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THE EFFECT OF VARIOUS FLUORIDE COMPOUNDS ON THE DEVELOPMENT OF EXPERIMENTAL ROOT SURFACE CARIES IN HAMSTERS

BY

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ABSTRACT

The aim of this investigation was to evaluate the effect of topical applications of various fluoride compounds on the development of root surface caries in hamsters. Male golden hamsters (n=115) were divided into 7 groups and were given a caries-promoting diet. Six groups were infected with *Actinomyces viscosus* ATCC 15987; and to 5 infected groups, distilled water (DW) and solutions containing 500 ppmF of acidulated-phosphate fluoride (APF), stannous fluoride (SnF_2), copper fluoride (CuF_2), and titanium tetrafluoride (TiF_4) were applied topically to the first mandibular molars once daily, 7 days a week, over a 24-week period. A grid method was used to assess the periodontal changes and root surface caries on the first molars. Plaque accumulation was reduced most by SnF_2; and alveolar bone loss was more reduced by SnF_2 and CuF_2. Root surface caries was significantly prevented in all fluoride groups when compared to an infected control group; and TiF_4 was the most effective fluoride compound followed by SnF_2, CuF_2, and APF. Root caries scores of the TiF_4 and SnF_2 groups were significantly lower than those of the DW group.

It was concluded that all fluoride compounds tested may prevent the development of root surface caries in hamsters.

Key words: Hamster, *A. viscosus*, Root surface caries, Prevention, Fluoride compounds

INTRODUCTION

The development of dental caries in elderly persons occurs mainly on the root surfaces (Beck [1], Surmont and Martens [2]). The occurrence, diagnosis and treatment of root surface caries has been attracting more attention as the elderly population increases (Nyvad and Fejerskov [3]). It is important that research on preventive measures be focused on controlling the development of this disease in the elderly (Bowen [4]).

It has been shown that fluoride reduces enamel demineralization by both *in vitro* and *in vivo* studies (Mellberg and Ripa [5]). The data on fluoride therapy to prevent root surface caries are recent and limited (Stookey [6]). A few studies using topical and systemic fluoride treatments in humans and animals have indicated an inhibitory effect on root surface caries (Newbrun [7]). Stamm and Banting [8] reported that the prevalence of root surface caries among life-long residents in a fluoridated area was about 50% less than that in a non-fluoridated area.

The caries-preventive effect on en-

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amel of topical fluoride therapy with sodium fluoride, acidulated-phosphate fluoride, stannous fluoride, and titanium tetrafluoride is well established (Wefel [9], Skartveit et al. [10]). A recent study has shown that copper fluoride has a caries-inhibitory effect when applied topically to the molar teeth of hamsters (Maltz and Emilson [11]).

In root surface caries studies, stannous fluoride, sodium fluoride, and acidulated-phosphate fluoride have shown anti-caries effects in rodents (Beiraghi et al. [12], Tsurumoto et al. [13]). Titanium tetrafluoride has been applied to exposed dentin of dogs (Tveit et al. [14]) and has shown promising anti-caries effects for root tissues *in vitro* (Hals et al. [15]). No report on the caries-preventive effects of copper fluoride or titanium tetrafluoride on root surface caries in rodents has been found in the literature.

The purpose of this study was to evaluate and compare the effect of daily topical applications of stannous fluoride (SnF_2), copper fluoride (CuF_2), titanium tetrafluoride (TiF_4), and acidulated-phosphate fluoride (APF) solutions at relatively low concentrations (500 ppmF) on the development of root surface caries in hamsters infected with *A. viscosus*, over a period of 24 weeks.

Materials and Methods

Animals and infection One hundred and fifteen 21-day-old male golden hamsters (Japan SLC, Inc.) were weighed and housed in cages made of solid plastic walls with a stainless steel bottom and a mesh lid (2–3 animals per cage). During one night, all the animals were given distilled water containing 0.02% streptomycin and were fed a solid diet CE-2 (Clea Japan, Inc.). On the following day, the animals were skin-marked and divided by littermate into 7 groups with balanced body weights. Caries-promoting diet 2000 (Keys and Jordan [16]) and tap water were given *ad libitum* to all hamsters. Afterwards, 6 groups were infected orally with strains of *Actinomyces viscosus* ATCC15987 and one group remained uninfected as a control (uninfected control).

Previously, strains of *Actinomyces viscosus* ATCC15987 suspended in Gam broth "Nissui"*[1] were incubated at 37°C for 48 hours. After incubation, the broth was centrifuged at 6000 r.p.m. for 5 minutes. The supernatant fluids were discarded, and the sediment solutions were repeatedly gently mixed and washed with sterilized 0.85% saline by means of a pipette, and centrifuged. A final solution with the bacterial strains (10^8 CFU/ml) suspended in saline was obtained and stored at −20°C.

Disposable mini-syringes were used to instill approximately 0.2 ml of the fluid containing the *A. viscosus* strains into the mouth and the cheek pouches of the animals. In addition, a stick applicator with a cotton pellet on the tip was also used to spread the fluid containing the bacterial strain in the molar region of the animals' mouth. These procedures were repeated daily for 10 successive days. Tap water was available *ad libitum* to all the groups, which were maintained at a room temperature of 23±2°C, air humidity of 60%, and light control (lights on from 6:00 a.m. to 8:00 p.m.). Cages were changed twice a week, the animals' weight measured weekly, and the diet and water intake monitored every other day.

A period of 10 days was allowed for recovery of the cariogenic bacteria. Plaque samples were taken by swabbing the molar teeth of the animals with a cotton pellet soaked in 1 ml of an aqueous solution of 1% peptone (pH7.0). The presence of *A.*

*[1] *Nissui Pharmaceutical Ltd., Tokyo, Japan*

viscosus was verified by the CFAT medium described by Zylber and Jordan [17].

Treatments When the animals became 6 weeks-old, the 6 infected groups were treated as follows. Four groups each received daily topical applications of solutions containing 500 ppmF of one of 4 kinds of fluoride compounds (Wako Pure Chemical Industries, Ltd., Tokyo): respectively, sodium fluoride (NaF) in the form of acidulated-phosphate fluoride (APF =1.11 g of NaF/l of deionized water +0.51 ml of 0.85% H_3PO_4, approximately pH 3.6); stannous fluoride (2.06 g/l of SnF_2, pH 2.8); copper fluoride (1.34 g/l of CuF_2, pH 5.4); and titanium tetrafluoride (0.82 g/l of TiF_4, pH 2.0). All fluoride solutions were prepared daily by dissolving each fluoride salt in 100 ml of deionized water (except for the APF solution which was prepared previously and stored in a plastic bottle). Of the other two infected groups, one received daily topical applications of distilled water (DW), and another remained untreated as a control (infected control). Immediately after the SnF_2, CuF_2, and TiF_4 solutions were prepared, topical applications with each of these 3 compounds, APF, and DW were performed once daily, 7 days a week. Disposable mini-syringes were used to flush 0.2 ml of each respective solution over the lingual cervical region of the left and right first mandibular molars for 10–15 seconds. After the topical applications were performed, all animals were prevented from eating and drinking for 40 minutes. These procedures were repeated for 24 weeks.

Sacrifices and evaluation methods All animals were killed by chloroform inhalation after 24 weeks of treatment with the topical applications. The animals' lower jaws were dissected out and the cervical region of the first mandibular molars was examined under a stereo-microscope at magnifications of 10× to 20× for any periodontal alterations below the cemento-enamel junction (CEJ). The chart illustrated in Fig. 1 was used to score the development of plaque accumulation, alveolar bone loss, and root surface caries in the first mandibular molars. The scoring chart was a modification of the grid method of Rosen et al. [18] for hamsters. The roots of the first mandibular molars were divided into equal sections as reported by Rosen et al. [18] and, coronoapically, the mesial, lingual, and distal surfaces, including the furcation, were divided into unit areas of about 0.15 mm^2 (0.3 mm×0.5mm of width). As shown in Fig. 1, on the root of the first molar, a total of 29 unit areas could be scored for the mesial, lingual, and distal surfaces (hereinafter these three surfaces will be referred to as lingual aspect). Previous results (Cordeiro [19]) had indicated that the lingual aspects of the first mandibular molars are most affected in the hamsters infected with strains of *A. viscosus*. For that reason, only the lingual aspects (Fig. 1: unit numbers 1 to 29) of the roots of the first molars of both left and right sides (a total of 58 unit areas per hamster) were assessed. The

Lingual View

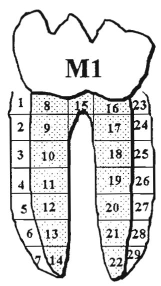

Fig. 1. Chart used for scoring plaque accumulation, alveolar bone loss, and root surface caries on the hamsters' first mandibular molars.

*[2] *Wako Pure Chemical Industries, Ltd., Tokyo, Japan*

evaluation of the unit areas affected by plaque accumulation, alveolar bone loss, and root surface caries was done under a stereo-microscope at magnifications of 10× to 20×. Each unit area was given a score of 1 only if more than half of the unit was involved on the root surface. A scale-drawing showing the contour of the roots of the first mandibular molar, as shown in Fig. 1, was inserted into the eyepiece of a stereo-microscope, and the cervical regions of the first molars were examined by adjusting the scale-drawing to the image of the first molar at the mesio-distal contour of the crown on the cervical region.

Plaque Accumulation Immediately after each animal was killed, their jaws were dissected out, slightly rinsed in tap water to remove the blood, and stored moistened in sealed plastic jars. Afterwards, the lower jaws were split and each mandibular quadrant was stained with a 1% solution of Neutral Red*[2] (Takashima [20]). Then, each stained quadrant was immersed and shaken for 10 seconds in water, dried with absorbent paper, and puffed with compressed air. Both mandibular quadrants were then mounted on blocks of utility wax fixed to glass Petri dishes and examined under a stereo-microscope at magnifications of 10× to 20×.

The first mandibular molars were examined with a stereo-microscope fitted with a scale-drawing inserted into its eyepiece. The scale-drawing was adjusted to the mesio-distal contour of the crowns on the cervical region of the first molar (Cordeiro [19]). When gingival recession was present, the number of unit areas affected by plaque accumulation between the cemento-enamel junction (CEJ) line of the scale-drawing and the internal margin of the gingiva receded apically on the root surfaces was recorded. A unit area was given a value of 1 only if more than half of the unit was affected by plaque. For each experimental group, the plaque scores were calculated by dividing the total number of unit areas affected by plaque accumulation (on the left and right sides of the hamster mandibles) by the number of animals per group. Student's t-test was used for the statistical analysis between groups.

Alveolar bone loss and root surface caries After the plaque accumulation had been scored, the lower jaws were autoclaved and soft tissues and debris removed and cleaned with a small soft toothbrush under tap water. Then, the cleaned jaws were placed into small bottles containing a 0.06% solution of Murexide*[3] (Navia [21]) and stored in a dark room for 24 hours. Later, the Murexide-stained samples were washed once in tap water and dried in an oven at 50°C. Afterwards, all samples were examined under a stereo-microscope fitted with a scale-drawing inserted into the eyepiece as described previously. Alveolar bone loss was scored by examining the unit areas exposed on the root surfaces between the CEJ and the level of the bone destroyed coronoapically. At the same time, root surface caries was scored only if a unit area on a root exposed by bone loss was stained red and could be penetrated by an explorer (Rosen et al. [18]).

For each experimental group, the bone loss scores were calculated by dividing the total number of unit areas exposed by bone loss (on the left and right sides of the hamster mandibles) by the number of animals per group. Likewise, the root caries scores were calculated as the average number of unit areas affected by caries on the lingual aspects (left and right sides) of the roots of the first molars for the hamsters of each experimental group. The scores for alveolar bone loss and root surface caries were analyzed statistically

*[3] *See footnote *2, page 107*

using the Student's t-test. In addition, to evaluate the development of root surface caries in the animals, the chi square test was used to compare the numbers of mandibular quadrants that developed carious lesions on the roots of the first molars in each experimental group.

Results

Animals' growth conditions The mean weight gain and number of animals killed after 24 weeks of treatment with the topical solutions are shown in Table 1. No significant differences in weight gain were found among the animals in the experimental groups.

Plaque Accumulation Plaque was observed affecting the lingual aspects of the roots of the first mandibular molars in the hamsters of all the 6 infected groups. Nevertheless, the groups treated with fluoride compounds showed lower plaque scores than did the infected control and DW groups. In the uninfected control group, no plaque accumulation was found. The plaque scores and standard deviations (±S.D.) for the lingual aspects (total 58 unit areas for the left and right sides) of the roots of the first mandibular molars in the hamsters of each group were as follows: uninfected control (0), infected control (4.13±4.35), DW (2.00±4.27), APF (0.94±1.88), SnF_2 (0.56±0.96), TiF_4 (1.63±3.03), and CuF_2 (0.68±2.75). The plaque scores for the lingual aspects of the roots of the first molars in all the experimental groups and their statistical relationships are illustrated in Fig. 2. The plaque scores for the animals treated with SnF_2 and CuF_2 were significantly lower than those of the infected control group's. There were no significant differences between the plaque scores of the animals treated with TiF_4 and those of the infected control group's. As shown in Fig. 2, plaque accumulation was also decreased in the DW group as compared with the infected control group, but no significant difference was found between these two groups. When the plaque scores of the DW-treated animals were compared with those of the fluoride-treated animals, only the plaque scores of the animals in the SnF_2 group were significantly lower than those of the DW group's (Fig. 2). There were no significant differences when the plaque scores of the 4 fluoride-treated groups were compared with each other.

Alveolar bone loss Alveolar bone loss occurred in all the animals. The mean bone loss scores and standard deviations (±S.D.) for the lingual aspects of the roots of the first mandibular molars of the hamsters in each group were as follows: uninfected control (10.44±0.89), infected control (18.50±4.50), DW (16.38±3.58), APF (14.00 ±2.61), SnF_2 (11.44±1.71), TiF_4 (12.63

Table 1. Weight gain(g) (mean±S.D.) after 24 weeks of treatment and number of animals per group.

Experimental Group	Infection with *A. viscosus*	Number of hamsters	Initial weight	Weight gain
Uninfected control	−	16	39.4±2.09	126.7±12.36
Infected control	+	16	39.4±2.53	128.8±8.47
DW	+	16	39.5±2.57	120.7±13.87
APF	+	16	39.6±2.91	124.9±10.32
SnF_2	+	16	39.6±1.52	120.6±11.97
TiF_4	+	16	39.6±2.68	129.3±10.89
CuF_2	+	19	39.6±2.75	121.0±9.36

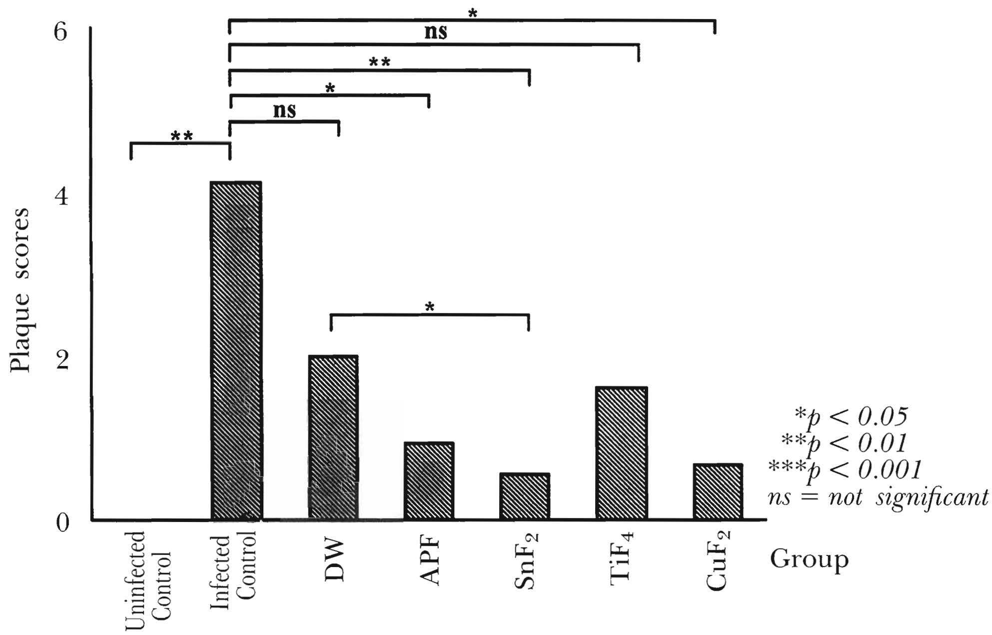

Fig. 2. Plaque scores for the lingual aspects of the first mandibular molars in each group.

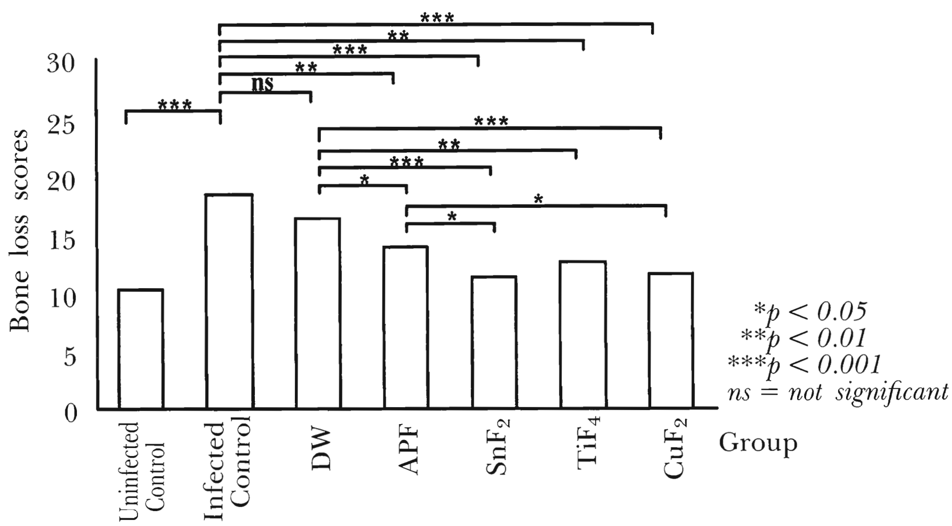

Fig. 3. Alveolar bone loss scores for the lingual aspects of the first mandibular molars in each group.

±2.63), and CuF_2 (11.63±3.35). The bone loss scores for the lingual aspects of the roots of the first molars in all the experimental groups and their statistical relationships are illustrated in Fig. 3. As shown in Fig. 3, except for the DW group, significant differences were found between the bone loss scores of the animals of the infected control group and those of the other experimental groups. The bone loss scores of the 4 fluoride-treated groups were lower than those of the infected control and DW groups. Alveolar bone loss was also significantly reduced in the fluoride-treated animals as compared with those of the DW-treated animals. Moreover, among the fluoride-treated groups, significant differences were found between the APF and the SnF_2 groups, and between the APF and the CuF_2 groups. There were no significant differences between the APF and the TiF_4 groups. The alveolar bone loss scores of the SnF_2, CuF_2, and TiF_4 groups were not significantly different when compared with each other.

Root surface caries Root surface caries developed in the animals in all the infected groups, except for those in the TiF_4 group. For each experimental group, data and statistical analyses upon the development of root surface caries on the mandibular quadrants (lingual aspects of the first molars only) are summarized in Table 2. Out of the 32 mandibular quadrants per infected group (38 quadrants for the CuF_2 group), the number of quadrants with carious lesions on the roots of the first molars were 17, 10, 6, 1, 0, and 1, respectively, for the infected control, DW, APF, SnF_2, TiF_4, and CuF_2 groups. In the animals of the uninfected control group, no carious lesions were found at the end of the experiment. The numbers of carious quadrants in the animals of the fluoride-treated groups were significantly lower than those of the infected control's. No significant differences were found between the numbers of carious quadrants of the infected control and the DW groups. When the numbers of carious quadrants of the DW group were compared with those of the fluoride-treated groups, significant differences were found between the DW and SnF_2, DW and TiF_4, and DW and CuF_2 groups. No significant difference was found between the DW and APF groups. When the fluoride-treated groups were compared with each other, there were significant differences between the numbers of carious quadrants of the APF and SnF_2, APF and TiF_4, and APF and CuF_2 groups (Table 2).

The root caries scores and standard deviations (±S.D.) for the lingual aspects (left and right sides) of the roots of the first mandibular molars in each group were as follows: uninfected control (0), infected control (4.75±4.19), DW(1.94±3.47), APF(0.63±1.15), SnF_2(0.06±0.25), TiF_4 (0), and CuF_2(0.47±2.06). The root caries scores of each group and their statistical relationships are illustrated in Fig. 4.

Table 2. Development of root surface caries on the mandibular quadrants in each group after 24 weeks.

Experimental Group	Number of mandibular quadrants	Number of quadrants with root surface caries
Uninfected control	32	0
Infected control	32	17
DW	32	10
APF	32	6
SnF_2	32	1
TiF_4	32	0
CuF_2	38	1

Note: Chi test (x^2)=*p<0.05; **p<0.01; ***p<0.001.

There were significant differences between the infected control group and the 4 fluoride-treated groups; however, no significant difference was found between the infected control and the DW groups.

Although the root caries scores of the 4 fluoride-treated groups were lower in value than those of the DW group, significant differences were found only between the SnF_2 and the DW groups, and between the TiF_4 and the DW groups. There were no significant differences between the DW and the APF groups or the DW and the CuF_2 groups.

Discussion

Previous reports have shown that the prevalence of root surface caries increases with age (Banting et al. [22]). The development of root surface caries is also positively associated with periodontal disease and gingival recession (Al-Joburi and Koulourides [23]); as well as with coronal caries (Vehkalahti [24], Papas et al. [25]). It has been shown that life-long consumption of fluoridated water significantly reduces the prevalence of root surface caries (Stamm and Banting [8]; Burt et al. [26]). Al-Joburi and Koulourides [23] have suggested that topical fluoride therapy should be used in older people, or particularly in patients receiving periodontal treatment whose teeth show vulnerably exposed cemental or root surfaces. A few studies have shown that fluoride administered at low concentrations in the drinking water of rats (Rotelie et al. [27], Rosen et al. [28]), or hamsters (Stookey [29]) may significantly reduce root surface caries but not alveolar bone resorption. Topical fluoride therapy with solutions of SnF_2 (1000 ppmF) or NaF (1000 ppmF) have reduced root surface caries in rats, but only SnF_2 reduced alveolar bone loss (Beiraghi et al. [12]).

The aim of this investigation was to evaluate the effect of topically applied solutions of several fluoride compounds on the development of root surface caries in hamsters. The results of the present study showed that daily topical fluoride

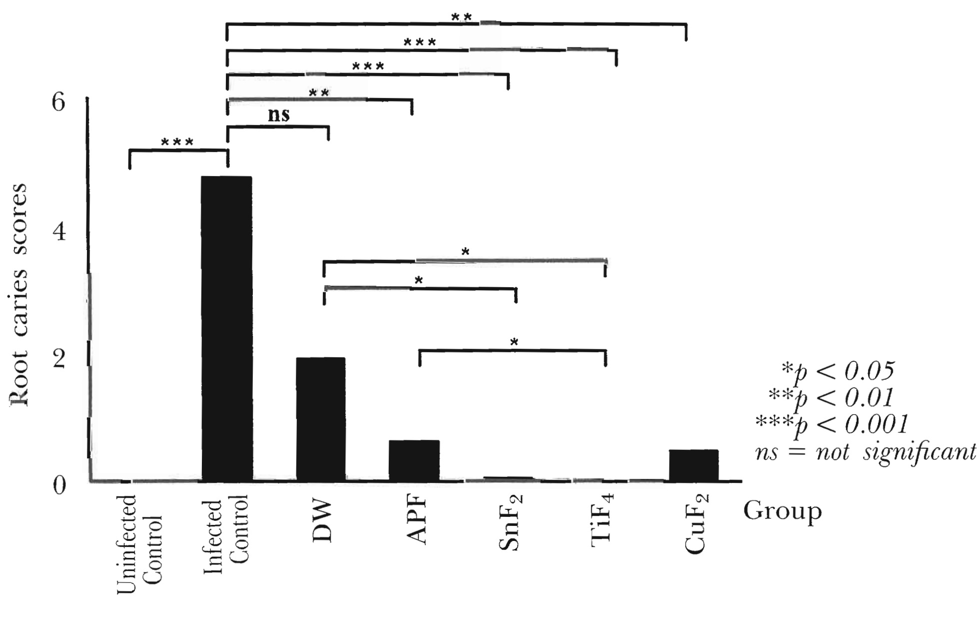

Fig. 4. Root caries scores for the lingual aspects of the first mandibular molars in each group.

therapy at relatively low concentrations (500 ppmF) may effectively reduce root surface caries in hamsters. In this study, although APF and CuF_2 significantly reduced root surface caries as compared with the infected control group, no significant differences were found when these two groups were compared with the DW group. In contrast, significant reductions of root surface caries were observed in the TiF_4 and SnF_2 groups as compared with the infected control and the DW groups.

As in the enamel, the penetration of fluoride into the root tissues will be greater if the fluoride concentration is increased and the pH is lowered (Hals et al. [16]), and if the frequency of applications is increased (Øgaard et al. [30]). In the present study, the groups of hamsters treated with solutions of lower pH (TiF_4: pH 2.0; SnF_2: pH 2.8) showed greater reductions in root surface caries than did the groups treated with solutions of higher pH (CuF_2: pH 5.4; APF: pH 3.6).

The mechanisms of action of topical fluoride compounds may vary greatly because of the cationic portion of these compounds or the pH of the treatment solutions (Wefel and Harless [31]). Tveit et al. [32] demonstrated that Sn and Ti metals form a glaze-like coating on the surface of the enamel or root surfaces. These authors also discussed the possibility of the Sn ion binding to Ca to create acid-resistant complexes.

In recent years, studies on titanium compounds have suggested that TiF_4 may have considerable potential as a caries-preventive agent (Stamm [33], Tveit et al. [14]). Hals et al. [15] studied the effect of TiF_4 (pH 1.0), NaF (pH 7.1), and APF (pH 1.0) solutions on root surfaces *in vitro* using electron microprobe analysis, microradiography, and polarized light microscopy. A glaze-like layer was found covering the surfaces of the TiF_4-treated specimens, and the fluoride uptake in the TiF_4 group was much greater than that in the NaF group. The fluoride uptake was also great in the APF-treated roots, but a marked demineralization subsurface was also found. These authors pointed out that topical application of TiF_4 may have advantages over that of NaF such as a higher uptake and penetration of fluoride, and a lower acid solubility of the root tissue. They also reported that, in the APF group, similarly to the TiF_4 group, an increased uptake and penetration of F may be attained without the occurrence of undesirable demineralization.

A similar *in vitro* study was conducted by Skartveit et al. [34] on the root surface reactions to solutions of TiF_4 and SnF_2. Root surface specimens were exposed for 1 min. and 4 min. to equimolar solutions of TiF_4 (native pH 1.0), SnF_2 (native pH 2.5), and SnF_2 (acidified pH 1.0) and examined by transmission electron microscopy (TEM). The results indicated that solutions of TiF_4 and SnF_2 at native pH caused only slight demineralization when applied topically to root surfaces.

It has been reported earlier that application of metal ions may increase the fluoride uptake on enamel surfaces, and Ti was reported to be a more potent complexer than Sn (MacCann [35]). Dérand et al. [36] observed a reduction of demineralization depths on the SnF_2- and TiF_4-treated roots. They reported that SnF_2 and TiF_4 formed depots in the cementum and increased the fluoride uptake on the treated root surfaces. From the results of the above reports, it is considered that the main preventive effect of fluoride on the development of root surface caries is to reduce demineralization of the root tissues. In the case of fluoride salts such as SnF_2, CuF_2, or TiF_4, the covering of the root surface by a metal ion coating will greatly reduce the demineralization of the root tissues.

In the present study, alveolar bone loss was significantly reduced in the 4 fluoride-treated groups as compared with the infected control and DW groups. Although treatment with TiF_4 was the most effective in reducing root surface caries, plaque accumulation was not significantly reduced as compared with the infected control group. The mechanisms of the TiF_4 action on plaque or alveolar bone loss are not yet understood.

CuF_2 has shown cariostatic effects in hamsters (Maltz and Emilson [11]), but its effect on root surface caries is still unknown. A few studies on plaque and oral bacterial metabolism have indicated an effect of Cu on reducing acidogenicity (Opperman [37]). Several studies have shown that metal ions such as Cu^{++} and Sn^{++} have bactericidal and anti-plaque properties (Scheie [38]). Opperman [37] has shown that divalent cations have major inhibitory effects on the formation of bacterial plaque.

Maltz and Emilson [39] showed *in vitro* that CuF_2 and SnF_2 are more bactericidal than NaF at the same fluoride concentration. Furthermore, they suggested that the inhibitory activity of NaF seems to be carried by the fluoride ion, while the metal component seems to play a major role in the bactericidal effect of SnF_2 and CuF_2. In the present study, plaque accumulation, alveolar bone loss, and root surface caries were significantly reduced in the hamsters of the CuF_2 group as compared with those of the infected control group. However, when the CuF_2 group was compared with the DW group, only alveolar bone loss was found to be significantly reduced. From the findings of previous studies (Costich [40]), it is possible that CuF_2 may prevent the periodontal disease and root surface caries. Nevertheless, further studies are required to understand the modes of action fo CuF_2, as well its toxicity, before it can be used in humans.

SnF_2 and NaF solutions have been the most frequently used anticaries agents in humans (Wefel [9]). Root surface caries studies in rodents have shown that topical applications of SnF_2 and NaF solutions at relatively high concentrations (1000 ppmF) reduce the severity of root surface caries (Beiraghi et al. [12], Rosen et al. [28]). However, reductions of alveolar bone loss were reported only when SnF_2 was used (Beiraghi et al. [12]), but not when NaF was used even at concentration of 5000 ppmF (Rosen et al. [28]). SnF_2 in the form of mouthrinse or dentifrice has been demonstrated to be effective in reducing plaque accumulation by metabolically disrupting plaque bacteria (Svanberg and Rølla [41]). Previous clinical trials have shown that SnF_2 prevented periodontal disease of adolescents (Boyd et al. [42]). Recent studies in humans have also shown that SnF_2 is effective as a plaque-inhibiting agent in the therapy of periodontal diseases in high risk patients (Newman et al. [43]). Maltz and Emilson [39] observed that actinomyces and streptococci species show similar sensitivity to SnF_2 and CuF_2 *in vitro*. In the present study, SnF_2 significantly reduced plaque accumulation, bone loss and root surface caries. It seems reasonable to consider that topical applications of SnF_2 remarkably reduced the plaque accumulation which resulted in lower levels of alveolar bone loss and prevented the development of root surface caries in the hamsters. This finding agrees with the significant reductions observed by Beiraghi et al. [12] for root surface caries and bone loss in rats treated topically with SnF_2 (1000 ppmF). They [12] also reported that SnF_2 may influence the development of root surface caries through remineralization and an antimicrobial effect of the stannous ion.

In this study, because 0.2 ml of each treatment solution was applied carefully once every day over the lingual cervical region of the first mandibular molar by flushing with a mini-syringe, even in the DW group plaque accumulation was reduced, but not significantly, as compared with the infected group. This reduction of plaque accumulation probably resulted in reductions of alveolar bone loss and root surface caries as compared with the infected control group.

In conclusion, the results indicated that daily topical applications of APF, CuF_2, SnF_2, and TiF_4 at relatively low concentrations (500 ppmF) may reduce the severity of periodontal disease and root surface caries in hamsters. However, collectively, the best benefits were observed in the SnF_2-treated group. Further studies on the effect of fluoride compounds upon root surface caries are required to develop means to enhance the preventive measures for the elder persons, particularly those with increased susceptibility to periodontal disease.

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