. All data were analyzed

Table 2. *Clinical findings over time by treatment group*

	Treatment group[a]	Day 0	Day 7	Day 14	Day 28
Gingival index	PT	1.9 – 0.5[b]	0.6 – 0.6[d]	0.2 – 0.4[d]	0.1 – 0.3[d]
	FDis	1.5 – 0.5 [g]	0.7 – 0.6[d]	0.4 – 0.5[d]	0.2 – 0.4[d]
Probing pocket depth (mm)	PT	6.3 – 1.5	5.2 – 1.5[d]	4.5 – 1.5[d]	4.4 – 1.5[d]
	FDis	5.5 – 0.7 [g]	4.7 – 1.1[c]	4.3 – 1.2[d]	3.7 – 1.0[d]
Probing attachment level (mm)	PT	6.5 – 1.5	5.3 – 1.6[d]	4.7 – 1.6[d]	4.6 – 1.8[d]
	FDis	5.8 – 1.0 [g]	4.9 – 1.2[c]	4.6 – 1.4[c]	4.0 – 1.4[d]
Presence of bleeding	PT	21	5[f]	3[f]	2[f]
on probing (number of sites)	FDis	21	12[e] [h]	9[f] [h]	6[f]
Plaque index	PT	2.2 – 0.7	0.6 – 0.5[d]	0.5 – 0.7[d]	0.5 – 0.6[d]
	FDis	2.3 – 1.5	0.9 – 1.1[d]	0.8 – 0.8[d]	0.4 – 0.5[d]

[a] PT = Professional toothbrushing, FDis = One-stage full-mouth disinfection.
[b] Data are expressed as mean values – (SD). (n = 24 teeth)
[c] $p < 0.01$, [d] $p < 0.001$ Wilcoxon signed-ranks test, compared to day 0.
[e] $p < 0.01$, [f] $p < 0.001$ Chi-square test, compared to day 0.
[g] $p < 0.05$ Mann-Whitney U-test, compared between the two groups.
[h] $p < 0.05$ Chi-square test, compared between the two groups.

using GeneAmp® 5700 SDS software (PE Applied Biosystems).

Clinical examination

At baseline (Day 0), and on Days 7, 14 and 28, the following clinical parameters were measured on the representative teeth by an examiner blinded to the treatment assignment: probing pocket depth, probing attachment level and bleeding on probing of the deepest pocket. The gingival index (L e & Silness 1963) and a modification of the Quigley-Hein plaque index (Turesky *et al.* 1970) were used to record gingivitis and plaque levels, respectively.

Blinding

Each patient was randomly assigned to one of the two treatment modalities by one researcher (JS). All other contributors to the study were blinded to the generation and implementation of the treatment assignment. The method of randomization was not revealed until all data had been analyzed. The clinical examiner (TY), the therapist (NT), microbiologists (BC, MT, SK, and KF) and the statistician (TT) were blinded to the treatment group.

Statistical analysis

Chi-square test and Wilcoxon signed-ranks test were performed using a statistical package (11.5 J for Windows, SPSS Japan, Tokyo, Japan) with the representative tooth as the unit of analysis.

Results

Bleeding on probing was less common in the professional toothbrushing group than in the one-stage full-mouth disinfection group on Days 7 and 14. There was significant clinical improvement from Day 0 to 7 in both the professional toothbrushing and one-stage full-mouth disinfection groups (Table 2).

As observed by PCR-DGGE, detection frequencies of anaerobic bacteria including *P. gingivalis*, *P. intermedia* and *T. forsythia* were reduced in both groups over time (Table 3). Detection frequencies of aerobic bacteria including *Neisseria mucosa*, *Rhizobium* JEYF14 and *Rothia dentocariosa* were increased. Real-time PCR (Fig. 1) showed that the percentages of some bacterial species including *P. gingivalis*, *P. intermedia*, *T. forsythia* and *T. denticola* decreased in both treatment groups over time.

The average (standard deviation) treatment time per day for the professional toothbrushing group was 15.7 min (2.7 min). The time required for one-stage full-mouth disinfection on the first two visits and subsequent visits was 90.3 min (4.5 min) and 29.6 min (1.9 min), respectively.

Discussion

The results of this study demonstrate that professional toothbrushing using the Toothpick method was more effective in reducing gingival bleeding than the one-stage full-mouth disinfection method. Gingival bleeding is an outer hemorrhage, which is caused by ulceration in periodontal pockets and increased capillary permeability of the gingiva. Mechanical stimulation with toothbrushing enhances proliferation of gingival epithelium and endothelial cells (Horiuchi *et al.* 2002) and thus can reduce

Table 3. *Number of teeth with each bacterial species at each time point according to PCR-DGGE*

	Professional toothbrushing (Day)				One-stage full-mouth disinfection (Day)			
	0	7	14	28	0	7	14	28
Aerobic bacteria								
Neisseria elongata	0[a]	0	0	0	0	3	1	2
Neisseria mucosa	2	6	7	8	0	11	8	9
Neisseria flava	0	0	0	0	0	5	5	4
Pedobacter sp. oral clone AV100	1	1	1	1	1	2	1	4
Rhizobium sp. JEYF14	0	0	2	4	0	2	3	4
Rothia dentocariosa	0	5	3	4	0	4	2	3
Anaerobic bacteria								
Aggregatibacter actinomycetemcomitans	2	2	4	4	0	0	0	0
Actinomyces naeslundii	2	3	2	3	0	2	5	7
Actinomyces odontolyticus	0	0	0	0	0	0	0	3
Corynebacterium durum	0	0	0	0	0	0	1	3
Corynebacterium matruchotii	1	1	3	3	1	2	6	10
Corynebacterium sp. oral clone AK153	0	0	0	0	0	1	1	2
Deferribacteres sp. oral clone D084	1	2	4	2	2	4	0	1
Deferribacteres sp. oral clone W090	0	0	1	3	0	2	4	1
Dialister pneumosintes	2	1	1	0	1	2	2	0
Eubacterium saphenum	0	0	0	0	3	1	2	1
Fusobacterium naviforme	0	0	0	0	2	0	1	1
Fusobacterium nucleatum	2	1	1	2	2	2	3	2
Haemophilus parainfluenzae	0	0	0	0	2	2	0	0
Lautropia mirabilis	10	10	9	4	6	10	8	9
Lautropia mirabilis	10[a]	10	9	4	6	10	8	9
Porphyromonas endodontalis	2	2	2	1	5	4	3	2
Porphyromonas gingivalis	23	9	9	7	24	9	14	8
Porphyromonas sp. oral clone BS077	3	4	6	3	0	0	0	0
Prevotella dentalis	0	0	0	0	2	2	0	0
Prevotella intermedia	7	4	2	0	4	3	0	2
Prevotella loescheii	4	2	2	2	0	0	0	0
Prevotella melaninogenica	3	1	2	1	0	0	0	0
Prevotella nigrescens	0	0	0	0	3	2	0	0
Prevotella odontolyticus	2	4	3	1	0	0	0	0
Prevotella oralis	1	0	0	0	4	3	3	4
Prevotella oris	4	2	1	3	5	5	1	1
Tannerella forsythia	8	8	3	1	3	2	1	0
Treponema denticola	3	2	0	0	0	0	0	0
Veillonella parvula	1	4	3	1	0	2	0	2

[a] Total number of teeth examined was 24.

gingival bleeding. These effects are observed when toothbrushing is performed with optimum force and duration in a limited area (Tomofuji *et al.* 2002, Sakamoto *et al.* 2003, Tomofuji *et al.* 2003). The Toothpick method (Morita *et al.* 1998), in which the bristles are inserted into interdental areas, was effective in reducing the gingival bleeding.

It seems unlikely that toothbrushing was able to directly remove subgingival plaque since the depth of the periodontal pockets was 5 mm or more and the filaments of the toothbrush were unlikely to reach that far into the pocket. The beneficial effects of professional toothbrushing are more likely due, in part at least, to supragingival plaque removal influencing the composition of subgingival microbiota. Some Gram-positive bacteria in supragingival plaque have been shown to serve as attachment sites for *P. gingivalis* and other gram-negative bacteria (Turesky *et al.* 1970). Moreover, bacteria in supragingival plaque produce compounds which are essential growth factors for subgingival bacteria (Slots & Gibbons 1978). Supragingival plaque removal by professional toothbrushing may also have disrupted the equilibrium of the subgingival flora by altering the supragingival environment.

The findings of this study are not in agreement with those of other studies which showed

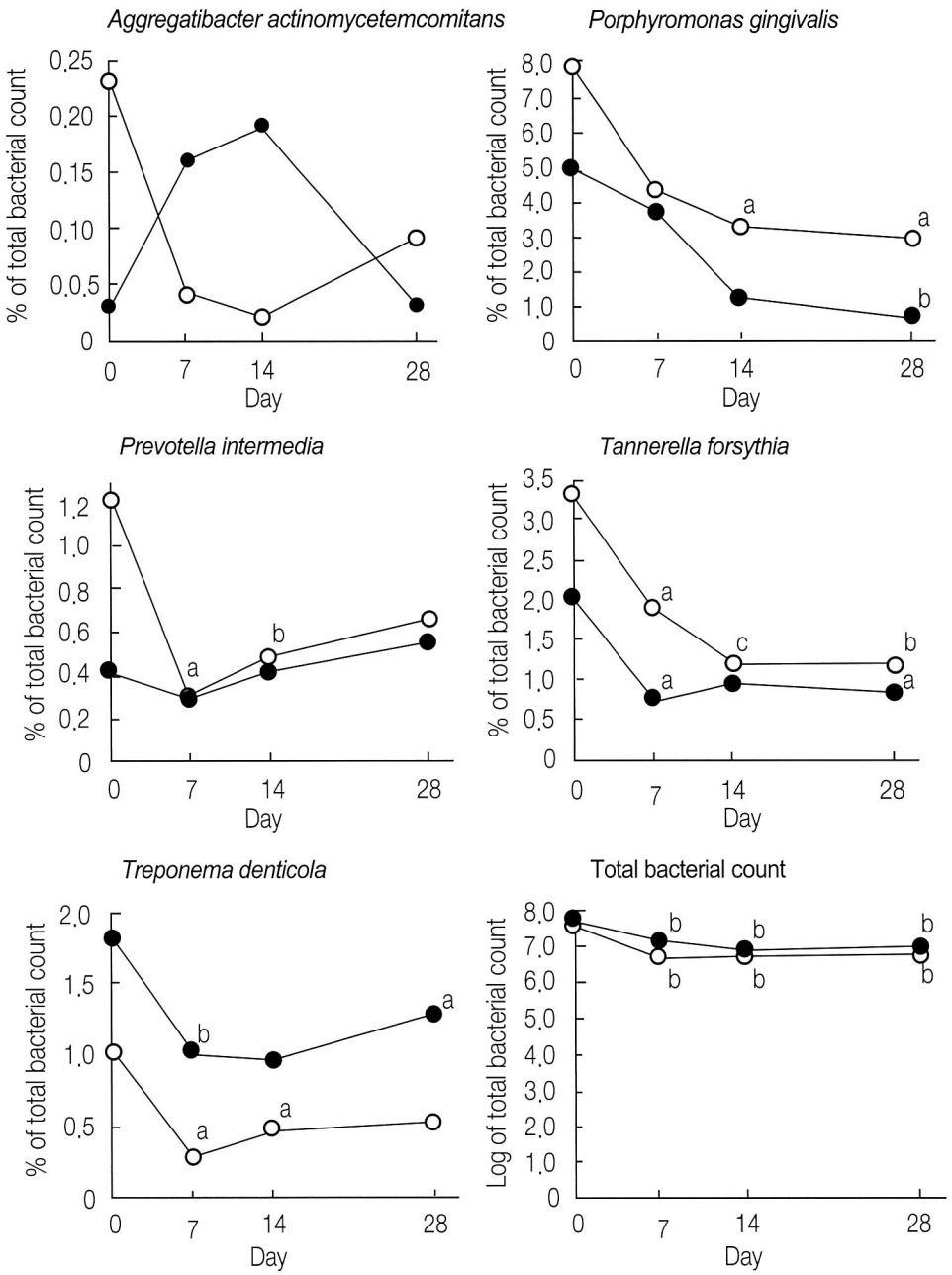

Figure 1. *Microbiological results for the professional toothbrushing group (open circle) and one-stage full-mouth disinfection group (closed circle) based on real-time PCR. Data are expressed as mean values (n=24 teeth). Data for each bacterial species are expressed as a mean percentage of the total bacterial count and data for the total bacterial counts in log of the mean value.* $^{a}p<0.05$, $^{b}p<0.01$, $^{c}p<0.001$ *Wilcoxon signed-ranks test, compared to Day 0.*

supragingival plaque control alone was inferior to supra- and subgingival therapy in improvement of clinical and microbiological parameters of periodontitis (Loesche 1968, Loos *et al.* 1988). In the abovementioned studies, toothbrushing was performed by the patients themselves rather than professionally. The average time that patients brush at a dental clinic has been reported to be only 56.7 sec for the whole mouth (range 15 to 155 sec) (Westfelt *et al.* 1998). In the present study, professional toothbrushing using the Toothpick method was performed for about 15 min. To enhance proliferation of gingival cells and collagen synthesis, the recommended optimum duration of mechanical stimulation by toothbrushing is 10 - 20 sec per tooth surface (Emling *et al.* 1981, Tomofuji *et al.* 2002). This is certainly longer than the time patients generally spend brushing at home, suggesting that professional toothbrushing may be a more effective means to treat periodontal disease.

The change of bacterial composition in the professional toothbrushing group, which was confirmed by real-time PCR and PCR-DGGE analyses, may be ascribed to enhancement of

defence mechanisms against infection via mechanical stimulation of the gingiva. The epithelial surface of the periodontal pocket is often ulcerated (Tomofuji *et al.* 2003), and these weakened areas bleed, supporting the growth of several anaerobic periodontal pathogens such as *P. gingivalis* (Carranza & Camargo 2002), *T. forsythia* (Kesavalu *et al.* 2003) and *T. denticola* (Wyss *et al.* 1993), which require blood components in growth medium. The effects of mechanical stimulation include the promotion of cell proliferation in the junctional epithelium of the periodontal pocket (Yamamoto *et al.* 2004) and vascular endothelial cells in gingiva (Sakamoto *et al.* 2003). Accelerated repair of ulcerated epithelium by mechanical stimulation might stop the bleeding and reduce haemoglobin-required bacterial growth. This explanation was supported by the present temporal relation, where in the toothbrushing group the decrease in bleeding on probing preceded the reduction in *P. gingivalis* count (Fig. 1).

Limitations of this study include the small sample size. However, the results suggest that changes in host immune mechanism affect the ecology of subgingival microbiota. Further studies are required to confirm the findings of this study.

In conclusion, professional toothbrushing was more effective in improving gingival bleeding than one-stage full-mouth disinfection. However, there was no significant difference in the microbiological and clinical parameters of periodontitis between professional toothbrushing and one-stage full-mouth disinfection. Professional toothbrushing might thus hold an important place in periodontal treatment in the future.

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Original Article

The relationship between the number of the residual tooth and the masticatory function

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Abstract

This study set out to examine the solidity of major foods consumed by the elderly in Korea, identify the kinds of food according to categories, calculate correlations between the food and number of remaining teeth, and provide some guidelines to maintain, manage, and preserve the healthy state of natural teeth. For this study, dentists and dental hygienists conducted oral exams and interviews on 586 male and female senior citizens aged 60 and above to investigate and analyze their chewing ability according to the types of food chewed only with natural teeth with or without dentures. Collected data were analyzed with SPSS WIN 14.0 program. Correlations between the number of the subjects remaining teeth and their chewing ability were examined with coefficients of correlations and regression analysis with the results as follows:

Significant correlations between the subjects chewing ability and oral state in tartar, teeth mobility, filling points, decay experience, number of extracted permanent teeth, number of implants, number of natural teeth, age, hypertension, diabetes, other diseases, and smoking were found($p<0.05$).

The elder the subject, the less chewing ability they had, and the more natural teeth they had, the more chewing ability they demonstrated.

The chewing ability = 79.417-(age x 2.281) + 3.813 x number of teeth($p<0.05$).

The subjects chewing ability was highly related to the number of their remaining teeth and was 3.817 x natural teeth +58.694($p<0.05$).

It was found that the elderly of Korea needed 18 remaining natural teeth to chew rice and kimchi, their principle foods, very well, 12 to chew well, at least 18 to chew meat well, and 25 to chew meat very well.

Key words: natural tooth, chewing ability, Korea food

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Introduction

Today Korea has entered the stage of an aging society. In 1980, those who were aged 65 or older accounted for 3.95% of its entire population, and the percentage went up to 7.2% in 2000. It s estimated to enter the stage of an aged society with 14.4% by 2019 and the stage of a super-aged society by 2026[1)]. At the second annual conference of the International Association of Gerontology, the process of human aging was defined involving interactions among physiological, physical, and psychological environments and behavioral changes. Kim characterized the aging process as a period in which changes to the physical and physiological changes would cause various kinds of disorders and weakened strength, alter the cerebrovascular, respiratory, and musculoskeletal system[2)].

With the recognized importance of teeth and chewing ability, Korea has undertaken measures recently to take care of natural teeth .Japan initiated the 8020 Movement about 20 years ago. Under the goal of maintaining 20 natural teeth till the age of 80, the Department of Oral Health at the Ministry of Health and Welfare has set an ultimate objective of better oral health for the elderly. Under the guidance of the Japan Dental Association, each local government has installed a devoted department to

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the movement and developed and implemented required projects for oral health. In Korea the focus has been on dental treatment to replace natural teeth instead of maintaining them and further on treatment rather than prevention. While there has been a lot of research on chewing ability in other countries, Korea has conducted little research on the importance of natural teeth.

Although there is a definite need for national policies for the elderly welfare, the reality just depicts a grim picture. Lee et al. maintained that a good health and nutrition state should be essential to the high quality of life of the elderly and that nationwide efforts should be made to improve their health and nutritional state[3].

Thus, in this study, research was conducted on examining the number of natural teeth of elderly Koreans aged 60 and above, the solidity of major foods consumed, and the kinds of chewable food in each food category. Correlations between the two were calculated as an indicator to maintain, manage, and preserve the healthy state of natural teeth and pursuing a goal of making a contribution to the better oral health of the nation.

Subjects and Methodology

Subjects

Participants were selected from 30 big cities, medium- and small-sized cities, eups and myeons in accordance with the population ratio of the samples used during the Korean National Oral Health Survey in 2003. From June to October 2008 subjects, elderly aged 60 and above, were surveyed via the dental hospital, dental clinics, and public health centers. After excluding 14 of the total 600 whose answers were insincere or wrongfully entered, total 586 questionnaires were used in the subsequent analysis.

Methodology

Survey form included an oral exam, exam of the entire body and more, the chewing food state with natural teeth, chewing food state with or without a denture, and a consent form. Based on literature on the frequency of food consumption in the Korean National Health and Nutrition Survey in 2005, ten food items were selected and investigations on the elderly chewing ability were put to solidity test and ranked accordingly. The ten foods included those that were in the No. 1 and 2 spot according to frequency of consumption in each category with these being tofu, rice, whites of boiled egg, biscuits, cabbage kimchi, meat, cubed white radish kimchi, apples, beans, dried squid, and candy. All subjects were tested in tensile strength with the compressive/tensile strength tester already and classified them by degree 1 to 10.

An oral exam was conducted in terms of decay, filling, extraction, implants, dentures, teeth mobility and tartar. In the exam of the entire body and more, investigations on oral disease, systematic disease, temporomandibular joint disease, smoking and drinking. were conducted. Dentists and dental hygienists undertook the exams and administered the tests and interviews. The subjects were offered enough information and explanation prior to exams for a full understanding of what would be undertaken. A questionnaire questionnaire was distributed and sent only to those places that agreed to participate in this study via e-mail, mail, fax, and personal visit (see Appendix).

Organization and evaluation of the chewing ability note

Chewing ability note was composed of three parts, first, the oral test included decay, filling, extracted teeth, implants, and a dentures. As for teeth mobility, only three points were recorded that involved 1mm or more horizontal mobility and vertical mobility to require an extraction. As for tartar, only a three-point tartar that covered all the three sections of a crown and the three-point ring-shaped subgingival calculus were recorded. As for decay, a scale of 1 to 4 was applied; C1 refers to decay limited to enamel, C2 to dentin, C3 to the tooth pulp, and C4 to the tooth root. The decay of C4 category requires an extraction due to an abscess. As for filling, a scale of F1 to F5 was employed; F1 refers to one filling side, F2 two, F3 three, F4 four, and F5 a crown. Using clone s First Moral Health Scale, 0.5 points were added for each stage. Thus the lowest points were 0.5 and the highest were 2.5. The combined points of decay and filling made the decay experience points. Since the study was to investigate the elderly chewing ability according to the number of their remaining teeth, set the standard a total standard of 32 teeth were set including the wisdom teeth. The number of remaining teeth was calculated by subtracting all the teeth that were extracted and not recovered, artificial teeth and implants. Secondly, subjects were examined for systematic diseases, hereditary oral disease, other kinds of oral disease, and temporomandibular joint disease in the exam of the entire body and more. And the smoking and drinking points were calculated by multiplying the amount of

Table 1. *Gender and age distribution of subjects*

Gender	Male	Female	Total(%)
60-69	156(26.6%)	168(28.7%)	324(55.3%)
70-79	90(15.4%)	113(19.2%)	203(34.6%)
80 or older	21(3.7%)	38(6.4%)	59(10.1%)
Total	267(45.7%)	319(54.3%)	586(100%)

Table 2. *Regional distribution of subjects (%)*

Region	Seoul	Gyeonggi	Gangwon	Chungcheong	Gyeongsang	Jeolla	Jeju	Total
In 60s	67	99	24	41	49	28	16	324
	(11.4)	(16.9)	(4.1)	(7.0)	(8.4)	(4.8)	(2.7)	(55.3)
In 70s	46	45	36	18	36	18	4	203
	(7.8)	(7.7)	(6.1)	(3.1)	(6.1)	(3.1)	(0.7)	(34.6)
In 80s	8	19	11	3	9	7	2	59
	(1.4)	(3.2)	(1.9)	(0.5)	(1.5)	(1.3)	(0.3)	(10.1)
Total	121	163	71	62	94	53	22	586
	(20.6)	(27.8)	(12.1)	(10.6)	(18.0)	(9.2)	(3.7)	(100)

Table 3. *Solidity and chewing ability points of each food*

Solidity ranks	Foods	Compression strength (gf)	Very good (5)	Good (4)	Average (3)	Poor (2)	Very poor (1)
0	Water/milk	0	0	0	0	0	0
1	Tofu	192.3	5	4	3	2	1
2	Rice	204.3	10	8	6	4	2
3	Whites of boiled egg	868.7	15	12	9	6	3
4	Biscuits/cabbage kimchi	1089.0 /1416.0	20	16	12	8	4
5	Meat(beef)	1843.0	25	20	15	10	5
6	Cubed white-radish kimchi	2790.0	30	24	18	12	6
7	Apples	3083.2	35	28	21	14	7
8	Peanuts/beans	4360.0 /4820.0	40	32	24	16	8
9	Dried squid	11500.0	45	36	27	18	9
10	Candy	25210.0	50	40	30	20	10

Total points: 275

smoking by the amount of drinking.

Thirdly, 0 to ten points were given to the 0 to ten stages of foods[4]. Chewing degree was divided into five steps with five points reserved for very good and one point for very poor. The subjects chewing ability was recorded when those with only natural teeth chewed foods and those who had a dentures chewed their foods with and without a dentures.

Statistical analysis

Using the SPSS(Statistical Package for the Social Science) WIN 14.0 program, the collected data were analyzed in frequency and percentage for general characteristics. Correlation analysis was carried out to calculate the relationships between natural teeth and chewing ability. t-test was conducted to understand the relationships between systematic disease and chewing ability. Regression analysis was conducted to examine the factors related to chewing ability.

Results

Gender and age distribution of subjects

Study involved a nationwide survey among the male and female elderly aged 60 or older. As shown in with Table 1, there were 156(26.6%) elderly males aged 60 or older and 168(28.7%) elderly females aged 60 or older; in the seventies group, male and female subjects were 90(15.4%) and 113(19.2%), respectively, and in the eighties group, male and female subjects were 21(3.7%) and 38(6.4%), respectively. A total of 586 were analyzed in this study.

Regional distribution of subjects

Return rate of questionnaires among subjects was proportionate to population ratio, Seoul(121) and Gyeonggi(163) accounted for 48.4%, Gyeongsang

Table 4. *Correlation coefficient between chewing ability of natural teeth and oral condition*

Factor	r	p
Tartar	.124**	.003
Teeth mobility	-.116**	.005
Decay points	.018	.655
Filling points	.112**	.007
Decay experience points	.101*	.014
Prosthetic points	.060	.145
Abutment prosthetics	.078	.058
pontic	.024	.566
Number of extracted teeth not recovered	-.640**	.000
Number of implants	.098*	.017
Number of natural teeth	.642**	.000

**P<0.01 *P<0.05

Table 5. *Correlations between smoking and drinking and chewing ability*

Factor	r	p
Smoking	-.086*	.037
Drinking	.053	.197

**P<0.01 *P<0.05

Table 6. *t-test between chewing ability and the overall body state*

		N	Mean	SD	t	p
Hypertension	No	396	133.79	74.18	4.13**	0.000
	Yes	190	107.63	66.08		
Diabetes	No	488	128.59	72.99	2.45*	0.015
	Yes	98	108.96	68.91		
Thyroid gland	No	578	125.88	78.64	0.54	0.586
	Yes	6	109.66	48.57		
Others	No	567	126.84	72.76	2.81**	0.005
	Yes	19	79.47	52.13		
Dry mouth	No	583	125.52	72.75	0.98	0.324
	Yes	3	84.00	22.51		

**P<0.01 *P<0.05

(94) 18.0%, Gangwon(71) 12.1%, Chungcheong(62) 10.6%, Jeolla(53) 9.2%, and Jeju(22) 3.7% respectively.

Table 3 shows the chewing ability results measured with a tensile/compression testing machine.

Tofu was ranked at the lowest Stage 1 with 192.3g; meat at the middle Stage with 51843.0 gf; Candy at the highest Stage 10 with 25210.0 gf. With these results, stages from 0 to 10 were determined. The lowest point was 0 and the highest was 10. Thus tofu received 1, rice 2, whites of boiled egg 3, biscuits/cabbage kimchi 4, meat 5, cubed white-radish kimchi 6, apples 7, peanuts(beans) 8, dried squid 9, and candy 10. The chewing ability was put on a scale of five points. Five points were given to very good, four to good, three to average, two to poor, and one to very poor. The food chewing ability was the multiplication of each food and its chewing ability. When a subject can chew all the foods very well, he or she will receive a total 275 calculated as $(1\times5)+(2\times5)+(3\times5)+(4\times5)+(5\times5)+(6\times5)+(7\times5)+(8\times5)+(9\times5)+(10\times5)$; if he or she finds it very difficult to chew all the foods, he or she will receive the lowest total of 55 calculated as $(1\times1)+(2\times1)+(3\times1)+(4\times1)+(5\times1)+(6\times1)+(7\times1)+(8\times1)+(9\times1)+(10\times1)$.

Correlation coefficient between chewing ability of natural teeth and oral condition

Table 4 shows the relationships between the chewing ability of natural teeth and oral condition. The chewing ability was significantly related to tartar, teeth mobility, filling points, decay experience points, number of extracted teeth not recovered, number of implants, and number of natural teeth ($p<0.05$). The greatest significance was observed in the number of natural teeth and extracted teeth not

Table 7. *Factors related to chewing ability 1*

1. Model-When two factors of age and number of natural teeth work	Unstandardized coefficient	
	B	Standard error
(Constant)	79.417	24.403
Age	-2.281	0.326
Number of natural teeth	3.813	0.201

Table 8. *Factors related to chewing ability 2*

2. Model-When only natural teeth work	Unstandardized coefficient	
	B	Standard error
(Constant)	58.694	4.050
Number of natural teeth	3.817	0.187

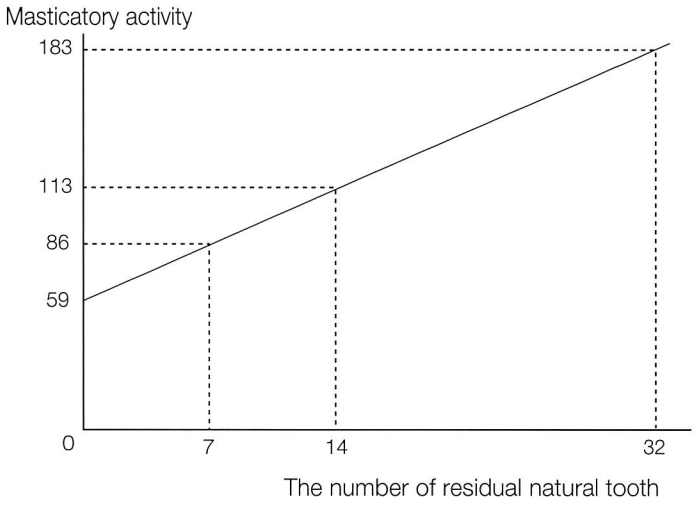

Figure 1. *Graph of the relations between the number of natural teeth and the chewing ability*

recovered ($p<0.01$), which were followed by tartar, teeth mobility, and filling points ($p<0.05$).

Correlations between smoking and drinking and chewing ability

Table 5 shows the correlations between smoking and drinking and chewing ability. While smoking was significantly correlated to chewing ability ($p<0.05$), drinking was not.

t-test between chewing ability and the overall body state

As seen in Table 6, chewing ability was significantly correlated to hypertension, diabetes and others with the greatest significance found with hypertension ($p<0.05$), which was followed by others and diabetes, respectively.

Factors related to chewing ability (Regression analysis 1)

When two of the factors related to chewing ability, which were age and number of natural teeth, came into play, results are shown in Table 7. The regression equation is as follows:

Regression equation of chewing ability
=79.417-(age x 2.281)+3.813 x number of natural teeth($p<0.05$)

Factors related to chewing ability (Regression analysis 2)

When only natural teeth of the factors related to the chewing ability came into play, the results are shown in Table 8. The regression equation is as follows:

Regression equation of the chewing ability
=3.817 x natural teeth + 58.694 ($p<0.05$)

Regression analysis results of relations between the number of natural teeth and chewing ability

In short, if a subject has no natural teeth left as shown in in Figure 1, his or her chewing ability points will be 59; has seven natural teeth left, the points will be 86; 14, the points will be 113; and 32, the points will be 183, respectively. The more natural teeth they have, the greater chewing ability they have.

Table 9. *Differences in chewing ability with and without dentures*

N	With denture	Without denture
272	139.92 – 48.32**	69.05 – 31.182**

**P<0.01

Food / Chewing ability points	1 Tofu	2 Rice	3 White of a boiled egg	4 Cabbage Kim chi	5 Meet(beef)	6 Cubed white-radish kimchi	7 Apple	8 Peanuts	9 Dried squid	10 Candy
185	5	5	5	5	5	5	4	3	2	1
155	5	5	5	5	5	4	3	2	1	1
129	5	5	5	5	4	3	2	1	1	1
107	5	5	5	4	3	2	1	1	1	1
89	5	5	4	3	2	1	1	1	1	1
75	5	4	3	2	1	1	1	1	1	1
65	4	3	2	1	1	1	1	1	1	1
59	3	2	1	1	1	1	1	1	1	1
56	2	1	1	1	1	1	1	1	1	1
55	1	1	1	1	1	1	1	1	1	1

Tooth	Chewing ability points
32	183
31	179
30	175
29	171
28	167
27	163
26	159
25	156
24	152
23	148
22	144
21	140
20	136
19	132
18	128
17	125
16	121
15	117
14	113
13	109
12	105
11	101
10	97
9	94
8	90
7	86
6	82
5	78
4	74
3	70
2	66
1	63
0	59

Figure 2. *Connections between the chewing ability and number of teeth*

Differences in chewing ability with and without dentures

As seen in Table 9, chewing ability increased with dentures and decreased without, and the difference between points was significant ($p<0.01$). Also significant was the difference in the chewing ability with and without denture($p<0.01$).

Regression analysis results of total table.

As seen in Figure 2 and 3, five points were given to very good chewing ability and one to very poor chewing ability.

One can chew meat very well with 25 remaining teeth and 155 chewing points and well with 18; and one can chew cabbage kimchi very well with 18 remaining teeth and 129 chewing points and well with 12, respectively.

One can chew whites of boiled egg very well with 12 remaining teeth and 107 chewing points and well with 8; one can chew rice very well with 8 remaining teeth and 89 chewing points and well with 4; and one chew tofu very well with 4 remaining teeth and 75 chewing points and well with 3. In conclusion, one needs 18 remaining natural teeth in order to chew rice and kimchi, the staple foods of Koreans, very well and 12 well, respectively.

Discussion

As society develops and the quality of life improves, the life span of humans is naturally

Natural Tooth	very well						well	nomal	poor	very poor
32	Tofu	Rice	Boiled egg	Cabbage Kimchi	Meat (beef)	Radish Kimchi	Apple	Peanuts	Dried squid	Candy
31	Tofu	Rice	Boiled egg	Cabbage Kimchi	Meat (beef)	Radish Kimchi	Apple	Peanuts	Dried squid	Candy
30	Tofu	Rice	Boiled egg	Cabbage Kimchi	Meat (beef)	Radish Kimchi	Apple	Peanuts	Dried squid	Candy
29	Tofu	Rice	Boiled egg	Cabbage Kimchi	Meat (beef)	Radish Kimchi	Apple	Peanuts	Dried squid	Candy
28	Tofu	Rice	Boiled egg	Cabbage Kimchi	Meat (beef)	Radish Kimchi	Apple	Peanuts	Dried squid	Candy
27	Tofu	Rice	Boiled egg	Cabbage Kimchi	Meat (beef)	Radish Kimchi	Apple	Peanuts	Dried squid	Candy
26	Tofu	Rice	Boiled egg	Cabbage Kimchi	Meat (beef)	Radish Kimchi	Apple	Peanuts	Dried squid	Candy
25	Tofu	Rice	Boiled egg	Cabbage Kimchi	Meat (beef)	Radish Kimchi	Apple	Peanuts	Dried squid	Candy
24	Tofu	Rice	Boiled egg	Cabbage Kimchi	Meat (beef)	Radish Kimchi	Apple	Peanuts	Dried squid	Candy
23	Tofu	Rice	Boiled egg	Cabbage Kimchi	Meat (beef)	Radish Kimchi	Apple	Peanuts	Dried squid	Candy
22	Tofu	Rice	Boiled egg	Cabbage Kimchi	Meat (beef)	Radish Kimchi	Apple	Peanuts	Dried squid	Candy
21	Tofu	Rice	Boiled egg	Cabbage Kimchi	Meat (beef)	Radish Kimchi	Apple	Peanuts	Dried squid	Candy
20	Tofu	Rice	Boiled egg	Cabbage Kimchi	Meat (beef)	Radish Kimchi	Apple	Peanuts	Dried squid	Candy
19	Tofu	Rice	Boiled egg	Cabbage Kimchi	Meat (beef)	Radish Kimchi	Apple	Peanuts	Dried squid	Candy
18	Tofu	Rice	Boiled egg	Cabbage Kimchi	Meat (beef)	Radish Kimchi	Apple	Peanuts	Dried squid	Candy
17	Tofu	Rice	Boiled egg	Cabbage Kimchi	Meat (beef)	Radish Kimchi	Apple	Peanuts	Dried squid	Candy
16	Tofu	Rice	Boiled egg	Cabbage Kimchi	Meat (beef)	Radish Kimchi	Apple	Peanuts	Dried squid	Candy
15	Tofu	Rice	Boiled egg	Cabbage Kimchi	Meat (beef)	Radish Kimchi	Apple	Peanuts	Dried squid	Candy
14	Tofu	Rice	Boiled egg	Cabbage Kimchi	Meat (beef)	Radish Kimchi	Apple	Peanuts	Dried squid	Candy
13	Tofu	Rice	Boiled egg	Cabbage Kimchi	Meat (beef)	Radish Kimchi	Apple	Peanuts	Dried squid	Candy
12	Tofu	Rice	Boiled egg	Cabbage Kimchi	Meat (beef)	Radish Kimchi	Apple	Peanuts	Dried squid	Candy
11	Tofu	Rice	Boiled egg	Cabbage Kimchi	Meat (beef)	Radish Kimchi	Apple	Peanuts	Dried squid	Candy
10	Tofu	Rice	Boiled egg	Cabbage Kimchi	Meat (beef)	Radish Kimchi	Apple	Peanuts	Dried squid	Candy
9	Tofu	Rice	Boiled egg	Cabbage Kimchi	Meat (beef)	Radish Kimchi	Apple	Peanuts	Dried squid	Candy
8	Tofu	Rice	Boiled egg	Cabbage Kimchi	Meat (beef)	Radish Kimchi	Apple	Peanuts	Dried squid	Candy
7	Tofu	Rice	Boiled egg	Cabbage Kimchi	Meat (beef)	Radish Kimchi	Apple	Peanuts	Dried squid	Candy
6	Tofu	Rice	Boiled egg	Cabbage Kimchi	Meat (beef)	Radish Kimchi	Apple	Peanuts	Dried squid	Candy
5	Tofu	Rice	Boiled egg	Cabbage Kimchi	Meat (beef)	Radish Kimchi	Apple	Peanuts	Dried squid	Candy
4	Tofu	Rice	Boiled egg	Cabbage Kimchi	Meat (beef)	Radish Kimchi	Apple	Peanuts	Dried squid	Candy
3	Tofu	Rice	Boiled egg	Cabbage Kimchi	Meat (beef)	Radish Kimchi	Apple	Peanuts	Dried squid	Candy
2	Tofu	Rice	Boiled egg	Cabbage Kimchi	Meat (beef)	Radish Kimchi	Apple	Peanuts	Dried squid	Candy
1	Tofu	Rice	Boiled egg	Cabbage Kimchi	Meat (beef)	Radish Kimchi	Apple	Peanuts	Dried squid	Candy
0	Tofu	Rice	Boiled egg	Cabbage Kimchi	Meat (beef)	Radish Kimchi	Apple	Peanuts	Dried squid	Candy

Very well well Normal Poor Very Poor

Figure 3. *Chart of elderly s chewing ability according to number of remaining teeth*

prolonged. People make every efforts to increase their standard of living in the course of their lives. There is no one that just wants a longer life in the increasingly aging society. All of us want an active and happy life as long as we live, and there are many conditions to be met to achieve this. The most important are a healthy body and soul. Health is the most critical across all the age groups, but the elderly especially find it more valuable and significant than other age groups. The elderly go through many physical, psychological and social changes, feel intensive basic desires for everything, and have a great interest in health[5].

In the old age populations oral health is closely related to overall health. The mouth is the starting point of the digestive system and performs diverse

functions including respiration, pronunciation, and sensation. Not only does it play a critical role in maintaining life, but also it has much to do with pleasures of life such as eating and speaking. Thus when one suffers pain in their teeth or oral mucosa due to decay or infection or has limited mobility in the lower jaw due to a disability caused by an accident or operation, he or she has enormous mental and physical inconveniences[6]. Miura et al. reported that there are close relationships between chewing ability and quality of life as well as overall health[7].

The elderly, compared to other age groups, lose many teeth. As they grow older, they have less remaining natural teeth and more extracted permanent teeth[8]. But there are often cases in which they regard having a tooth extracted, losing one, or suffering from physiological phenomena by periodontal disease as a natural process of getting old, give up seeking dental treatment[9], and endure inconvenience. As a result, whatever oral disease they have eventually worsens and causes a problem to the entire body. When a tooth is extracted or lost, the sensory receptors on the periodontal membrane are removed as well. As a result, they lose the senses of food touching teeth and controlling chewing ability reflectively and have difficulty with regulating chewing ability[10].

Watanabe maintained that loss of a tooth has much effect on an individual s social roles as well as food consumption, communication, taste, pronunciation, and appearance[11]. These days the rapid advancement in artificial teeth or implants has been reported in the mass media and press. Thus many people believe that they can have new teeth through implants even though they lose their natural teeth. But that s false and misleading, of course, implants generate better results than no teeth at all, but this is not the only aspect that one should see. It s important to educate people about the differences between natural teeth and implants and the importance of the former. The most important thing is to chew food with natural teeth, which should be promoted among the nation.

Thus the study set out to examine the solidity of major foods consumed by the elderly in Korea, identify the kinds of food according to the categories, calculate correlations between food and the number of their remaining teeth, and provide some guidelines necessary to maintain, manage, and protect the healthy state of their natural teeth. A total 586 of 600 questionnaires were returned. The high return rate can be attributed to the fact that participants and/or subjects were personally asked by dentists and dental hygienists in the field to participate and given thorough explanations about what was being asked of them. In addition throughout this study the methods and methodology were explained either through personal communication by phone, mail, and subsequent checking was undertaken on the progress at certain intervals. Also, a variety of means were undertaken to collect the questionnaires to include but not limited to e-mail, fax, mail and personal collection. Regarding the general characteristics of the subjects, a total of 324 were in their sixties (156 males and 168 females), this group accounting for the most; and 203 and 59 in their seventies and eighties, respectively rounded out the total number. The age distribution can be explained by the fact that the sixties group was more active with social activities than the other age groups.

The age distribution can be explained by the fact that the sixties group was more active with social activities than the other age groups.

As for the relationships between chewing ability of natural teeth and oral condition, there were significant relationships according to tartar, teeth mobility, filling points, decay experience points, number of extracted teeth not recovered, number of implants, and number of natural teeth($p<0.05$). The biggest significance was found in the number of natural teeth and the number of extracted teeth not recovered ($p<0.01$), which were followed by tartar, teeth mobility, and filling points in the order ($p<0.05$). Only a three-point tartar that covered all three sections of a crown and the three-point ring-shaped subgingival calculus were recorded because three points mean there are many periodontal pockets, the periodontal tissues have many problems due to tartar, and there is a chewing issue. For teeth mobility, only three points were recorded that involved 1mm or more horizontal mobility and vertical mobility to require an extraction because three points suggest a chewing issue.

There was significant correlation between chewing ability and the overall state according to hypertension, diabetes and others. The greatest significance was observed in hypertension ($p<0.05$), and there were diverse systematic diseases found since the subjects were elderly. The most frequent hypertension and diabetes are common in the elderly. Hypertension occurs when an adult s diastolic pressure exceeds 140/90mmHg in a resting supine position. As people grow older, blood pressure

increases. Diabetes refers to a prolonged state of high blood sugar and the accompanied metabolic disorder due to absolute or relative lack of insulin secretion or the decrease of insulin s biological effects. It is accompanied by a disorder with the small and large blood vessels[12]. Other diseases include dementia, blood disease and liver disease. The results indicate that once the elderly contract these types of disease, they cannot help but to focus on treating itand, thus are less concerned with their oral health. Since these diseases affect the entire body, it s natural to assume close relationship between oral health and any disease. Diabetes, for instance, accompanies complications, which make it more dreadful. From a dental aspect, diabetes often absorbs alveolar bone and postpones injury healing. There are many studies that report on the influences of those systematic diseases on chewing ability. Thus it s important to remind the elderly the importance of overall health along with oral health. As for smoking and drinking, smoking was significantly related to chewing ability($p<0.05$).

There are many reports on this close relationship with periodontal and bone loss and on the connection between a smoker s daily smoking and entire smoking period and his or her periodontal loss[13-16]. It s also well known that a smoker is prone to infection with microbial pathogenic organs and doesn t respond to the treatment of antibiotics well. Everyone s knowledgable of the fact that smoking causes many diseases and does the body no good. But there are still many people who smoke; why is that? One reason can be habit. But in the case of Korea, it is a , it is a custom and a virtue among men to smoke cigarettes and/or drink alcohol. There is also strange logic that smoking and drinking earn one better points socially and career wise in their life. In fact, Korean society is laced with such logic[17]. Such a social atmosphere has caused smoking to be accumulated for ages, which may cause a problem to the chewing ability of the elderly. Despite no significance finding, drinking is also a formidable factor in overall physical and oral health. Since smoking and drinking worsen the malnutrition of the elderly and increase the risk of a chronic disease, it is of great necessity to educate the elderly on how to curb these habits[18].

When the two factors related to chewing ability, which are age and the number of natural teeth, came into play, a regression equation was obtained:

chewing ability=79.417-(age× 2.281)+3.813 × number of teeth($p<0.05$).

When only natural teeth came into play,

chewing ability=3.817× natural teeth+58.694 ($p<0.05$).

What was undertaken in this study was to show and prove how natural teeth affected chewing ability. The results indicate that the more natural teeth they had, the better chewing ability they demonstrated. In short, they need 155 points and 25 remaining teeth to chew meat very well, 129 points and 18 remaining teeth for cabbage kimchi, 107 points and 12 remaining teeth for the whites of a boiled eggs, and 75 points and 4 remaining teeth for tofu. In conclusion, Koreans need 18 remaining teeth in order to chew rice and kimchi, their staple foods, well.

Goto reported that there was a huge difference in meals and satisfaction with chewing between 20 and 2 remaining natural teeth. Hirano showed that the more remaining teeth they had and the less tooth loss they suffered, the better their chewing ability was[8]. Those results seem so natural. There is an old saying in Korea, The gum can come into play when you have no teeth. Although it doesn t strictly apply to teeth and chewing, perhaps this saying should be changed to No gum can replace teeth In order to chew, man needs saliva secreted in the mouth, and saliva protects the mouth in various ways. In particular, it removes sugar in the mouth, suppresses the growth of acidophil oral microorganisms, plays an important role in increasing PH in the mouth[19], and gets rid of various toxins. Thus a good chewing habit can protect the mouth from various harmful matters and prevent cancer[20], which means that chewing ability is extremely important to the health of the elderly.

The test results also show that the subjects demonstrated better chewing ability with dentures than without. Dentures are under the influence of various factors including quality, oral state, mental and psychological factor, socioeconomic factor, relationships between patient and dentist, edentulous period, former experience with dentures, and recovery state of opposing teeth arches. It s also reported that the patient s feeling and emotions that are subject to constant changes can also play a part[21]. Considering all those, it s for certain that a dentures increases chewing ability significantly and they can be helpful in the absence of natural teeth.

According to the Korean National Oral Health Survey in 2003, the elderly population aged 65-75 had 12.1 permanent teeth left, which was a huge drop from 16.3 in 2000[22]. With better times and

improved medical technology this should guarantee more teeth remain in the elderly in the future. Then what can explain the drop? Is it partly because they have promoted alternatives to natural teeth too much through advertising? In Korea research has focused heavily on implants and dentures rather than how to preserve natural teeth. The current dentistry community in Korea evolves around treatment. If it s true that implants or dentures can replace natural teeth perfectly, no one will bother to make efforts to preserve natural teeth and thus easily choose such method over preventing natural teeth to decay. But if this is the case, the elderly with economic difficulties will continue to suffer from poor chewing ability. But is it really true that implants or dentures can perform all the functions of natural teeth? Receiving promotional highlight implants are regarded as a revolution in dentistry. But natural teeth can regulate chewing ability with the periodontal ligaments, which cannot be done by implants. We, however, should not underestimate implants. The success rate of implants is not 100% and it is reported they can replace natural teeth in terms of prognosis[23]. Although implants are a boon to those who have lost natural teeth, it is not desirable to perceive them as the only solution. Therefore, it must be stressed here that taking preventive measures before removing a tooth is more valuable and important, which should be the aim of education to the entire nation. In addition, the dentistry community should not focus on implants or pricy design dentures to earn high revenue.

Eating foods just to feel full is the custom of old age. Today chewing represents much more including enjoying food, tasting food, and keeping one s health. This is one of the most essential functions of life. The elderly have no reason to give up leading a satisfying life or enjoying foods just because they have lost a few teeth. Rather they should pay more attention to eating for good health. Japan started the 8020 Movement, 20 years earlier than Korea. In Korea, they usually recommend alternatives to loss of teeth and have conducted little researches on how to take care of and preserve natural teeth. It is a more healthy movement to recommend preventive measures than treatment. As for the limitations of this study, a more nationwide occlusal evaluation was necessary. With a more objective testing machine, more accurate measurements would have been possible. Also, it would have been better in terms of results if the subjects had been evenly distributed among the ages of those in their sixties, seventies and eighties groups. In the future more objective supplements and systematic research on these aspects are something to be dealt with.

The dentistry community, the public, and the government should work together and take active interest in helping the elderly maintain and manage their natural teeth in today s aging society. There are also enough grounds to take interest in oral health and investigate how the preservation of natural teeth affects chewing ability, nutritional state, and overall health and engage in active promotional activities for the elderly. In planning a program for public health and medicine and the better health of the elderly, it s important to induce more interest in the dental domain and encourage a primary consideration of it. When those efforts are implemented, greater interest in and better results for oral health will be guaranteed.

Conclusion

There were significant relationships between chewing ability and oral condition according to tartar, teeth mobility, filling points, decay experience points, number of extracted teeth, number of implants, number of natural teeth, age, hypertension, diabetes, other diseases and smoking($p<0.05$).

The regression analysis results suggest that the older they became, the less chewing ability they demonstrated and that the more natural teeth they had, the greater chewing ability they demonstrated. The regression equation was chewing ability= 79.417-(age × 2.281)+3.813 × number of teeth ($p<0.05$).

According to the regression analysis results, the chewing ability points were the most related to the number of natural teeth, and the regression equation was chewing ability=3.817 × natural teeth+58.694 ($p<0.05$).

In short, the elderly of Korea need 18 remaining natural teeth in order to chew rice and kimchi, the staple foods, very well and 12 in order to chew them well; and they need 25 remaining natural teeth in order to chew meat very well and at least 18 in order to chew meat well.

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Original Article

Clinical Study about Oral Environmental Improvements Using Dental-washing solutions including IPMP or GK2

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Abstract

The objective of this study was to determine the oral environment improvement effects and safety of IPMP/GK2 mouthwash.

This double-blind study was carried out in 30 healthy Korean adults to evaluate the oral environment improvement effects and safety after application of the mouthwash containing IPMP and/or GK2 in given amounts over 4 cycles (2 weeks of carry-out effect period, and 2 weeks of IPMP/GK2 mouthwash use, total 4 weeks/cycle, group X 4 groups), compared with the placebo group.

After 4 cycles, IPMP/GK2 groups showed statistically significant reductions in oral environmental indexes, such as halitosis, GCF, periodontal pocket depth and plaque index, when compared to the placebo group. In particular, there were statistically significant reductions in the total counts of *S. mutans*, *S. sobrinus*, *P. gingivalis* before and after application of the mouthwash containing IPMP 0.02% and GK2 0.02%, using RT PCR method.

In summary, the safety and clinical effectiveness of IPMP/GK2 mouthwash in this study were identified in terms of its indirect bactericidal activity in the control of the deep-inside of plaque, as shown in the reduction of periodontal pocket depth and GCF as well as effective inhibition of oral microorganisms.

Key words: Dipotassium Glycyrrhizinate (GK2), Gingival Crevicular Fluid (GCF), Isopropyl Methylphenol (IPMP), Mouthwash

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Introduction

In the recent years, more attention has been focused on maintaining adequate levels of oral hygiene due to better living conditions, thus resulting in growing demand of mouthwashes for adjunctive use of oral environmental items. Accordingly, mouthwashes incorporating a variety of active ingredients have been studied to prevent dental caries and periodontal disease[1]. Of these ingredients contained in mouthwashes, sodium fluoride is generally used at the concentration of 0.05%[2]. Mouthwashes are also used to get a pleasant, refreshing taste and mask bad breath (halitosis)

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temporarily. Individuals may carry mouthwashes for more frequent use. Therefore, the long-term safety profile of mouthwashes should be guaranteed, along with their therapeutic efficacy[3].

Of edible salts, Korean bamboo roasted salt is sun-dried salt on west coast of Korea. It is sealed with yellow ocher and pine resin and heated to 850 ~ 1,500℃ in a clay furnace. The salt is then grinded. The bamboo salt has been shown to be effective for alleviating tooth pain and inflammatory periodontal tissue.

Held[4] reported that edible salt is effective in alleviating inflammation in the periodontal tissues.

Keyes and McCable[5] demonstrated that a mixture of sodium bicarbonate, hydrogen peroxide and edible salts reduce inflammation, tooth mobility, periodontal pocket and plaque in patients with periodontal disease.

Ursodeoxycholic acid (UDCA) is one of the bile acids produced by bear. UDCA has been used in the treatment of liver disease as it dissolves fat and inhibits the disposition of fats in the liver. UDCA is an effective agent for dissolving cholesterol gallstone[6]. Hydrophilic UDCA was chemically synthesized from cholic acid in 1952 and brought to market. UDCA may be also used in the prevention and treatment of periodontal disease based on the mechanism that it inhibits interleukin-1§, produced from immune cells by cellular toxicity stimulation, as well as the production of prostaglandine E_2 and collagenase[7,8]. Naturally occurring *Curcuma xanthorrhiza* belongs to the Zingiberaceae family and is a traditional herbs in Indonesia. Xanthorrhizol (1,3,5,10-bisabolatetraen-3-ol) contained in the root is sesquiterpenoid compound[9-11] cetylpyridinium chloride (CPC) has been proven to exhibit an inhibitory action against dental microorganisms. Xanthorrhizol is an active ingredient in the extract and its minimum inhibitory concentration (MIC) against dental caries-related strain *Streptococcus mutans*(*S.mutans*) is similar to that of chlorhexidine[12,13]. Brown stains between the teeth in CPC are less visible than those found in chlorhexidine, with better taste[14].

Triclosan has been shown to exhibit potent antimicrobial and antifungal actions with less harm to the human body. It is used in cosmetics and toothpastes. With its inhibitory action against bacteria or fungi, triclosan has been widely used as active ingredient for anti-acne product, antimicrobial bath-product and disinfectant[15].

So far, mouthwashes containing a variety of naturally occurring substances are being under development to ensure oral environmental improvements of the conventional products.

Based on the research model[16], we designed this study by developing a mouthwash containing isopropyl methylphenol (hereinafter called IPMP) and dipotassium glycyrrhizinate (hereinafter called GK2) as active ingredients to evaluate some of oral environmental indexes. IPMP was used by the research of Haruo[17] who demonstrated that non-ionic agents with adequate partition coefficient, such as isopropylmethylphenol, penetrated into biofilm at the high rate and killed bacteria in the biofilm strongly. GK2 was proven by Odaka Akito et al.[18] as having an anti-inflammatory property.

The results showed that the use of IPMP or GK2 gave significant impacts on halitosis, plaque formation and *S. mutans*, *S. sobrinus* and *P. gingivalis* counts within specific plaque, as well as changes in gingival crevicular fluid (hereinafter called GCF).

Materials and Methods

Subjects

Of those patients who visited Dept. of Preventive Dentistry, College of Dentistry, Dankook University, a total of 30 Korean healthy male or female volunteers aged 20 to 35 years were enrolled for this study. Eligible male or female subjects with periodontal disease had to have no drug allergy through oral diagnosis and preliminary oral test; had no a documented history of an antibiotic for the recent 1 year; were not included in the following exclusion criteria. In addition, patients had to demonstrate more than 0.5 of gingival index, when measured by the gingival index of L e & Silness[19]. This study was conducted by procedures of the Institutional Review Board (IRB No: KHUSD 0804-01).

Exclusion criteria:

(1) Patients equipped with orthodontics system or denture in part in whole

(2) Patients with serious pathological findings in oral tissue, such as oral cancer

(3) Patients with dental caries

(4) Patients with more than 5 teeth in the mouth requiring immediate treatment

(5) Patients who participated in other clinical trials within the last 6 months

(6) Patients with oral disease that require dental treatment by the investigator judgment during the

Table 1. *Subjects*

Gender	N	Age (year)			
		Mean	S.D.	Min.	Max.
Total	30	25.5	2.2	21	31
Male	18	25.6	2.0	21	29
Female	12	25.4	2.6	22	31

Table 2. *Composition of Standard Toothpaste (Unit: %)*

Function	Ingredients	Contents
Abrasive	Silica	18.0
Thickeners/Binders	Silica, etc.	4.3
Humectants	Sorbitol Solution, PEG	38.0
Others	Foaming Agent, Preservatives, Sweetener, Flavor, Colorant Purified Water	to be 100

Table 3. *Composition of study mouthwash (Unit: %)*

Ingredients	Placebo	Group II	Group III	Group IV
IPMP	-	0.02	0.02	-
GK2	-	0.02	-	0.02
Sodium Fluoride	0.022	0.022	0.022	0.022
*Others	to 100.0	to 100.0	to 100.0	to 100.0

*Others Sweetener/Humectant/pH Controller/Solubilizer/Flavor/Preservative, etc

study period

Methods

This double-blind study was carried out in a form of cross-over design which may be applicable to the same subjects. We examined the regular dietary habits of each subject.

Following the commencement of this study, subjects in Group I were instructed to use a toothpaste (hereinafter called standard toothpaste or base) over a 2-week carry-out effect period. The composition of standard toothpaste is as follows table 1.

Also, the composition of the study mouthwash is as follows table 2. Then, subjects were advised to use a placebo containing no IPMP and GK2 over 2 weeks to determine the first oral index (baseline). In the same procedures as above, subjects in group II were also instructed to use the standard toothpaste over a 2-week carry-out effect period. Then, subjects were advised to use a mouthwash containing 0.02% IPMP and 0.02% GK2 over 2 weeks to determine the second oral index. Subjects in group III were instructed to use the standard toothpaste over a 2-week carry-out effect period. Then, subjects were advised to use a mouthwash containing 0.02% IPMP without GK2 over 2 weeks to determine the third oral index.

Subjects in group IV were instructed to use the standard toothpaste over a 2-week carry-out effect period. Then, subjects were advised to use a mouthwash containing 0.02% GK2 without IPMP over 2 weeks to determine the fourth oral index. During the study period, subjects were instructed to brush their teeth using the standard toothpaste, twice daily (after breakfast and supper). As shown in Table 2, subjects were also instructed to use each mouthwash, thrice daily (12~15 ml once) 2 hours after breakfast and supper and within 3 minutes after lunch.

Oral examination

We examined the regular dietary habit of each subject and conducted several tests to assess the changes in halitosis, periodontal pocket, GCF and plaque deposits. During the oral examination, one of dentists in this study was responsible for taking charge of all evaluable items hereto to minimize the possible bias among investigators.

Halitosis

(1) Measuring procedure using OralChroma

Hold the mouth for at least 3 minutes prior to measurement to collect the volatile sulfur compounds (VSC). Insert the plastic syringe that comes with the product deep into the oral cavity and hold it lightly between the lips. Be careful to avoid touching the tongue with the syringe, blowing or

inhaling air through the syringe. Then, slowly pull the plunger, push it in again, and pull it for the second time before removing the syringe from the mouth.

If the top of the syringe is wet, wipe it dry with a tissue. Attach the dedicated needle and eject the sampled oral gas to 0.5 mL by pushing the plunger. Inject the remaining oral gas into the inlet on the main unit of the OralChroma. Then, after a signal sound, the measurement will start automatically and take out the plastic syringe after pressing the plunger with fingers.

Eight minutes later, record each component of VSC hydrogen sulfide (H_2S), methyl mercaptan (CH_3SH) and dimethyl sulfide $(CH_3)_2S$).

(2) Bad breath checker (B/B checker)

Halitosis is measured in the following procedures, using bad breath checker (TM mBA-4, TAIYO instrument INC, Japan):

Connect B/B checker to a power adaptor and wait for a 5-minute warming-up after automatic turn-on. Press OG button prior to measuring halitosis in the mouth. Then, a 180-second countdown will start. Hold the mouth for 3 minutes to collect the mouth gas via nasal breath. Once a signal sound is heard after 3 minutes, insert the measurement sensor equipped with a mouthpiece into the mouth and measure bad breath for 15 seconds in a breathless state. Insert the measurement sensor and mouthpiece into oral cavity by about 4 to 5 cm and hold it lightly between the teeth and lips. Record the measuring values marked at liquid crystal display.

To measure exhaled halitosis, press EGbutton and hold the mouth for a 30-second countdown. Once a signal sound is heard after 30 seconds, insert the measurement sensor equipped with a new mouthpiece into the mouth and measure bad breath for 15 seconds in a prolonged exhalation state. In the same manner as above, record the measuring values marked at liquid crystal display.

Periodontitis

(1) Periodontal pocket depth test

The periodontal pocket depths of 6 teeth (right maxillary No. 1 molar teeth, right maxillary central incisor, left maxillary No. 1 molar teeth, left mandibular No. 1 molar teeth, left mandibular central incisor and right mandibular No. 1 molar teeth) were measured and recorded using a periodontal probe.

(2) GCF measurement

GCF volume was measured using Periotron 8000¤ micro-moisture meter (Oraflow Inc., USA) that has been designed to measure GCF, periodontal pocket fluid (PPF), salivary flow and viscosity. For measuring larger volumes of GCF fluid, Periopaper strip (Oraflow Inc., USA)was used. Target teeth were separated between lips and vestibular organ using a cotton stick and dried by compressed air at the opposite direction of gingival pocket to maintain the influx of GCF for 30 seconds. Then, the sterilized periopaper strip picked by a pincette was cautiously inserted to a pure-cotton gingival pocket of the target teeth until a mild resistance was became apparent in gingival pocket. GCF volume was collected for 30 seconds without any movement. The periopaper strip removed from the gum was immediately placed between the two electrodes of Periotron 8000¤ to record the measuring figures. Periopaper was sealed and stored in a dried standard envelope to avoid errors in the measuring values, when being exposed to wetness during storage.

Plaque

Plaque deposits were measured at six surfaces by dividing the facial surfaces of each subject into proximal, distal and central surfaces, in accordance with Quigley and Hein s Plaque Index as modified by Turesky[20]. Individual plaque indexes were calculated by dividing total score into number of surfaces examined. Target teeth included No. 16, No. 11, No. 26, No. 46, No. 31and No. 36.

0: no plaque
1: separate flecks of plaque at the cervical margin of the tooth
2: a thin continuous band of plaque (up to 1 mm) at the cervical margin of the tooth
3: a band of plaque wider than 1 mm covering less than one third of the crown of the tooth
4: plaque covering at least one-third but less than two thirds of the crown of the tooth
5: plaque covering two-thirds or more of the crown of the tooth

Changes in oral microorganism through real-time reverse-transcription polymerase chain reaction

To determine the number of *P. gingivalis*, *S. mutans* and *S. sobrinus* that are oral microorganisms in the two major dental diseases (periodontal disease and dental caries), paraffin wax was chewed to collect the saliva-stimulating samples for 5 minutes.

Table 4. *Oralchroma value (unit:ppb)*

Group	N	HS			MM			DMS		
		Mean	ˇ(%)	S.D.	Mean	ˇ(%)	S.D.	Mean	ˇ(%)	S.D.
Base	30	44.23	-	77.1 [a]	27.03	-	38.4 [a]	17.77	-	21.7 [a]
Placebo	30	32.63	0	64.2 [ab]	27.17	0	53.4 [ab]	22.23	0	51.1 [a]
Group II	30	16.97	47.99	37.4 [b]	11.53	57.55	20.8 [b]	18.13	18.44	33.5 [a]
Group III	30	14.43	55.77	26.9 [b]	14.77	45.64	59.8 [ab]	23.47	-5.55	64.4 [a]
Group IV	30	13.07	59.96	23.1 [b]	7.30	73.13	15.9 [b]	15.50	30.28	33.6 [a]
p-value by Friedman test		0.0247			0.0171			0.3627		
p-value by Repeated measured ANOVA		0.0911			0.0568			0.9689		

[ab]: No significantly difference in the same letter(by Bonferroni test)

Table 5. *B/B checker value (unit:BBV)*

Group	N	B/B checker value		
		Mean	ˇ(%)	S.D.
Base	30	29.9	-	12.7 [a]
Placebo	30	24.6	0	13.7 [ab]
Group II	30	22.1	10.15	8.4 [b]
Group III	30	19.8	19.62	13.4 [b]
Group IV	30	21.9	10.98	11.9 [b]
p-value by Friedman test		0.0880		
p-value by Repeated measured ANOVA		0.0089		

[ab]: No significantly difference in the same letter(by Bonferroni test)

The amounts of mRNA were assayed through the real-time reverse-transcription polymerase chain reaction. *S. mutans* Ingbritt and *S. sobrinus* 6715-7 was incubated in Todd-Hewitt broth at 37℃ under ventilation condition. To determine the individual total amounts of *S. mutans* Ingbritt and *S. sobrinus* 6715-7, these strains were stained in safranin O for 5 minutes at room temperature and calculated by Petroff-Hauser counting chamber (Hausser Scientific Co., U.S.A). Bacterial DNA (2 l) was then mixed with SYBR Premix Ex *Taq*, ROX reference Dye (Takara Bio, Otsu, Japan) and each primers (0.2 M). The condition for the real-time RT-PCR reactions were 40 cycles of denaturation at 94℃ for 15 s, annealing at 60℃ for 15 s, and extension at 72 ℃ or 33 s and performed using ABI PRISM 7500 Sequence Detection System (Applied Biosystems, Darmstadt, Germany). Dissociation curves, which were verified by melting curve analysis, were obtained to confirm non-specific amplification of DNA. The quantification of target bacteria was determined using standard curve which were plotted for each primer pairs using the threshold cycle (Ct) values obtained by amplifying 10-fold serial dilutions of *S. mutans* and *S. sobrinus* of known concentration (10^2 to 10^7cells). The sequences of the primers used were 5 -CTA CAC TTT CGG GTG GCT TG-3 and 5 -GAA GCT TTT CAC CAT TAG AAG CTG-3 for *S. mutans* 5 - AAA ACA TTG GGT TAC GAT TGC G-3 and 5 - CGT CAT TGG TAG TAG CCT GA-3 for *S. sobrinus*.

Data analysis

All of the collected data were stored using SPSS software (SPSS 12.0KO for Windows, SPSS Inc, Chicago, USA). The changes in each group were analyzed by repeated measured ANOVA and the Friedman test. The significance level was considered by 0.05.

Results

Measuring results of halitosis

As a result of measuring the collected VSC hydrogen sulfide (HS), methyl mercaptan (MM), and dimethyl sulfide (DMS) using the OralChroma (No. CHM-1, Abilit, Japan), HS values in Group II, III and IV were statistically reduced significantly by 47.99%, 55.77% and 59.96%, respectively, compared to the placebo group. MM values in Group II, III and IV were statistically reduced significantly by 57.55%, 45.64% and 73.13%, respectively, compared to placebo (Table 4). DMS values in Group II and IV provided 18.44% and 30.28% reductions, respectively, compared to placebo.

Halitosis in this study was additionally measured using B/B checker, with which each gas

Table 6. *Periodontal pocket depth and GCF value*

Group	N	GCF			depth(mm)		
		Mean	ˇ(%)	S.D.	Mean	ˇ(%)	S.D.
Base	30	28.4	-	5.2 [a]	3.7	-	0.4 [a]
Placebo	30	27.1	0	5.0 [ab]	3.6	0	0.4 [a]
Group II	30	25.3	6.88	5.7 [c]	3.4	5.56	0.3 [b]
Group III	30	26.5	2.21	6.3 [bc]	3.5	2.78	0.3 [b]
Group IV	30	26.9	0.74	5.8 [abc]	3.5	2.78	0.3 [b]
p-value by Friedman test		0.0003			<0.0001		
p-value by Repeated measured ANOVA		0.0002			0.0050		

[abc]: No significantly difference in the same letter(by Bonferroni test)

Table 7. *Plaque index*

Group	N	Plaque index		
		ˇM	ˇ%	S.D.
Base	30	16.6	-	4.0 [a]
Placebo	30	16.3	0	4.5 [a]
Group II	30	13.4	17.95	4.3 [b]
Group III	30	14.3	12.26	4.7 [b]
Group IV	30	13.9	14.68	4.1 [b]
p-value by Friedman test		<0.0001		
p-value by Repeated measured ANOVA		<0.0001		

ˇM : (baseline - 4th test result)/baseline*100

ˇ% : (Placebo M - each group M)/Placebo M*100

[ab]: No significantly difference in the same letter(by Bonferroni test)

concentration in the nasal cavity and throat could be displayed. Halitosis values were statistically reduced significantly by 10.15%, 19.62% and 10.98%, respectively, compared to the placebo group (Table 5).

Measuring results of GCF and periodontal pocket depth

GCF values in Group II, III and IV provided 6.88%, 2.21% and 0.74% reductions, respectively, compared to the placebo group. There was statistically significant reduction in GCF values of Group II (Table 6). This results was somewhat anticipated before the study as it was judged that the chronic disease gingivitis could be easily or rapidly recovered to normal state and longer period than the study period should be required for visible sign of efficacy. The changes in periodontal pocket depth of Group II, III and IV were noted as 5.56%, 2.78% and 2.78%. There was statistical significances between IPMP/GK2 groups and placebo, and IPMP/GK2 groups showed a trend of improvement over periodontal disease.

Dental plaque index

Dental plaque is a biofilm that is made up of microorganisms, food debris, mucin and dead epithelial cells on the teeth. If not removed regularly, it can lead to dental caries or periodontal problems. Plaque can be detected only via special staining. Accordingly, plaque was measured using plaque checker gel-BR (GC Co, Japan). The results showed that plaque index in Group II, III and IV were statistically reduced significantly by 17.95%, 12.26% and 14.68%, respectively, compared to the placebo group (Table 7). Accordingly, the IPMP/GK2 mouthwash is basically effective in inhibiting plaque formation, a causative agent of dental caries or periodontal diseases.

Total counts of oral microorganisms

Assay of S. mutans

The number of *S. mutans*, which was calculated by a standard graph (Fig. 1) using regression equation of R^2=0.999, is described in Table 8. Changes in total counts of *S. mutans* were expressed by percentage over the difference between number of microorganisms at baseline (M1) and number of microorganisms immediately after 2- week use of given mouthwash (M3) [M1= number of microorganisms at baseline, M2= number of microorganisms measured after 1 week of given mouthwash (M2), M3= number of microorganisms measured after 2-week use of given mouthwash].

Table 8. *Changes in the number of S. mutans on average after 2-week use of given mouthwash in each sample (Unit:%)*

Group	Sample ID	Bacteria (M1-M3)/M1 x 100%	Mean	S.D.
Base	sample 1	-2.78	2816.2	96.9
	sample 2	2.71	321565.7	8822.9
	*A(%)	-0.04		
Placebo	sample 3	5.99	14995.3	1551.6
	sample 4	-13.14	397267.4	24550.1
	*A(%)	-3.58		
Group II	sample 5	43.02	621733	170572.4
	sample 6	55.80	3219.0	211.9
	*A(%)	49.41		
Group III	sample 7	34.19	8121.6	1665.6
	sample 8	39.39	60451.2	15216.7
	*A(%)	36.79		
Group IV	sample 9	9.16	8520.8	411.8
	sample 10	-4.20	360138.2	24251.4
	*A(%)	2.48		

* A(%): Changes in the number of oral microorganisms in the same group of mouthwash

Table 9. *Changes in the number of S. sobrinus on average after 2-week use of given mouthwash in each sample*

(Unit:%)

Group	Sample ID	Bacteria (S1-S3)/S1 x 100	Mean	S.D.
Base	sample 1	-10.26	150.64	13.11
	sample 2	-32.03	10532.82	3256.47
	*A(%)	-21.15	-	-
Placebo	sample 3	-16.21	67.05	19.53
	sample 4	5.12	3347.34	252.17
	*A(%)	-5.54	-	-
Group II	sample 5	49.13	88.53	34.63
	sample 6	51.37	1666.92	731.77
	*A(%)	50.25	-	-
Group III	sample 7	57.84	7.80	3.28
	sample 8	19.57	596.41	65.96
	*A(%)	38.70	-	-
Group IV	sample 9	65.32	8.64	4.44
	sample 10	51.74	65.96	22.58
	*A(%)	58.53	-	-

* A(%): Changes in the number of oral microorganisms in the same group of mouthwash

The results showed that the number of microorganisms in sample 1 after 2-week use of standard toothpaste in all subjects was increased by 2.78%, while there was a 2.71% reduction in sample 2; the number of microorganisms in placebo was decreased by 5.99% (sample 3), while there was a 13.14% increase (sample 4); the number of microorganisms in Group II was decreased by 43.02% (sample 5) and 55.80%(sample 6), respectively and the number of microorganisms in Group III was decreased by 34.19% (sample 7) and 39.39% (sample 8), respectively. By contrast, the number of microorganisms in Group IV was decreased by 9.16% (sample 9), while there was a 4.20% increase (sample 10).

Assay of S. sobrinus

The number of *S. sobrinus*, which was calculated by a standard graph using regression equation of R^2=0.982, is described in Table 9. Changes in total counts of *S. sobrinus* were expressed by percentage over the difference between number of microorganisms at baseline (S1) and number of microorganisms immediately after 2-week use of given mouthwash (S3) [S1= number of microorganisms at baseline, S2= number of microorganisms measured after 1 week of given mouthwash (S2), S3= number of microorganisms measured after 2-week use of given mouthwash]. The results showed that the number of microorganisms after 2-week use of standard

Table 10. *Changes in the number of P. gingivalis on average after 2-week use of given mouthwash in each sample (Unit:%)*

Group	Sample ID	Bacteria (G1-G3)/G1 x 100	Mean	S.D.
Base	sample 1	-33.027	40.966	6.052
Placebo	sample 2	9.300	14.069	6.581
Group II	sample 3	72.104	360.167	281.376
Group III	sample 4	57.520	610.256	242.267
Group IV	sample 5	39.083	43.519	11.603

toothpaste in all subjects was increased by 10.26%(sample 1) and 32.03%(sample 2) the number of microorganisms in placebo was increased by 16.21%(sample 3), while there was a 5.12% decrease (sample 4); the number of microorganisms in Group II was decreased by 49.13% (sample 5) and 51.37% (sample 6), respectively and the number of microorganisms in Group III was decreased by 57.84%(sample 7) and 19.57%(sample 8) reductions, respectively. By contrast, the number of microorganisms in Group IV was decreased by 65.32% (sample 9) and 51.74%(sample 10), respectively, depending on the samples.

Assay of P. gingivalis

The number of *P. gingivalis*, which was calculated by a standard graph using regression equation of $R^2=0.999$, is described in Table 10. Changes in total counts of *P. gingivalis* were expressed by percentage over the difference between number of microorganisms at baseline (G1) and number of microorganisms immediately after 2-week use of given mouthwash (G3). The results showed that the number of microorganisms after 2-week use of standard toothpaste use in all subjects was increased by 33.027%. The number of microorganisms in placebo, Group II, III and IV was decreased by 9.300%, 72.104%, 57.520% and 39.083%, respectively. In consequence, Group II, III and IV provided 62.804%, 48.220% and 29.783% reductions in the number of microorganisms, respectively, compared to placebo.

Discussion

Mouthwash is a liquid solution use for prevention of an oral condition such as dental caries, periodontal disease or halitosis as well as for oral hygiene. Mouthwash exhibits bactericidal, antimicrobial and bacteriostatic activity by acting on microorganisms in the mouth. Mouthwashes are usually classified into one of two categories - commercial mouthwash (cosmetic gargle) and therapeutic mouthwash (therapeutic gargle). In general, mouthwash or gargle means a commercial mouthwash. Mouthwash may be concurrently used in line with common oral hygiene practices, such as daily brushing and flossing. It rinses away the remaining dental plaque, thus providing a pleasant, refreshing taste. Mouthwash also functions to mask halitosis[16].

More recently, cetylpyridinium chloride (CPC), a cationic quaternary ammonium compound, has been widely used as active ingredient in toothpastes as well as in mouthwashes. In addition to chlorhexidine, CPC has been shown to be effective in preventing dental plaque and reducing gingivitis, which have been reported in a number of clinical trials.

In 2007, Biesbrock et al.[22] reported that mechanical plaque control method using toothpaste and triclosan (TCS)-containing mouthwash demonstrated statistical effectiveness after 4 weeks in a double-blind study. They also claimed that the mouthwash significantly inhibited microorganism growth and reduced gingival index and plaque index.

Kraivaphan et al.[23] conducted a double-blind study in 180 women at three months of pregnancy in Southeast Asia and reported that after five months, a triclosan dentifrice significantly reduced plaque formation, gingivitis and gingival bleeding by 40.5%, 22.5% and 35.3%, respectively, compared to placebo group.

So far, many studies on a variety of active ingredients in the mouthwash have been carried out. Lee and his colleague[24] reported that a mouthwash containing NaF, CPC and UDCA prevented dental plaque and reduced gingivitis. Bae et al.[25] also reported that the mouthwash containing NaF, CPC and green tea/pine leaf extract was effective in preventing halitosis and dental caries, compared to placebo. As noted in the previous studies, the mouthwash exhibits bactericidal, antimicrobial and bacteriostatic activity by acting on microorganisms in the mouth. On top of that, it can be used to

provide a pleasant, refreshing taste to the mouth and mask halitosis. It is important to recognize that the remaining amount of drug at the surface of oral cavity may enhance the effectiveness in the micro-environment of the target area. The twice-daily regimen of a mouthwash with long-lasting effects should be recommended at the target area.

Korean bamboo roasted salt is sun-dried salt on west coast of Korea. It is sealed with yellow ocher and pine resin and heated to 850 ~1,500 ℃ in a clay furnace. The salt is then grinded. The bamboo salt has been shown to be effective in indigestion, intoxication, trauma and bleeding as one of folk remedies.

Of several antimicrobial agents, CPC used in this study is a cationic quaternary ammonium compound[26,27] and has been widely used as active ingredient in mouthwashes. The applicable concentration of CPC in mouthwash ranges from 0.024% to 0.1%. The antimicrobial property and oral retention time of CPC is similar to those of chlorhexidine, but the reported plaque reduction effects vary. Unlike chlorhexidine, teeth discoloration has not been reported in CPC and its inhibitory actions against plaque and gingivitis are usually encouraging despite conflicting study results[28,29]. It is difficult to make a definite conclusion due to different study designs and principles. Subjects who participate in clinical trials will be further encouraged to maintain adequate levels of oral hygiene, so a certain degree of prejudice in the study results may be inevitable. As a results of measuring the collected VSC hydrogen sulfide (HS), methyl mercaptan(MM), and dimethyl sulfide (DMS) using the OralChroma, HS values in Group II,III and IV were statistically reduced significantly by 51.07%, 55.77% and 59.96%, respectively, compared to the placebo group. However, Kim et al.[30] claimed that OralChroma could detect specific halitosis-inducing substances. Thus, Halitosis in this study was additionally measured using B/B checker, with which each gas concentration in the nasal cavity and throat could be displayed. Halitosis values were statistically reduced significantly in Group II, III and IV, compared to the placebo group. Both IPMP and GK2 inhibited halitosis.

Meanwhile, the presence of gingivitis may be detected by measurement of GCF exposure time after probing or GCF volume. Further, the periodontal pocket present in collapsed periodontium implies periodontal disease in patients with periodontal disease[31].

In consideration of the fact that there is no clinically or theoretically definite classification between gingivitis and periodontal disease and gingivitis, an initial stage of periodontal disease, is regarded as periodontal disease when the inflammation can, over the years, cause deep pockets between the teeth and gums and loss of bone around teeth[32], the main reason for measuring GCF volume or periodontal pocket depth in this study lies in the fact that all patients have gingival index of more than 0.5. Accordingly, GCF measurement is an element to evaluate the changes in gingivitis. We attempted to assess the changes in periodontal disease as those in periodontal pocket, which is produced between gums and periodontal ligament. Now that gingivitis and periodontal disease are chronic oral diseases, the assessment of their changes during the study period lacks a rationale, but we included such changes in gingivitis and periodontal disease as one of the evaluable items, since the sustained use of mouthwash may predict some changes and preventive measures.

In the measurement of changes in GCF volume as a reversible oral environment index to check gingival index directly, there was statistically significant reduction in GCF values (6.88%) of Group III over placebo and increased GCF levels by stimulation is translated into an initial condition of periodontal disease[33]. It was noted that IPMP and GK2 are effective in alleviating the initial condition of periodontal disease. The increased periodontal pocket depth, which is in parallel with deteriorating periodontal disease, appears to be a reversible index that may assess gingival index directly to some degree. The periodontal pocket depth in Group II, III and IV was reduced by 4.37%, 3.26% and 3.16%, respectively, to placebo. However, there was no statistically significant difference. Lee and Shin[24] included the inhibition of plaque formation as an evaluable efficacy item of mouthwash. The efficacy assessment item included in the study showed that any mouthwashes containing IPMP or GK2 had an inhibitory action against plaque formation.

As a result of analyzing all counts of *S. mutans*, *S. sobrinus* and *P. gingivalis* through real-time PCR reactions, total number of *S. mutans* and *S. sobrinus* was reduced in Group II using IPMP and GK2. Further, number of *S. mutans* and *S. sobrinus* in IPMP or GK2 group was statistically reduced, compared to the placebo group. The number of *S. mutans* was more reduced in the group using IPMP.

In consideration of biological properties that *S. mutans* prefers an aerobic environment, the results of this study confirm the finding of Haruo kadoya et al.[17] who demonstrated that the mouthwash kills bacteria at the deep-inside of plaque.

Total counts of *P. gingivalis* were more reduced in the group using IPMP or GK2, compared to the placebo group. The fact that IPMP-containing mouthwash was superior to GK2-containing mouthwash, suggesting that IPMP-containing mouthwash has a potent bactericidal action against *P. gingivalis* present at the deep-inside of plaque.

The side effects found in the study included is comfort during gargling due to strong stimulation of a mouthwash, damage in oral mucosa, hypersensitivity in the teeth, headache due to taste or flavor of toothpaste, tooth discoloration, tongue stimulation or malfunction of tongue or vomiting. The investigator checked the daily mouthwash of each subject on a weekly basis at his/her clinic visit. With the regular monitoring of oral environmental index test on a month interval, the investigator made oral diagnosis on taste or flavor of the mouthwash used in the study, including subject s subjunctive symptoms, but there no complaint about side effects or other discomfort. The majority of subjects completed the study, and there was no severe side effect during oral examination. Therefore, the mouthwash used in the study is safe as there was no clinical sign in the teeth, tongue, gingiva and oral mucosa.

In conclusion, the mouthwash is an oral item designed to ensure oral washing or tooth-brushing effects easily in the modern society. Although a mouthwash has the defect to maintain inadequate levels of oral hygiene or prevent periodontal disease, manufacturers have tried to widen the function of mouthwash by combining CPC or fluoride into the mouthwash. More recently, fluoride has been widely used in the prevention of dental caries as well as oral environmental improvements. Accordingly, with the expanded recognition of fluoride, 99% of locally manufactured toothpastes contain a proper amount of fluoride, and sodium fluoride in mouthwash. Furthermore, In the final operation following teeth whitening, fluoride is applied to avoid partial teeth hypersensitivity. IPMP or GK2 has spotlighted in the foreign clinical trials using fluoride-containing mouthwashes as it inhibits bacterial in biofilms effectively. We obtained the aforementioned results using proper formulation of 4 different mouthwashes containing fluoride and other ingredients over 20 weeks. During the study period, we noted that IPMP/GK2 mouthwash delivered clinically relevant benefits in the control of bacteria in biofilms, while reducing plaque deposits and halitosis as well as prevention of periodontitis. The safety and clinical effectiveness of the mouthwash used in the study were best determined as GCF volume and periodontal pocket depth was significantly reduced in Group (IPMP 0.02% + GK2 0.02%) and there was no side effect and subjects complaint.

In consideration of the fact that periodontal pocket depth and total counts of *P. gingivalis* and *S. mutans* was significantly reduced in the group using IPMP mouthwash, it was noted that IPMP exhibits an bactericidal action against oral microorganisms present at the deep-inside of plaque.

Conclusion

This double-blind study was designed to evaluate the inhibitory actions of the IPMP/GK2 mouthwash on halitosis, plaque and oral microorganisms within specific plaque as well as changes in GCF over 20 weeks. The mouthwash in an appropriate amount of IPMP or GK2 was given to 30 subjects over 4 cycles (2 weeks of carry-out effect period, and 2 weeks of IPMP/GK2 mouthwash use, total 4weeks/ cycle,group x 4 groups), compared with the placebo group.

Halitosis in this study was additionally measured using B/B checker, with which each gas concentration in the nasal cavity and throat could be displayed. Halitosis values were statistically reduced significantly in the groups using IPMP/GK2 mouthwash, compared to the placebo group. Both IPMP and GK2 inhibited halitosis.

For a measurement of GCF values to assess reversible oral environmental index to check gingival index directly, there was a statistically significant reduction in GCF values in IPMP/GK2 mouthwash only.

For a measurement of periodontal pocket depth as drug irreversible index to evaluate gingival index directly, periodontal pocket depth was slightly reduced in IPMP or GK2 mouthwash, compared to the placebo group, but with no statistical difference.

Plaque index was significantly reduced in IPMP or GK2 mouthwash group, compared to placebo.

Total counts of *S. mutans* and *S. sobrinus* were statistically reduced significantly in Group II (mouthwash containing IPMP 0.02% + GK2 0.02%) during comparison before and after application. The number of *Streptococcus mutans* was reduced in

IPMP or GK2 group, compared to placebo. The number of *P. gingivalis* was statistically reduced significantly in the groups using IPMP or GK2 or both.

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Original Article

The Effect of Denture Cleanser on the Reduction of Oral Malodor and Denture Cleansing Behaviors among Korean Elderly

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Abstract

The purposes of this study were to evaluate a denture hygiene method and the attitudes of proper cleansing method of denture among Korean elderly. In this study, the effect of denture cleanser in the reduction of oral malodor was measured by OralChroma (Abilit, Japan), and the investigation of denture cleansing behaviors among Korean elders wearing dentures drawn from dental clinics in Seoul. The differences between VSCs production seen in comparisons of the denture cleanser and water (control) were statistically significant (At 30 minutes, p=0.01; at 1 hour, p=0.001). The differences in VSCs production after incubation were statistically significant (after 24 hours, p<0.05). Denture cleanser showed a large reduction in viable count (>99.5% kill) and a reduction of the VSCs in denture wearers. As a result, the denture cleanser (Polident) was more effective than water in reducing VSCs and microorganisms. The questionnaire included information about self-reported denture hygiene and denture hygiene-related behavior items. The majority of respondents reported cleaning their dentures with water or a toothbrush and toothpaste. More information and training on denture hygiene is required to raise awareness of the potential importance of cleansing dentures and reducing oral malodor.

Keywords: Denture cleanser, oral malodor, denture, Polident, Biofilm, denture cleansing agent

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Introduction

Some denture wearers experience difficulty when cleaning their dentures and a consequence many elderly people do wear dirty dentures (Harrison *et al.* 1997). Dirty dentures contribute to malodor (Neill 1968). A thorough cleaning of dentures may rate low on the priority list of some denture patients. Due to the artificial nature of the denture, many edentulous patients express concern that they may produce a distinct malodor (Fiske *et al.* 1995). The nature of malodor in denture wearers is ill defined (Verran 2005).

Once the teeth are lost, edentulous adults return to an oral microflora that closely resembles that of the infant prior to the eruption of teeth (Socransky *et al.* 1971). But the presence of dentures in edentulous patients creates yet another environment with its own microflora. Denture plaque is known to harbor increased proportions of *Candida* spp. (main etiological agent in denture stomatitis) in comparison to dental plaque. And it also affects oral malodor. The increased likelihood of the presence of *Candida* spp. in the mouth may contribute yeasty odors (Verran 2005).

It is not clear whether this relationship between volatile sulfur production and oral malodor in the denture wearer is comparable to patients with teeth. A denture wearer s oral malodor has been described as sweet but offensive (Manderson *et al.* 1975). If it could be found out a strong correlation between the VSCs and denture malodor, it is indicating a possible causative relationship.

Incubation of whole saliva at body temperature results in the generation of a malodor similar to what arises and emits from the human mouth as bad

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breath (Tonzetich 1971). The incubation of retainer or denture wetted by saliva would produce a malodor similar to bad breath.

The purpose of this study was to develop an appropriate denture hygiene method by access the basic data of proper cleaning method of denture.

Materials and Methods

The reduction of VSCs of denture wearer after rinsing with denture cleanser

Study Population

A total of 20 subjects were eligible who were edentulous and had a complete acrylic upper denture. Those who had used a denture cleanser within the previous 1 week were not eligible. Mean age was 71 (standard deviation 7.8) years old.

Clinical Examination

The purpose of the study was fully explained to the subjects who demonstrated their willingness to participate. A questionnaire about dental and medical history related to malodor and malodor history were collected first. After signing the consent form, each subject surveyed a complete oral examination. Subjects exhibiting poor dental or general health that could potentially interfere with compliance or evaluation measurements, use of medications with xerostomia or taste alternation adverse effects, recent use of antibiotics, or regular use of antimicrobial oral products were excluded from the study.

Treatment Protocol

For all patients, water only was used to clean dentures for initial one-week period before the study started. Subjects were not allowed to use scented personal products (including soap and antiperspirant) on screening and treatment days. Then water (control) was used to clean dentures in the first visit. In the crossover design, the denture cleanser (Polident¤, GlaxoSmithKline) was used in the second visit separated by a 1-week wash-out (during which water only was used). Cleaning with the Polident was performed according to the manufacturer s instruction. For water (control), dentures were soaked in the same condition. Subjects were instructed to eat breakfast earlier than 2 hours prior to visit time in the morning of each visit day, to prohibit smoking within 3 hours of visits, consuming alcoholic beverages and eating highly seasoned/spicy. Subjects were instructed to abstain from eating strong-smelling foods at least for 24 hours. Subjects attended in the morning period, the measurement was conducted between 9:00 a.m. and 12:00 a.m..

1st visit (control measurement)

(1) For those qualified subjects, a baseline OralChroma measurement was conducted with subject s dentures in place. Each subject place his/her mouth over one end of a clean syringe (1 cc). Subject should kept oral cavity closed for 30 seconds while the syringe was in the mouth. While the subject continued to hold breath, the syringe withdrew air from the mouth. 1cc of oral air was aspired twice by the syringe. Because of the dead-space effect in the syringe, the first aspirated sample was discarded, and 0.5 cc of the syringe was inserted into the injection port of the OralChroma. The syringe was provided by Abilit (Japan). Total amount of VSCs was measured.

(2) Subjects ingested odor-causing food (granulated garlic powder 1g). The VSCs were measured after 30 minutes. Subjects were asked to keep their mouths closed while breathing through the nose.

(3) Dentures were soaked in a glass of 200 ml tap water for 5 minutes.

(4) Dentures were rinsed for 10 seconds with tap water and returned to the subject for insertion.

(5) Subjects then wore denture continuously for an hour. Post-rinsing OralChroma evaluations were conducted 30 minutes and 1 hour after denture insertion.

2nd visit (experimental measurement)

The same procedures {(1)(2)(3)(4)(5)} as the control except for adding a tablet of denture cleanser (Polident) in water were conducted in the 2nd visit.

The reproduction of VSCs of retainers rinsed with denture cleanser after incubation

Study Population

A total of 10 healthy subjects (over 50 years old of age) were prepared with removable upper maxillary retainers.

Clinical Examination

The same procedures as 1. 2 were conducted.

Treatment Protocol

Water (control) was used to clean retainers in the first visit. In the crossover design, the denture cleanser (Polident) was used in the second visit with new retainer. Cleaning with the Polident was

Table 1. *Mean difference in VSCs in denture wearers.*

Treatment group	Baseline Mean (SD)	After 30 minutes of ingesting powder	After 30 minutes of rinsing (SD)	After 1 hour of rinsing (SD)
Denture cleanser	11.34 (15.99)	100	25.34 (13.62)	15.04 (7.99)
Control (Water)	6.15 (5.03)	100	37.20 (32.56)	23.67 (16.37)

Those values represent a reduction in VSCs relative to the peak value (100).

performed according to the manufacturer s instruction. For water (control), retainers were soaked in the same condition. Subjects were instructed to prohibit smoking within 3 hours of visits, consuming beverages and eating. Subjects were instructed to abstain from eating strong-smelling foods at least for 24 hours. For Oralchroma, the gas from retainer was collected in polypropylene-coated sampling bags and 1 cc of gas was collected out of the bags into sampling syringe. 0.5 cc of the syringe was inserted into the injection port of the OralChroma. Total VSCs were measured. Two same retainers were prepared in each subject. (One for the control, one for the experiment) Soft denture liner was applied to inner surface of denture to improve comfort and fitness. This liner gave more porosity, and may thus entrap and accumulate more plaque biofilm. All retainers were lined with denture liner (CoeSoft) in inner superficial layer.

1st visit (control measurement)

(1) For basement measurement, subject ingested odor-causing food (granulated garlic powder 1g). Subjects were asked to keep their mouths closed while breathing through the nose. After an hour, retainers were packed into sampling bags. VSCs were measured after 30 minutes of packing.

(2) VSCs were measured after incubation of retainer at 37℃ for 24 hours.

(3) VSCs were measured after 30 minutes from rinsing with water and after 24 hours incubation of rinsed retainers.

2nd visit (experimental measurement)

Same procedures {(1) (2) (3)} as the control except for adding a tablet of denture cleanser (Polident) in water were conducted.

The CFU reduction in dentures rinsed with denture cleanser

Ten subjects with dentures or retainers were participated in this study. In the crossover design, the denture cleanser (Polident) was used in the second visit separated by a 1-week wash-out.

1st visit (control group)

(1) Subjects were wearing their dentures or retainers at least for 2 hours.

(2) The denture was soaked in DW for 30 seconds and 1 drop of the solution was suspended in BHI plate. (brain heart infusion agar) BHI was incubated in 37℃ for 48 hours.

(3) It was soaked in a glass of 200 ml tap water for 5 minutes and rinsed with tap water for 10 seconds.

(4) After rinsing, it was soaked in DW for 30 seconds 1 drop of the solution was suspended in BHI plate. BHI was incubated in 37℃ for 48 hours.

(5) The number of colonies was calculated by multiplying the dilution magnification.

2nd visit (experimental group)

The same procedures {(1) (2) (3) (4) (5)} as the control except for adding a tablet of denture cleanser (Polident) in DW were conducted.

The behavior of denture cleansing habits

50 Korean denture patients (mean age: 65–4.1 years old) were questioned including information about self-reported denture hygiene and denture hygiene-related behavior items. Denture hygiene (cleansing, immersion) and wearing habits (overnight removal) were also recorded.

Results

Paired t-test was used to evaluate the differences between subjects with denture cleanser and with water statistically.

Subject baseline characteristics were generally well-balanced among treatment groups. The relative VSCs percents for the denture cleanser were 25.34 % at 30 minutes and 15.04 % at 1 hour and for water (control) 37.20% and 23.67 %, respectively. While smaller in magnitude, the differences in VSCs production seen in comparisons of the denture cleanser and water (control) were statistically significant (At 30 minutes, $p=0.01$ and at 1 hour, $p=0.001$).

At evaluation time-points following denture cleansing (30 minutes and the incubation for 24

***Table 2**. Mean difference in the reproduction of VSCs after incubating the rinsed retainers for 24 hours.*

Treatment group	Baseline Mean (SD)	After 30 minutes of ingesting powder	After 30 minutes of rinsing (SD)	After 24 hour of rinsing (SD)
Denture cleanser (Polident)	22.39 (23.29)	100	16.01 (21.39)	30.56 (37.75)
Control (Water)	14.88 (23.39)	100	23.40 (31.05)	65.65 (42.91)

hours), retainers rinsed with denture cleanser exhibited statistically significantly lower mean in the 24 hours incubation of rinsed retainer. (The peak value of VSCs after the incubation was adjusted to 100. And the other values were adjusted relative to the peak value of 100.) The relative VSCs percents for the denture cleanser were 16.01 % at 30 minutes and 30.56 % after the incubation and for water (control) 23.40% and 65.65 %, respectively. The differences in VSCs production seen in comparisons of the denture cleanser and water (control) were statistically significant in the incubation of rinsed retainers (After 30 minutes, $p>0.05$ and after 24 hours, $p<0.05$).

In the control group, the CFU was reduced by 75.50% and with the denture cleaner the CFU was reduced by 99.50%. There was a significant difference between cleanser and control. ($p<0.001$)

Over 50% of participants reported to store dentures by soaking in water overnight. The majority of participants reported cleaning denture with water rinsing (48.5%) and with a toothbrush and toothpaste (20%). Less than 20% cleansed dentures using denture cleanser. Over 21% reported the denture odor caused by the oral deposits and microorganisms.

Discussion

Production of malodorous compounds is dynamic and changes in response to nutrient sources (Loesche *et al.* 2002). Salivary protein itself could not lead to the occurrence of malodor. Biofilms containing populations of anaerobic species are highly active with regard to VSC production from cysteine. However, without cysteine, the H_2S level is relatively low (Spencer *et al.* 2007). According to this result, granulated garlic powder containing cysteine was ingested as odor-causing food. *In vivo*, production of VSCs was induced upon provision of peptides and amino acids in the mouth (Waler 1997). The biofilm response to cysteine pulses was similar to the response of the human mouth with rinses of cysteine (Kleinberg *et al.* 2008). Without this method, the magnitude of the VSCs levels detected with the OralChroma was hovering closely to the lower limit of detection for the instrument and between regimen differences were very small. The mean baseline VSCs for this denture population (Table 1) were noticeably lower than those seen in dentate populations (Gerlach *et al.* 1998). Thus some variation can be expected between measurements, especially below the cognitive thresholds. After 3 hours of ingesting granulated garlic powder, the VSCs level of subject tends to return to the level of baseline measurement in the pilot study. So the VSCs levels were measured within this time period.

Inter-subject variability is quite high regardless of the sample collection method used, so it is less important to focus on the strict quantitation of VSCs and microorganisms as it is to focus on the general trend for one subject. The wash-out periods were intended to allow *Candida* spp. and bacteria to repopulate the dentures and to eliminate the effects of the previous cleanser. In this study, tap water was used as control because most of denture users soak dentures in tap water rather than diluted water (DW).

Oxone which is major ingredient in Polident can be used to oxidize H_2S (hydrogen sulfide), mercaptans, sulfides, disulfides, sulfides (www.Dupont.com). Although this effect is weak, it can attribute some reduction of VSCs. Analytic testing must be used to determine the effect on oral malodor.

In total, the experiments were treated with denture cleanser showed a large reduction in viable count (>99.5% kill) and a slow reduction in the VSCs reproduction rate, consistent with the reduction in microorganisms rather than direct inhibitory effects on the biotransformation steps, and a reduction of the VSCs in denture wearers. It is important to concede the mere presence of a VSC-producing bacterium is not sufficient to explain denture bad breath.

The results of this study showed the denture cleanser (Polident) were more effective than water in reducing VSCs and microorganisms from denture acrylic resin. This study has significance in measuring the VSCs reduction effect of denture cleanser by reliable digital readout equipment (OralChroma). More information and training on

denture hygiene is required to raise awareness of the potential importance of cleansing dentures and reducing oral malodor.

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ASIAN FLUORIDE COMMISSION

Date 15 Sep. 2007
Seoul Educational & Cultural Center
Asian Academy of Preventive Dentistry

Asian Academy of Preventive Dentistry

Asian Fluoride Commission
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Chairman Dr, Prathip. Phantumvanit (Thailand)

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Session 2, Water Fluoridation
Dr. Kim, Jin-bum,and Dr. Kim Jhong-Bai (Korea)

Session 3, Fluoride Mouthrinsing
Dr. Kobayashi, S. ,Dr. Sakai.O, and Dr. Miyazaki H. (Japan)

Session 4, Fluoride Salt, Milk Fluoridation
Dr. P. Phantumvanit (Thailand), and Dr. Maki, Y.(Japan)

Session 5, Professional Fluoride Application (Topical application at dental clinics)
Dr. Shin, S.C. (Korea)and Iijima, Y.(Japan)

Participants:
Dr. Yupin (Thailand)
Dr. Bahar (Indonesia)
Dr. Rahimah (Malaysia)
Dr. James Lee (Brunei)
Dr. Paik Y. D (Korea)
Dr. Paik (Korea) Dr. Ogawa (Japan)
Dr. Monse (philippine)
Dr. Bella Monse (philippine) Dr. Cho J. W (Korea)

Sample format to implement guidelines

Asian Fluoride Commission

Title:

Tasks:

1. Definition

2. Background and History

3. Objective

4. Criteria
   - Oral health status (dental caries, self-care practices etc.)
   - Fluoride sources I Fluoride mapping
   - Economics I Effectiveness
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5. Application method
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   - Effectiveness
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9. References

Title: Fluoride Toothpaste Usage and Recommendation for Asia

Tasks:

1. Definition

Topical fluoride delivery from toothpaste products help to strengthen tooth enamel and remineralize tooth decay and will be effective against caries in all individuals at risk for caries.

2. Background and History

Fluoridated toothpaste has been proven effective and safe in caries prevention by numerous scientific researches and the daily oral hygiene practices of billions of people around the world over the past 50 years.

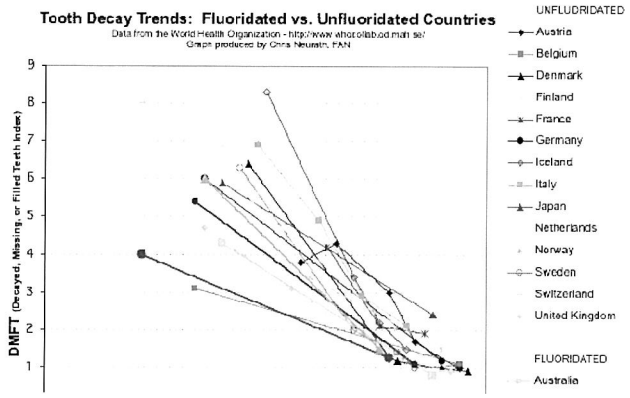
Anti-caries Mechanism

Fluoride is well known for it's ability to prevent caries. Pre-eruptively, ingested fluoride is incorporated into the developing enamel hydroxyapatite crystal where it has the effect of reducing enamel solubility(Beltran&Burt,1988). Post-eruptively, the frequent presence of low concentrations of fluoride into the oral cavity, such as through drinking fluoridated water or brushing with a fluoridated toothpaste, enhances remineralization, e.g. rebuilding the enamel matrix when demineralization has occurred in the early stages of the carious process (Koulourides,1990).

Topical exposure from fluoridated toothpaste results in fluoride uptake, as partially fluoridated hydroxyapatite, into the enamel. This incorporation of fluoride into the tooth improves enamel stability and increases its resistance to subsequent acid challenge. There is increasing evidence that the cariostatic activity of fluoride is primarily due to its topical effects on teeth. The frequency of use and concentration of fluoride in toothpaste is critical [SCCP/0882/05]

Efficacy

From the following figure, it is clear to see the decline in caries rates with increasing use of fluoride dentifrice in countries with and without fluoridated drinking water.



Fluoride dentifrice products have been demonstrated effective in the presence of water fluoridation [Jackson et al, 1995].

Opinions from Experts

After comprehensive evaluation and consideration, the efficacy and safety of fluoridated toothpaste has been endorsed by global regulatory agencies and dental associations. As a result, fluoridated toothpaste has been widely applied around the world.

During the Global Consultation on "Oral Health through Fluoride", which was jointly convened by the World Health Organization (WHO), FDI World Dental Federation and the International Association for Dental Research (IADR) on 17-19 November 2006, a panel of experts on fluoride emphatically made clear that prevention by using fluoride is the only realistic way of reducing caries burden in populations. Moreover, the experts reaffirmed the efficiency, cost effectiveness, and safety of the daily use of optimal fluoride and confirmed that universal access to fluoride for dental health is a part of the basic human right to health. The three authoritative agencies especially emphasized that governments should develop necessary programs and encourage suppliers to improve availability of effective affordable fluoride toothpaste for disadvantaged populations [1].

3. Objective

To prevent tooth decay, which is particularly high in children, since that most tooth decay remains untreated which can result in dangerous infection, pain and loss of school days.

To decrease the burden caused by tooth decay, which affects children, adults and the elderly, disrupts life and causes considerable pain, suffering and economic hardship.

Exposure to appropriate fluoride, in particular through fluoride toothpaste, will improve quality of life and enhance the achievement of the Millennium Development Goals by reducing the high dental disease burden of entire populations, especially children.

4. Criteria

Early clinical trials in the 1950s used stannous fluoride as the active ingredient; later studies with stannous fluoride toothpastes reported caries reductions of 15-30% among children over periods of 2-3 years; Subsequent trials of other fluoride compounds, added to a number of different abrasive systems, have generally produced similar results. Toothpastes have now been successfully tested with stannous fluoride , sodium fluoride, sodium monofluorophosphate and amine fluoride as the active ingredient. Most products today contain sodium fluoride or sodium monofluorophosphate as the active ingredient, usually in concentrations of 1000-1100 mg F/g, though toothpastes with 1500 mg F/g have been successfully tested and are marketed in Europe and USA[2]. According to the clinical data, fluoridated toothpaste containing 1100ppm-1500-ppm fluoride can significantly reduce the incidence of cavities for both children and adults [Marinho et al, 2003; Rolla et al, 1991]. Dental Hospital of Peking University conducted a clinical trial in 1300, 3-5 years old children in Beijing. After 2 years of using 1100ppm fluoridated toothpaste, the cavity incidence was reduced by 21% vs control group [P&G].

5. Application Method

Target population: Everyone with natural tooth;

Frequency: brush teeth daily: Fluoride toothpaste should be used daily by everyone with natural tooth.

6. Utilization

Fluoride toothpaste remains the most widespread and significant form of fluoride used globally and the most rigorously evaluated vehicle for fluoride use. The effectiveness of fluoride toothpaste has been assessed since the 1940's in over one hundred clinical trials and the anti-tooth decay (anti-caries) efficacy of fluoride toothpaste has been confirmed.

Fluoride toothpaste is safe to use irrespective of low, normal or high fluoride exposure from other sources. There are no adverse health consequences, everyone, even in areas with high natural fluoride, should have access to fluoride dentifrice to protect against tooth decay. Fluoride toothpaste is cost-effectiveness of the daily use of optimal fluoride.

In many industrialized counties fluoride-containing toothpastes make up more than 95% of all toothpaste sales, while the situation in developing country is various.

The use of fluoride is a primary reason why oral health has improved so much in the developed world over the last generation or so.

The use of fluoride toothpaste in China was increasing rapidly during the past two decades[3]. According to the second and third national oral health survey in China, 18.5% of the 12-year-old student brushed their teeth with fluoride toothpaste in 1995 [4], while the percentage in 2005 was 45.9%.

7. Consideration

Fluoride toothpaste has been the most rigorously evaluated of the various vehicles for fluoride. Daily use of fluoride toothpaste may reduce the DMFT 3-year increment by 25 per cent[5,6]. The effect of fluoride toothpaste increased with higher baseline levels of D(M)FS, higher fluoride concentration, higher frequency of use, and supervised brushing, but was not influenced by exposure to water fluoridation. However, an important limitation is that the effectiveness of fluoride toothpaste depends on the behavior of the individual and the family in purchasing and regularly using the products.

It should be emphasized that "topical" fluorides such as toothpaste can also have a "systemic" effect when they are inadvertently ingested by young children. Recent evidence suggests that use of fluoride toothpaste from an early age is associated with higher levels of very mild fluorosis, and this might result from the recent research result that young children inadvertently swallow a considerable proportion of the toothpaste they use[7, 8]. Because the fluorosis recorded in these studies was confined to the very mild grades and was not aesthetically compromising, the use of fluoride toothpaste should continue to be promoted in young children[9]. It is hoped that lower-fluoride toothpaste may reduce the risk of fluorosis while substantially retaining caries-preventive benefits. Products containing fluoride 400 ppm have been marketed for years in some countries as "children's toothpaste", but no clinical trials with these products have been conducted.

The following suggestion should be concerned for young children: Fluoride toothpaste tubes should carry advice that for children under the age of 6 years brushing should be supervised and only a very small amount (less than 5mm, or pea-sized) should be placed on the brush. Let the children to spit out the remnant toothpaste and rinse mouth thoroughly after toothbrush. The use of toothpaste with candy-like flavors or containing fluoride at 1500 ppm or more by children under 6 years of age should not be encouraged.

8. Recommendation

Public education is needed to promote using Fluoride toothpaste correctly and effectively, especially amount children, who have higher risk of caries. Specific recommendations for the use of fluoride toothpaste in very young children should follow guidelines from the respective national authority.

Governmental institutions promoting oral health and general health, the medical and dental professions, the educational system (e.g. health promotion in schools) and industry should take action to ensure that populations know the benefits of regular use of fluoride toothpaste and that fluoride toothpaste is made accessible and affordable.

Encourage governments to reduce or remove taxation and tariffs on fluoride toothpaste for dental health [1]

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Session2: Water Fluoridation / Dr. Kim , Jin-bum / Dr. Kim Jhong-Bai

Title: Water Fluoridation

Tasks:

1. Definition

Water Fluoridation is the deliberate upward adjustment of the natural trace element, fluorine (in the ionic form of fluoride), using guidelines developed by scientific and medical research, for the purpose of promoting the public's health through the prevention of tooth decay.

2. Background and History

A young dentist, Frederick S. McKay, practicing in Colorado Spring, US, noticed brownish stained teeth from many patients. He caned it "Colorado Brown Stain". In 1908, he initiated a study and this condition was prevalent throughout the surrounding EI Paso County. Dr. McKay and Dr. O. V. Black wrote detailed descriptions of mottled teeth.
In the 1920's, Dr. McKay, along with others, concluded that something either in or missing from the drinking water was causing the mottled enamel, In the late 1920's, Dr. McKay made another discovery. Teeth with mottled enamel were essentially free of dental caries. In 1931, fluoride was identified as the element in the drinking water that caused mottled enamel, but also inhibited dental caries.

In the 1930's, Dr. OJ. Cox, a dental researcher was the first person to propose adding fluoride to the drinking water for the prevention of dental caries.
Under the direction of Dr. H. Trendley Dean fluoridation began in 1945, in Grand Rapids, Michigan; in Newburgh, New York; in Brantford, Ontario, Canada.
In 1963, Franz J. Maier, a sanitary engineer, published the first comprehensive book on the technical aspects of fluoridation; the "Manual of Water Fluoridation Practice". In 1972, Ervin Bellack, a chemist with the US Environmental Protection Agency published the "Fluoridation Engineering Manual."
In1958, World Health Organization published the "First Report of Expert Committee on Water Fluoridation" as a Technical Report Series, No. 146 and recommended; 1. Drinking-water containing about 1 ppm fluoride has a marked caries-preventive action. Maximum benefits are conferred if such water is consumed throughout life. 2. There is no evidence that water containing this concentration of fluoride impairs the general health. 3. Controlled fluoridation of drinking water is a practicable and effective public health measure.
Over the past 50 years, continuous studies have been conducted on fluorides and fluoridation.

3. Objective

The main objective of water fluoridation is the prevention of dental caries on deciduous and permanent teeth to all population from young children to aged elderly people.

4. Criteria

Fluoride can be found in a solid form in minerals such as fluorspar, cryolite, and apatite. Fluorspar(also called fluorite) is a mineral containing from 30 to 98% calcium fluoride(CaF2). Cryolite(Na3AIF6) is a compound of aluminum, sodium, and fluoride. Apatite[CalO(P04,C03)6,(F, CI, OH)2] is a deposit of a mixture of calcium compounds. Apatite contains from 3 to 7% fluoride and is the main source of fluorides used in water fluoridation as the present time. The raw material of apatite is also used for phosphate fertilizers.

Theoretically, any compound that forms fluoride ions in water solution can be used for adjusting the fluoride content of a water supply. However, there are several practical considerations involved in selecting compounds. First, the compound must have sufficient solubility to permit its use in routine water plant practice. Second, the cation to which fluoride ion is attached must not have any undesirable characteristics. Third, the material should be relatively inexpensive and readily available in grades of size and purity suitable for their intended use.

Three basic fluoride chemicals are commonly used for fluoridation drinking water supplies; sodium fluoride, sodium fluorosilicate, and fluorosilicic acid. Sodium fluoride (NaF) is a white odorless material available either as a powder or in the form of crystals of varies sizes. Sodium fluorosilicate (Na2SiF6) is a white odorless crystalline powder. Fluosilicic acid (H2SiF6) is a 20 to 35% aqueous solution and exhibit a low pH (1.2).

Fluosilicic acid is mainly manufactured as a by-product of phosphate fertilizer manufacture. Sodium fluoride and Sodium fluorosilicate are mainly converted from fluosilicic acid.
Water fluoridation programme is the most effective and equitable method in preventing dental caries of the whole population. It would benefit most the socially and financially disadvantage group.
The reduction of dental caries on permanent teeth through water fluoridation was reported as 50¬60% in 1950's in US. However, the reduction rate was reported as 30-40% at the community where most population used the fluoride-containing toothpaste before the initiation of water fluoridation. The reduction of dental caries on deciduous teeth through water tluoridation at the community where most population used the fluoride-containing toothpaste was reported as approximately 30% compared to control group using fluoride-containing toothpaste.
Benefit population includes the elderly population as well as children owing to the reduction of root caries.
The cost of water fluoridation per person each year reported as 0.3-0.5 US dollar. Benefit Cost Ratio was reported as 8.64 by the saving money requiring to filling on permanent dentition of children in Korea.
In most countries, the prevalence rate of dental caries is the top level among chronic disease.
The requiring money to treat dental caries is so much that lots of poor population lives without proper treatment and prosthetics,

5. Application method

Dental caries occurs on any aged population. Water fluoridation programme is the most cost effective programme to prevent dental caries for all ages. However, new occurrences of dental caries are more in the children, adolescents and the elderly group. Many occurrences of root caries are found in the elderly group by the insufficient salivary flow rate.
The first benefited group is children and adolescents, and the second; the elderly. Nowadays the number of elderly population increases sharply.

6. Utilization

Water fluoridation programme is recommended for the prevention of dental caries where the public water systems have been well developed all over the world. For the community where public water systems have been well developed, the other tool of fluoride use can be considerable.

7. Consideration

There is no scientific evidence to show that water fluoridation programme has a injurious impact on general health. There is no scientific evidence to show that water fluoridation programme is harmful to the environment. However, to prevent the increase of very mild fluorosis, education is required for the children to use only small quantity of fluoride-containing toothpaste in the fluoridated area. These days we can encounter anti-fluoridationists. Most of them don't believe in the modern scientific findings and have a tendency to ignore the science. Some of them live in the crowded urban area and enjoy modem equipments for the daily living. However, they apparently say they like the rural life without modem equipments. Lots of them insist on any artificial material or system is harmful to health. They have a tendency to consume vegetables of organic farming without artificial fertilizer. Someone says they seem to be patients of paranoia.

8. Recommendation

For the protection from anti-fluoridationists, we unceasingly have to show the public the scientific data about the caries reduction effect and safety of water fluoridation. The international co-operation on the research and exchange of data will be very helpful to develop the water fluoridation program. In Asia, Singapore, Malaysia, Viet Nam, Israel, Korea and Hong Kong implement the artificial water fluoridation program at the moment.
However, some communities or countries use the naturally optimum-level fluoridated water.
For the education of water fluoridation to the public, the data from the community of naturally optimum-level fluoridated water can be very useful.

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Session3: Fluoride Mouthrinsing / Dr. Kobayashi / Dr. Sakai / Dr. Miyazaki

Title: Fluoride mouth rinse

Tasks:

1. Definition

2. Background and History 1)

In the early 1940s, it was appreciated that tooth enamel could take up fluoride ions from water solutions and that this rendered the enamel more resistant to acid solution. Over the next 50 years this topical methods has proved very successful at preventing dental caries, and different combinations of the fluoride concentration in the solution, the type of fluoride compound used, and the frequency and mode of application have been studied.

The first trial of fluoride mouth rinse was carried out in the USA. Since then, results of clinical trials at least in 14 different countries have been reported. These trials have been sufficiently favorable for dental public health officials to adopt fluoride mouth rinse as the main alternative to water fluoridation in community oriented program in many countries, e.g. Sweden, Norway, Denmark, Ireland, USA, and Cuba. Heifetz(1978) ranked fluoride mouth rinse as the most cost-effective community procedure out of six alternatives for topical fluoride therapy, and most recent articles have been concerned with this aspect. The regimen of fluoride mouth rinse is useful and practical also as home use.

3. Objective

Dental caries prevention for permanent teeth

4. Criteria

- Oral health status
- Fluoride sources
- EconomicsfEffectiveness : The most cost-effective community procedure out of six alternatives for topical fluoride therapy
- Social-demand

5. Application method

- Target: Preschool children, school children, junior high school children in school, adults at home
- Frequency: Daily method; for preschool children, in school, for adults at home Weekly method: for primary and junior high school children in school
- Rinse solution: for preschool children
  5-7ml of 0.05% sodium fluoride solution for school children and adults:
  7-10ml of 0.2% sodium fluoride solution

6. Utilization:

Japan
School-based FMR

A total of about 500,000 children participated in the school-based FMR at Japanese national level in 2006. Limited to the nursery and kindergarten schools, the number of children accounted for about 30% of all children practicing FMR.

Table 3 Number of schools and children participating, the rate of financial in the School-based FMR by grade level

Grade level	Number of school (%)*	Number of children (%)**	Financial support	
			Governments***	The others****
Nursery school & Kindergarten	3,313 9.1(%)	143,413 6.4(%)	84.1	15.9
Primary school	1,528 6.6(%)	300,912 4.2(%)	89.2	10.8
Secondary school	262 2.4(%)	45,508 1.3(%)	90.7	9.3
Special school, etc $	28 2.8(%)	1,501 2.7(%)	34.8	65.2
Total	5,131 7.2(%)	491,334 3.8(%)	85.5	14.5

* Number of school in the School-based FMR / Total school by grade level in Japan
** Number of children in the School-based FMR / Total children by grade level in Japan
*** The prefectural or municipal governments and/or deucational committee
**** The other (School, Parent, Dental Association, Jointed combination, etc)
$ The school for physically handicapped or mentally related children and the others

(Investigated by NPO-JPUF in 2006)

For home use:

In Japan there are three medicines of fluoride mouth rinse available on prescription of dentist. They have been used in daily method. Children as many as those participated in the school-based FMR would practice fluoride mouth rinse at home.

Taiwan

In 2000, 1,410,000 primary school children, 73.4% of eligible children throughout the country, enrolled in the school"based fluoride .mouth rinse program. The solution of 0.2% NaF wasused weekly.(Chin-Shun Chang. Oral Health in Taiwan, J.Dent. Health. 51(4),340-343,2000
(for other countries bellows need the additional survey)

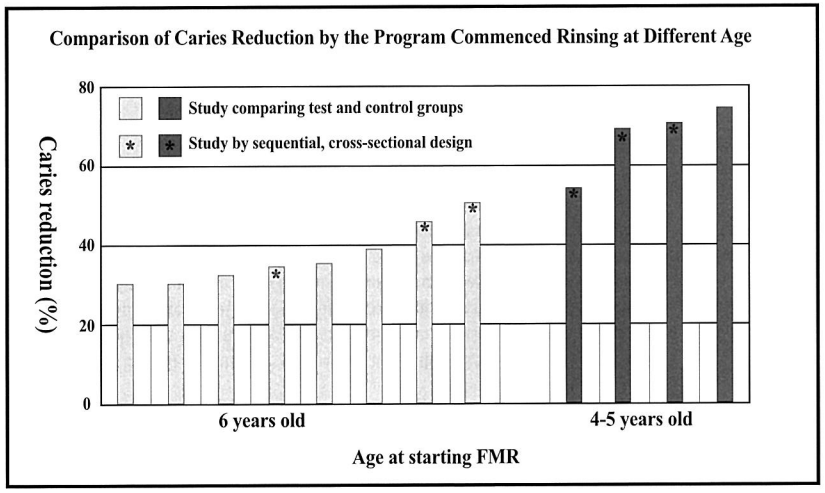
Korea
China
Thailand
Cambodia
Others

Tonga(being supported by Japanese voluntary group)
In 2006, total of 5,986 children in 6 kindergartens and 41 primary schools, enrolled in the school-based fluoride mouth rinse program. The solution of 0.05% NaF (Miranol) was used daily in kindergartens, and the solution of 0.2% NaF was used weekly in schools.Gayuri Kawamura. Spreading effects of oral health program in Kingdom of Tonga. J.Dent. Health. 2007.)

7. Consideration

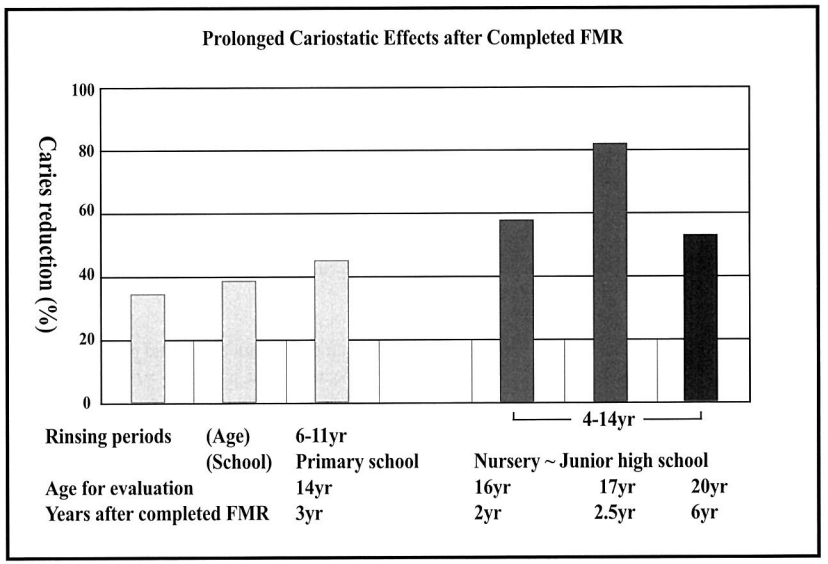
• Effectiveness

Comparison of caries reduction by the program commenced rinsing at different age According to clinical studies2'lO) in Japan, caries prevention for the first molars is much
more effective when the FMR is initiated at 4 years of age than doing so after the child enters primary school.



Rip a [11] also reviewed some historical comparison studies and observed that the earlier the children are introduced into a rinsing program and the longer they rinse, the better are the results.

Prolonged cariostatic effects after completed FMR [4, 12-16]
The cariostatic effects would be prolonged for more than 5 years after completed FMR.
The effects (53.6- 83.1%) which were obtained from FMR since preschool age were also higher than that (37.4 - 46.3%) of since 6 years old.



(Additional papers in other countries will be referred)

• Safety

The amount of fluoride retained after fluoride mouth rinse.

According to the inve stigation 17) of the amount of fluoride retained after FMR in preschool children practicing daily method (using 7ml of 0.05% NaF solution) , 12.0% of the administered dose was retained in younger group (mean age of 4 years and 10 months), corresponding to mean amount of fluoride of O.19mg. In the older group (mean age of 5 years and 4 months) the figure was 10.7% and 0.17mg. No child swallowed all ofthe solution. Of all subjects, 99.2% retained a fluoride amount that was less than O.5mg recommended (FDI 1993 and the American Dental Association 1994) for 3-5-year-olds living in a fluoride deficient area as dietary fluoride supplement.

Only six children having relatively larger values than 0.5mg retained also significantly less than the. recommended limit in an extra survey implemented after a
week.

Nowjack-Raymer et al. [18] found that there was no significant difference in the prevalence or severity of dental fluorosis among the preventive regimens, that included school-based FMR, fluoride tablet, or combined regimens, in children who began the regimens at ages 5, 6, or 7, or by eruptive status of teeth.

Yagi et a1.19) conducted an epidemiological study by application of the Fluorosis Risk Index to determine the prevalence of dental fluorosis among Japanese 5th and 6th grade elementary schoolchildren (10-12 years of age) in three communities with different exposure to fluoride. The children were exposed to fluoride as follows; FMR group who had participated in the daily program since 4 years of age in preschool (weekly rinsing with 0.2% sodium fluoride solution in an elementary school) in a fluoride deficient community; control group with no exposure to fluoride. The prevalence of enamel opacity of the FMR

group was lower than that of the control group. They found that FMR for preschoolers was not indicated to be a risk factor for dental fluorosis in fluoride deficient communities.

Acute toxicology

Probably toxic dose (PTD): doses exceeding 5mg/kg (body weight)

The symptoms of acute fluoride poisoning are nausea, vomiting and gastrointestinal pain, followed later by muscular weakness, spasms and tetany as the fluoride combines with blood Ca ions. Emergency treatment consists of emptying the stomach by stomach pump or emetic and providing Ca-containing solutions to precipitate the fluoride and to restore blood Ca. It is suggested that if the doses exceeds 5mglkg body weight, the patient should be sent to hospital. (cf; Fluorides in caries prevention third edition J.J.Murrey A.J.Rubb-Gunn and G.N.Jenkins p.333)

If children weighing 10 kilograms would drink off 32 cups containing 7 ml of 0.05 % sodium fluoride solution, the amount of fluoride ingested would exceed PTD of them. But it is unrealistic to suppose, under the supervised condition, they drink 32 cups of fluoride mouth rinse solution at a time. The supervised fluoride mouth rinse program
would be performed safely.

8. Recommendation

- Education
- Research
- Future aspects

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Session4: Fluoride Salt, Milk Fluoridation / Dr. Phantumvanit / Dr. Maki

Title: Salt Fluoridation

Tasks:

1. Definition:

Administration of fluoride via salt intake is an alternative w here the local infrastructure is not suitable for water fluoridation. One of the attractions of fluoridated salt is that it can be sold for human consumption alongside a non-fluoridated alternative.

2. Background and History:

The history of salt fluoridation spans more than half a century, mainly in Europe and the Americas. Since 1955, Zurich, Switzerland was the founder in adding both iodide and fluoride in salt for consumption and is now sold all over the country. Salt fluoridation produced benefits similar to those of water fluoridation, i.e. 50-60% caries reduction. The effectiveness depend on the fluoridated salt reaches the consumer through domestic household salt, meals at schools, large kitchens and in breads.

3. Objective:

Consideration of salt fluoridation to prevent dental caries with no side effect,
Conditions for practical salt fluoridation in Asia

4. Criteria:

- Oral health status:
  - Increasing trend of dental caries status High caries status especially in children
  - More high caries risk population with poor self care oral health

- Fluoride sources !Fluoride mapping:
  - Natural fluoride content in water source (fluoride mapping) Percentage use of fluoridated toothpaste in rural & urban population Other fluoride delivery system use in the population

- EconomicslEffectiveness
  - Cost of domestic salt and estimation of salt intake Cost of fluoridated toothpaste and availability Cost of dental restoration, extraction

- Social-demand
  - Percentage of iodized salt use Number of salt factory and condition of the salt factory
  - Legislation for fluoridated salt
  - Cost of fluoridated salt

5. Application method:

- Target
  - Total population with emphasis on young school children
- Frequency
  - Life long application
  - Several exposures of fluoridated salt per day
  - (not promoting use of more salt consumption)

6. Utilization:

Dose of 250 mgFIKg salt Cheaper use of NaF than KF
Practical use of dry method than wet method for addition of fluoride Applicable together iodized and fluoridated salt
Daily quality control and check for fluoride content in fluoridated salt Spot sample check in the market for fluoridated salt quality control

7. Consideration:

- Safety
  - Urinary fluoride excretion follow-up study in young children
  - Other fluoride biomarker for monitoring fluoride intake
  - Dental fluorosis status follow-up
  - Urinary fluoride in salt factory workers who handle fluoride addition

- Effectiveness
  - Epidemiological surveillance for dental caries and fluorosis
  - Baseline, 5-year and periodic follow•up
  - Cost benefit and cost effectiveness study

- Implications for organization of community fluoride programmes

- AntiF
  - Additional fluoride intake unnecessary
  - Side effect of fluoride intake

8. Recommendation

- Education
  - Oral health through fluoride
  - Salt fluoridation - alternative delivery fluoride
  - Support from dental schools and dental association
  - Agreement with the Ministry of Health, Food & Drug Administration
  - Educating the salt factories and Ministry of Commerce
- Health systems research
  - Availability of fluoride intake through urinary fluoride in children
  - Evaluation of community programmes and surveillance
  - Community impact to health
  - Safety of the community health
- Future aspects
  - Model for others
  - Expansion to other communities

9. References

Asian Fluoride Commission

Title: Fluoridated Milk

Tasks:

1. Definition:
Fluoridated milk has been used as a fluoride source, especially for young children through school programme.

2. Background and History:
The potential of milk as an alternative vehicle for fluoride was first reported from Switzerland in 1962. Further programmes were reported in Scotland and Hungary. Various channels have been used including kindergartens, schools as well as powdered milk and milk- cereal. It is emphasized that it is important to start the programme in early childhood to ensure an optimal effect on the deciduous teeth.

3. Objective:
Consideration of fluoridated milk to prevent dental caries with no side effect, Conditions for practical fluoridated milk in Asia

4. Criteria:
- Oral health status:
  - Moderate to high caries status especially in children
  - Increasing trend of caries status in school children
  - Poor oral health self care

- Fluoride sources !Fluoride mapping:
  - Natural fluoride in water in the region (fluoride mapping)
  - Availability of fluoride tablet provision
  - Fluoride toothpaste use and dosage
  - Fluoride intake in young children through urinary fluoride

- Economics/Effectivenese:
  - Availability of school milk programme
  - Cooperation from local diary plant
  - Monitoring of fluoride in milk in the dairy plant and milk
  - Cost effectiveness of fluoridated school milk

- Social-demand:
  - Parent-teacher association agreement
  - Cooperation from school and teachers
  - Support from local dental association and dentists

5. Application method:
- Target:
  - Primary school children Kindergarten children
  - Possibly at community mother and child care

- Frequency
  - Daily at school day
  - Higher benefit with more frequent daily consumption

6. Utilization:
- 200 ml of 2.5 ppm fluoride in school milk daily
- Fresh, pasteurized, UHT or powdered milk
- Sodium fluoride addition to milk
- Daily fluoride analysis in milk at dairy plant
- Spot samples checked for fluoride content in fluoridated milk

7. Consideration:
- Safety:
  - Urinary fluoride monitoring for fluoride intake
  - 24-hour urine or 8-hour urine in young children

- Effectiveness:
  - Baseline, 5-year and periodic follow-up
  - Cost benefit and cost effectiveness study

- Implications for organization of community fluoride programmes

- Anti-F:
  - Side effect of fluoride intake

8. Recommendation:
- Education:
  - Oral health through fluoride
  - Fluoridated milk - alternative delivery fluoride
  - Support from dental schools and dental association
  - Agreement with Food & Drug Administration
  - Educating the dairy plants and Ministry of Education

- Health systems research
  - Availability of fluoride intake through urinary fluoride in children
  - Evaluation of community programmes and surveillance
  - Community impact to health
  - Safety of the community health

- Future aspects
  - Model for others
  - Expansion to other communites

9. References:

Session5: Professional Fluoride Application (Topical application at dental clinics)

Dr. Shin / Dr. Iijima

Title: Professionally applied topical fluorides in dental clinics

1. Definition

Professional application of fluoride is defined as fluoride topical coating on the tooth surface for individuals with relatively high concentration of fluoride at the dental clinic or public health center, by dental professions.

2. Background and History

Topically applied fluoride varnish and gel applications have been used extensively as an operator-applied caries-preventive intervention for over two decades. The effectiveness of fluoride varnish and gel applications has been well established in caries prevention trials involving permanent teeth.

3. Objective

The purpose of this review is to summarize the recent published evidence regarding efficacy and safety of professionally applied topical fluorides, mainly used with fluoride varnish and fluoride gel, as well as fluoride solution, in caries prevention in dental clinics.

4. Criteria

Fluoride varnish: According to a recent published Cochrane Review of fluoride varnish in the prevention of dental caries in children up to 16 years during at least one year, the main outcome was as follows1). Nine studies were included, involving 2709 children.
For the seven that contributed data for the main meta-analysis, the D(M)FS pooled prevented fraction estimate was 46% (95% CI, 30% to 63%; p<O.OOOl; Table1). The pooled d(e/m)fs prevented fraction estimate was 30% (95% CI, 11% to 48%; p<O.OOOL Table2). No significill1t association between estimates of D(M)FS prevented fractions and baseline caries severity or background exposure to fluorides was found In meta-regression.
The authors concluded that the review suggested a substantial caries-inhibiting effect of fluoride varnish in both the permanent and the deciduous dentitions based largely on trials with no treatment controls.
There was little information concerning acceptability of treatment or possible side effects in the included trials. Given the relatively poor quality of most of the included studies and the wide confidence intervals around the estimates of effect, there remains a need for further trials.

Fluoride gel: Randomized or quasi-randomized controlled trials with blind outcome assessment, comparing fluoride gel with placebo or no treatment in children up to 16 years during at least one year2).
The main outcome was caries increment measured by the change in decayed, missing and filled tooth surfaces (D(M)FS). Twenty-five studies were included, involving 7747 children. For the 23 that contributed data for meta-analysis, the D(M)FS pooled prevented fraction estimate was 28% (95% CI, 19% to 37%; p<O.OOOL Table 3).

The effect of fluoride gel varied according to type of control group used. Only two trials reported on adverse events.

Fluoride Solution; Solution typed fluoride has been used for a long time traditionally and iontophoresis technique with 2 % NaF solution has been available in recent by lots of evidence based as ionizing NaF to F- Ion III order to increase the more uptake of fluoride at the tooth surface.

2% NaF I 8-10% SnF2, 1.23% APF, Sodium Silicofluoride, 4% Potassium Fluorstannite, Ag(NH3)F2, Amine Fluoride or other product can be used with solution, gel or thixotropic type, with some variations for concentration according to patient's state or operator's selection.

5. Application method

1) Targets
Most children over age 3 to young adults would be included in the indication for caries prevention on primary or permanent teeth. Adults and middle aged people could be involved to the indication for caries prevention or desensitization of tooth, according to patient's oral state or symptoms.
Aged peoples could be included in the indication for root caries prevention.

2) Frequency
Frequency of application is basically depends on the individual patient's oral state and operator's selection, by consideration of such factors as water fluoridated area and caries risk frequent applications might be needed at the focal potion of the target tooth surface for desensitization, by the operator's decision and patient's symptom.

3) Method
1, Painting or coating for several times with fluoride solution or vanish, about for 3-5 minutes at dried teeth surfaces with isolated from the saliva and gum, after cleaning the teeth.
2, Gel type or iontophoresis technique 1S also available according to patient's state or operator's choice.

4) cleaning
Three clinical studies 5,6,7) have reported that a cleaning or prophylaxis is not necessary before the application of topical fluoride. They indicated that no significant differences in caries reduction were found between patients who received a cleaning before application and those patients who did not receive a cleaning (Table 5). There is a little doubt that three are eventually considered to be of adequate validity and quality. The reason 1S that we can not eradicate publication bias. Cleaning and prophylaxis is necessary before the application of topical fluoride not only for caries prevention but also for gingivitis prevention.

6. Utilization

Solution type, gel type, and vanish type are prevalent used at the dental clinics in each countries. according to patient's oral status and dentist choice.

7. Consideration

High-concentration of fluoride varnish and gel are effective in preventing dental caries, but because application requires professional expertise. That is the reason why they are more expensive than the other self-applied methods such as brushing with fluoride toothpaste. In many countries including USA and Japan, fluoride varnish use for caries preventing agents is considered "off-labe1" because fluoride varnish is approved for treating hypersensitivity by the authorities . However, if the benefit-to-risk ratio i s favorable and there are many clinical trials to support the efficacy of the agents(Table 4),it is important to introduce the "off-label agents to the patients. The patients and dental hygienists preferred fluoride varnish togeI3).
Although fluoride varnish is "off-label" agents, consequently ,varnishes may be a better alternative to fluoride gels, except for persons withan history of allergic tocoiophony[14] .
Iontophoresis method with 2 % NaF is suggested to apply to children with routine preventive care or the patient with the fixed type Othodontic appliance.
Three clinical studies 5,6;7) have reported that a cleaning or prophylaxis IS not necessary before the ap-

plication of topical fluoride. They indicated that no significant differences in caries reduction were found between patients who received a cleaning before application and those patients who did not receive a cleaning (Table 5). There is a little doubt that three are eventually considered to be of adequate validity and quality. The reason IS that we can not eradicate publication bias. Cleaning and prophylaxis is necessary before the application of topical fluoride not only for caries prevention but also for gingivitis prevention.

8. Recommendation

Although the efficacy of fluoride application is well accepted, it is also true that the ingestion of fluoride during the enamel development can cause dental fluorosis. In the past also today there are several published studies that have measured or estimated total fluoride intake from various significant sources combined including topical application8,9). Research is needed in each country to accurately evaluate the exposure to and intake of fluoride from all sources with prevention of dental caries and risk of dental fluorosis.
It is known that caries prevention effect has been reported as about 30% to 70%, depending on such various factors as the kinds of fluoride selection, patient's caries activity, method of application or operator's selection and skill, etc.
Fluoride topical application is effect for caries prevention as well as for desensitization of hypersensitive dentin and contribute to tooth color recovery on the decalcified enamel through the re-mineralization effect.

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Table 1 Fluoride Varnish versus Placebo/No-treatment, D(M)FS increment (SMD) - nearest to 3 years (7 trials)

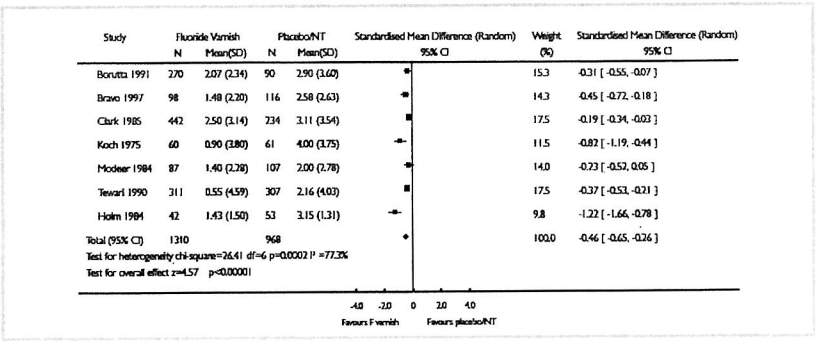

Table 2 Fluoride Varnish versus Placebo/No-treatment, d(e/m)fs increment (SMD) - nearest to 3 years (3 trials)

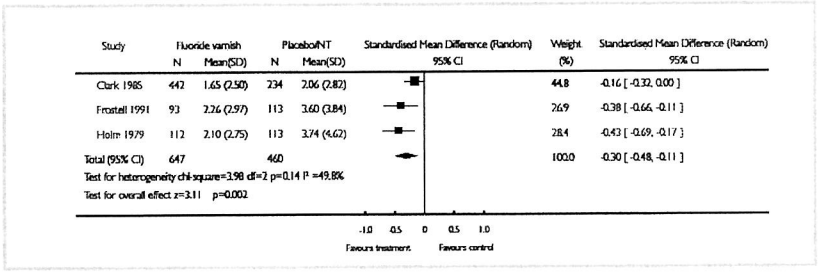

Table 3 Meta-analysis prevented fractions by fluoride gel on each condition

Analysis	No. studies	r.e. estimate	95% c. i.	Meta-analysis p-val	Heterogeneity test
D(M)FS - all studies	23	28%	(19% to 37%)	p<0.0001	Q=135 (22 d.f.); p<0.0001
D(M)FS - placebo controlled	14	21%	(14% to 28%)	p<0.0001	Q=22.5 (13 d.f.); p=0.05
D(M)FS - no-treatment control	9	38%	(24% to 53%)	p<0.0001	Q=62 (8 d.f.); p<0.0001
D(M)FT - all studies	10	32%	(19% to 46%)	p<0.0001	Q=103 (9 d.f.); p<0.0001
D(M)FT - placebo controlled	4	18%	(9% to 27%)	p<0.0001	Q=3.2 (3 d.f.); p=0.37
D(M)FT - no-treatment control	6	43%	(29% to 57%)	p<0.0001	Q=48 (5 d.f.); p<0.0001

Table 4 Compsion of professionally applied applied topical fluoride varnish gel and solution.

	Caries prevention	Clinical application	Fluoride ingestion	Cost	Acceptability	Advantage	dis advantage
varnish	Effective in high risk children (permanent teeth)	Easy Application time varies	Lowest risk Moisture can be better controlled than gel	Most expensive	Preferred by patients and hygienists Compared with gel	Effective	High cost
Gel	Effective in high risk children (permanent teeth)	Easy 4-Minute application time	% retained can be substantial Procedure must be followed to reduce risk	Low cost	Well-tollerated by patients, but varnish is preferred	Low cost easy application	Acidic
Solution with Iontopho - resis	Routine Preventive Cares	Complexity 4-Min F-ION	Low risk	Low cost sol Expensive unit	Routine Dental patients Orthodontic applianced	Safety Low cost. Material	High cost Device

Table 5 Professionally applied topical fluoride its effects with / without prior prophylaxis.

Houpt et al, 1983	APF Gel	2Y DMFT invrease 0.2 ppmF area. Randomized. no difference (ΔDMFT ca. 2-2.5)
Ripa et al, 1984	APF Gel	3Y DMFT increase 0.2 =or < ppmF area. Randomized. no difference (ΔDMFT ca. 2.0)
Johnston et al. 1995	APF Gel	3Y DMFT, increase Randomized. - Blind, no difference (ΔDMFS ca. 2.4)

Dietary Reference Intakes of Fluoride in Japanese

WHAT ARE DIETARY REFERENCE INTAKES?

ORis are reference values that are quantitative estimates of nutrient intakes to be used for planning and assessing diets for healthy people. They include RDAs as goals for intake by individuals, but also present 3 new. types of reference values (see Figurel). These include the Adequate Intake(AI), the Tolerable Upper Intake

Level (UL). and the Estimated Average Requirement(EAR).

Take1 Dietary Reference Intakes of Fluoride: Recommended levels for individual intake

	Fluoride (mgF / day)					
	Males			Female		
	AI (mg)	UL (mg)	Standard Body Weight (kg)	AI (mg)	UL (mg)	Standard Body Weight (kg)
0-5 (mo)	breast-feeding 0.01	0.66	6.6	breast-feeding 0.01	0.61	6.1
0-5 (mo)	breast-feeding 0.33	0.66	6.6	breast-feeding 0.31	0.61	6.1
6-11 (mo)	0.44	0.88	8.8	0.41	0.82	8.2
1-2 (y)	0.60	1.19	11.9	0.55	1.1	11.0
3-5 (y)	0.84	1.67	16.7	0.8	1.6	16.0
6-7 (y)	1.15	2.3	23.0	1.08	2.16	21.6
8-9 (y)	1.40	2.8	28.0	1.36	2.72	27.2
10-11 (y)	1.78	6.0	35.5	1.79	6.0	35.7
12-14 (y)	2.50	6.0	50.0	2.28	6.0	45.6
15-17 (y)	2.92	6.0	58.3	2.5	6.0	50.0
18-29 (y)	3.18	6.0	63.5	2.5	6.0	50.0
over 30y	3.40	6.0	68.0	2.64	6.0	52.7

Take2 Dietary Reference Intakes of Fluoride: Recommended levels for pregnancy and Lactaion

Pregnancy / Latation	AI (mg)	UL (mg)
Pregnancy	2.5	6.0
Lactaton	2.5	6.0

Recommended Dietary Allowance (RDA)

The average daily dietary intake level that is sufficient to meet the nutrient requirement of nearly al I (97% to 98%) healthy individuals in a group.

Adequate Intake (AI)

A recorrmended daily intake level based on observed or experimentally determined approximations of nutrient intake by a Group (or groups) of healthy people. It is used when an RDA cannot be determined.

Tolerable Upper Intake Level (UL)

The highest level of daily nutrient intake that is likely to pose no risks of adverse health effects to almost all individuals in the general population. As intake increases above the UL, the risk of adverse effects increases.

Estimated Average Requirement (EAR)

A nutrient intake value that is estimated to meet the requirement of half the healthy indi viduals in a group. It is used to assess adequacy of intakes of population groups and. along with knowledge of the distribution of requirements, to develop RDAs.

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THE EXECUTIVE MEETING ON ASIAN ACADEMY OF PREVENTIVE DENTISTRY

Date 16 Sep. 2007
Seoul Educational & Cultural Center

Asian Academy of Preventive Dentistry

President's Message

It's my great honor and pleasure to have this opportunity to invite the executive members of AAPD to Seoul and have a meeting.

As you know, 16 years has passed since the Asian Academy of Preventive Dentistry was established in Japan. Seven (7) honorable presidents have made significant contributions to the AAPD.

I would like to express my sincere thanks and respect to Dr. O Sakai, Dr. Kim J.B, Dr.Zhang. B.X., Dr. Phantumbanit, Dr. Chawla, Dr. Bahar and Dr. Watanabe who have lead this Academy with dignity and success.

Although they have made many accomplishments, there still remain many things to take care of in order to promote upgrade our Academy.
We have to establish our regulations or by-laws to standardize our activities, as well as to prepare the Academic Conference focusing on preventive dentistry and public oral on a grand scale and encourage more active participation in our Academy.

Therefore, to this end, it is my desire in preparing the 8th International Conference that we have many active, prominent speakers join in on symposiums focusing on the clinical and academic areas in our fields.

Otherwise, I think it unfortunate that we have no opportunities to have key-note speeches.

I have re-produced the AAPD flag and set-up an Internet homepage for the AAPD, in order to establish the Academic form, symbol, and correspondence with each other.

Additionally, it's my great honor to have this chance to hold the Asian Fluoride Commission Meeting in Seoul.

I hope, all our efforts will bear fruit to help us further develop the preventive dentistry field, in co-operation with each of our respective countries.

Thank you.

Shin Seung-chul

Shin, Seung Chul DDS.Ph.D
President.
Asian Academy of Preventive Dentistry

8TH. CONFERENCE OF ASIAN ACADEMY OF PREVENTIVE DENTISTRY

Advising Committee

Dr. Kim, Ju -Hwan:
former Dean of Seoul National University former President of Korean Dental Health Association

Dr. Kim, Jong-Bai:
Emeritus Professor of Seoul National University former President of Korean Dental Health Association

Dr. Kim, Dong- Ki:
Vice President of Korean Dental Association President of Korean Academy of Clinical Preventive Dentistry

Dr. Choi, Yu- Jin:
Emeritus Professor of Kyunghee University former President of Korean Academy of Oral Health

Dr.Yoon, Heung-Rvul:
former President of Korean Dental Association former President of FDI

Dr. Lee, Jae-Hyun:
former Dean of Dankook University / former President of Korean Dental Health Association

Dr. Paik, Dai-il:
Professor of Seoul National University
President of Asian Society of Oral Breath 8th. President of AAPD
President of Korean Dental Health Association

Organizing Committee

Dr. Shin, Seung-Chul:
former Dean of Dankook University Professor, Department of Preventive Dentistry,
Department of Oral Health, Dankook University

General Affairs
Dr. Cho, Eung-Hwee: Director of Dr. Cho's Dental Clinic

*** Planning**
Dr. Jung, Young-Bok: Director of Dr. lung's Dental Clinic
General Affairs, Seoul, Dongiak Dental Association President, Dr. Prevent Company

Finance
Ms. Chang, Yean-Soo: Lecturer, Dept.of Oral Health Graduated School of Dankook University Director,
Dental Expert Nurse Academy

Publishing
Dr. Cho, Ja-Won : Assistant Professor of Dankook University, Chairman of Clinical Dept. Preventive Dentistry

Academic Affairs
Dr. Park, Yong- Deuk: Assistant Professor of Kyunghee University Academic Affairs of Korean Academy of Oral Health

Advertizing
Dr. Jin, Bo-Hyoung Associate Professor of Seoul National Univ.

Editing
Dr. Park, Deok-Young Dean, College of Dentistry, Kangnung University.

Others: Some Active persons will be joined together one by one,
Most of Department of Preventive Dentistry, Dental Schools will support