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Development and Testing of Bioadhesive, Fluoride-containing Slow-release Tablets for Oral Use

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Abstract—The bioadhesive characteristics of tablets for oral use made from modified starch, polyacrylic acid (PAA), polyethylene glycol (PEG) and sodium carboxymethylcellulose (CMC) were investigated. Adhesion force and energy were determined in-vitro and maximal adhesion time was evaluated in-vivo in human subjects. In-vitro, PAA showed the best bioadhesive properties, followed by modified maize starch and PEG with a mol. wt of 300 000–400 000 daltons. The presence of 0·1 mg of fluoride as NaF did not lead to significant differences in adhesion force and energy for the same formulation. The in-vivo bioadhesion was not strongly correlated to the in-vitro data. PAA, despite its excellent adhesion, proved to be irritating to the mucosa. PEG with a mol. wt of 200 000 daltons was subject to erosion. CMC showed good bioadhesive properties but the mechanical strength of the tablets was low. Modified maize starch tablets containing 5% (w/w) PAA and PEG with a mol. wt of 300 000 daltons proved to be the most suitable formulations for a fluoride-slow-release tablet with bioadhesive properties. In-vitro, the tablets released all of the fluoride within the 8 h period, with a high initial release. The release rate was related to the water absorption rate of the tablets. The PAA-containing formulations and the CMC formulations had the fastest release. In-vivo, fluoride levels with a minimum of 150 and a maximum of 1000 $\mu g\ mL^{-1}$ were maintained for 8 h in the oral cavity. These fluoride levels were sustained significantly longer than those obtained with the administration of fourfold the amount of fluoride in the form of a fluoride-containing toothpaste. The release characteristics in-vivo exhibited a high variation. The use of bioadhesive polymers in oral pharmacotherapy seems promising.

The beneficial effect of fluoride for the prevention of dental caries is well documented (Margolis & Moreno 1990) and several pharmaceutical forms have been developed to administer fluoride to individuals or population groups. Among these, mouth-rinses, gels, varnishes and tablets are the most widely used forms (Grøn 1977). Water fluoridation is effective (Backer-Dirks 1963) but not accessible for large populations, especially in western Europe, due to political controversies. The advantage of individual fluoride application is the possibility to adapt it to the needs of age, caries risk, and oral health problems.

Many fluoride preparations such as gels or varnishes contain high concentrations of fluoride which may be toxic, especially in children after accidental swallowing (LeCompte & Whitford 1985). They also lead to superficial calcium fluoride precipitation which is not regarded as the most effective fluoride-enamel reaction product (Arends & Christofferson 1990). As recent studies have shown that fluoride is effective in small concentrations when present long enough in the fluid surrounding the enamel (Margolis et al 1986), an oral slow-release fluoride administration seems to be the method of choice (McKnight-Hanes & Hanes 1986).

Using bioadhesive formulations as carriers for drugs such as fluoride offers the possibility of achieving that goal. Several polymers including cellulose derivatives (Gurny et al 1984), polyethylene glycol (Chen & Cyr 1970) and polyacrylic acid (Ponchel et al 1987) have been described as bioadhesive polymers. Experimental methods for the in-vitro evaluation of bioadhesive properties have been proposed (Duchêne et al 1988). The method described by Ponchel et al (1987) for the determination of the tissue-polymer bond force and energy in-vitro was used in this study to evaluate the adhesive characteristics of thermally modified corn starches, polyacrylic acid, polyethylene glycol of different molecular weights, and sodium carboxymethylcellulose, as well as some of their combinations. The same method was used to determine the influence of the addition of a small amount of fluoride on the adhesion. Additionally, the fluoride release was determined in-vitro. Finally, a clinical evaluation of these polymers on volunteers was performed. The maximal retention time of all the fluoride-free formulations was determined and the fluoride levels in saliva were determined after administration of two bioadhesive slow-release formulations and compared with the effects of a fluoride-containing toothpaste.

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Materials and Methods

Tablet formulation

The polymers used in this study are commonly used in the food or pharmaceutical industry. The formulations were prepared from drum-dried waxy maize (DDWM, mol. wt 4 000 000) and spray-dried waxy maize (SDWM, mol. wt 4 000 000) (Cerestar, Vilvoorde, Belgium), polyethylene glycol (PEG: mol. wt 2 000 000; PEG 750: mol wt 3 000 000; PEG 3000: mol. wt 4 000 000; PEG 301: mol. wt 4 000 000 and PEG Coagulant: mol. wt 5 000 000, Polyox, Amerchol, Vilvoorde, Belgium), polyacrylic acid (PAA, Carbopol 934P: mol. wt 3 000 000, W. F. Goodrich Co, Cleveland, OH, USA) pure or with hydroxypropylmethylcellulose (HPMC, K4M, Colorcon, Orpington, UK) and sodium carboxymethylcellu-

lose (CMC, Tylose 1000: mol. wt 1000 and Tylose 10000: mol. wt 10000, Tylose, Hoechst, Frankfurt, Germany). The formulations are summarized in Table 1. The polymer powders were blended in a Turbula mixer (type T2A, W. A. Bachofen, Basel, Switzerland) for 5 min. Tablets of 150 mg without or with 0·1 mg sodium fluoride (p.a., Merck, Darmstadt, Germany) were prepared. They were compressed using 7 mm flat punches at a pressure of 150 MPa on an eccentric compression machine (Korsch, type EKO, Frankfurt, Germany).

Swelling rate

The swelling rate of the tablets was evaluated for six tablets of each formulation. These were weighed and placed separately in a preweighed basket made of stainless steel mesh and dental matrix tape (Dentaurum, Pforzheim, Germany). This basket was placed in a plastic vessel containing 4 mL of demineralized water. The whole was stored in a water saturated atmosphere at room temperature (22°C). The weight was determined at 0·5, 1, 2, 4, 6, 8, 24 and 48 h. The swollen weight/initial weight ratio was calculated and the parabolic weight-ratio/time curve was linearized. The swelling rates, determined as the slope of the plots, were converted to relative swelling rates with the value of 1 assigned to the slowest swelling polymer (PEG80).

In-vitro determination of bioadhesion

Bioadhesion was determined on porcine attached gingiva (Bottenberg et al 1989). In this area the mucous membrane of the attached gingiva and the periosteal layer are closely connected to a tissue with a very high mechanical resistance. The tissues were taken from pigs after slaughter and stored at −80°C until use. Before the experiments the tissue was thawed and kept in a phosphate buffered saline solution (2·38 g Na_2HPO_4, 0·19 g KH_2PO_4 and 8·0 g NaCl 1000 mL demineralized water, all reagents p.a. grade). For the in-vitro adhesion test the apparatus of Ponchel et al (1987) was applied. This consisted of an Instron 1026 universal testing instrument equipped with a #A 30-1001 type 2512-115 load cell (Instron Ltd, High Wycombe, UK). The data were recorded on an x-y plotter. The equipment was located in an air-conditioned room at 25°C and 60% relative humidity. A piece of mucosa was glued to a bronze stud with cyanoacrylate adhesive (Framet Loctite, Ireland). The tablet was glued with the same adhesive to a teflon support (burette cock) fitted into a tapered glass cone welded to the bottom of a 300 mL glass beaker. The mucosa and the tablet were then pressed together with a force of 0·5 N for 5 min. After initial contact the beaker was filled with the buffer solution in order to wet the mucosa and to act as a counterweight. Then tablet and mucosa were pulled apart with a crosshead speed of 5 mm min^{-1} until complete rupture of the tablet-mucosa bond. A force vs time diagram was constructed. The adhesive force could be read directly from the recording and the energy of adhesion was calculated as the area under the curve.

Surface pH of the tablets

The surface pH of the tablets was determined to investigate possible side-effects of the tablets in-vivo. The tablets were allowed to swell for 2 h in 1·0 mL of demineralized water (pH 6·3 ± 0·06). A combined glass pH electrode (Ingold, Urdorf, Switzerland) was brought into contact with the tablet and pH was measured after 1 min equilibration.

In-vivo evaluation of the adhesive behaviour

The adhesion time and the behaviour of the tablets were determined in a blind cross-over study in which 20 healthy volunteers (10 male, 10 female, ages ranging from 21 to 42 years) participated. Only the fluoride-free tablets were used in the clinical trial. The formulations were identical to those used for the in-vitro study. Informed consent from the volunteers and the approval of the medical ethics committee of the Medical Faculty, Free University of Brussels were obtained. Before the study, the salivary flow rate was determined in order to evaluate a possible relationship with the adhesion characteristics (Tucker et al 1989). Flow rate was measured unstimulated and stimulated by chewing on a piece of parafilm (American Can Company, Dixie, CT, USA). Saliva was collected over 10 min in a graduated polyethylene tube (Falcon 2070, Becton-Dickinson, Lincoln Park, USA). The volunteers received the tablets in numbered glass vials together with a written instruction sheet. They were instructed to test one tablet per day and to insert the tablet preferentially in the morning. One tablet at a time was placed on the attached gingiva in the region of the upper canine. The tablet was not moistened before application and had to be pressed onto the mucosa for about 30 s. Then the tablet and upper lip were moistened with saliva by tongue movements. The volunteers were asked to record the time of tablet insertion and the time and circumstances of the end of adhesion (erosion or detachment of the tablet). An index was used to describe the side effects of the tablets: 1, no irritation or hindrance; 2, taste alteration, dry mouth or excessive swelling of tablet without pain; 3, slight pain or irritation of the mucosa; 4, severe irritation or pain which necessitated removing the tablet; 5, severe pain or irritation resulting in a small mucosal lesion. When a tablet had to be removed due to irritation, this case was not taken into account in calculating the mean maximal adhesion time. In the case of mucosal irritation the volunteer was allowed to interrupt the experiment until the lesion had disappeared.

In-vitro fluoride release

The kinetics of fluoride release was determined using the dissolution method according to the United States Pharmacopoeia XXI (1985) with four tablets of each formulation. The dissolution apparatus consisted of four round-bottom polypropylene flasks placed in a warm water bath at 35 ± 1°C. A stirring apparatus was used which drove simultaneously 4 paddle-type stirrers at a rate of 70 ± 1 rev min^{-1}. Since the current formulae for synthetic saliva all contain calcium which could lead to CaF_2 precipitation, the isotonic phosphate buffered saline solution described above was used. The fluoride content of this solution was below the detection limit. Samples of 200 μL were taken before the experiment, after 30, 60, 90, 120, 150, 180, 210, 240, 300, 360, 420 and 480 min, and the last sample was taken after 24 h.

The fluoride activity was determined using an ion-selective electrode (Orion 96-06) connected to a digital mV meter (Orion Ion analyser 901, Orion Research, Cambridge MA, USA). The samples were diluted with an equal volume of

TISAB (total ionic strength adjustment buffer, consisting of 1 M NaCl, 0·25 M acetic acid, 0·75 M sodium acetate and 0·05 M CDTA (*trans*-1,2 diamino cyclohexane-*N*,*N*,*N*,*N*,-tetra-acetic acid), all reagents p.a. grade, Merck, Darmstadt, Germany). A standard curve was established before each series of measurements with standards from 10 to 1000 $\mu g\ L^{-1}$ prepared from NaF in water/TISAB 1:1. The error of the method in this range of concentrations was 5%. Standards were measured regularly between measurements in order to detect electrode potential drift. The fluoride concentrations were calculated and cumulative release curves were established. In order to simplify the comparison between the different formulations and to facilitate statistical analyses, the curves were linearized using the Weibull distribution (Langenbucher 1972). This linearization procedure resulted in two numbers characterizing the release kinetics of each formulation: t_d (time after which 62·3% of the total fluoride was released) and β, which describes the form of the release curves ($\beta < 1$ describes an exponential, $\beta > 1$ a sigmoid release curve).

In-vivo fluoride release

Two formulations were tested in-vivo: DDWMC and PEG750, each containing 0·1 mg of fluoride as NaF (these formulations had the best results for adhesion time and the lowest irritation in-vivo). Sixteen healthy volunteers of either sex (11 male and 5 female, ages ranging from 20 to 35 years) participated in the cross-over study. Informed consent from the volunteers and permission from the medical ethics committee of the Medical Faculty, Free University of Brussels were obtained. Before the release experiment, salivary secretion rate was determined as described above. The volunteers were instructed to avoid the consumption of fluoride-containing food and drinks such as seafood or tea, and to use a fluoride-free toothpaste the evening before the experiment and the day of the experiment. During the experiment, eating and drinking was limited to a minimum and allowed only after one of the samplings. Toothbrushing or mouthrinsing was not allowed during the experiments. Before insertion of the tablet a saliva sample was taken for evaluation of the fluoride background level. The salivary fluoride background is reported to be fairly stable at all secretion rates (Shannon 1977). The tablet was placed on the attached gingiva in the region of the upper canine, held for about 30 s with slight pressure and then moistened with the tongue. Samples were taken 1, 2, 3, 4, 5, 6, 7, 8 and 24 h after the insertion of the tablet. Saliva was collected over 5 min in a graduated polyethylene tube and stored at −20°C until analysis. Salivary fluoride was measured with fluoride-specific electrode using the method of Ekstrand (1977); 225 μL of saliva were mixed with 25 μL of a concentrated TISAB buffer (7·5 M acetate buffer, pH 5·0 and 2% CDTA). Each sample was measured three times. A standard curve with NaF standards in water/TISAB 9:1 was established before each series of measurements. The variation in the salivary fluoride determination was 6%. Two groups of 6 volunteers received a fluoride-free tablet of PEG750 or DDWMC as a control. Saliva samples were taken before the experiment and then every 2 h for 8 h. The experimental conditions and the fluoride analyses were identical with the procedures described above.

To compare the bioadhesive slow-release tablets with a widely used method of oral fluoride application, eleven of the volunteers brushed their teeth for 2 min with about 0·4 g of a fluoride-containing toothpaste. The toothpaste contained 1000 mg kg^{-1} of fluoride as NaF in a silica gel/water/cellulose base. The amount of fluoride thus administered was about 0·4 mg. After the toothbrushing the volunteers rinsed their mouths for 10 s with 20 mL of demineralized water which was expectorated into a polyethylene tube. Saliva samples were taken during toothbrushing (1 min after the start of the experiment), after rinsing and after 10 min, 30 min, 1 h and every hour over 8 h and analysed as described above.

The initial fluoride concentration was subtracted from the measured value and the maximal salivary concentration (C_{max}) and the time to reach the maximal salivary fluoride concentration (t_{max}) was determined from the concentration-time graphs. The area under the curve (AUC) was calculated by the trapezoidal rule.

Statistical analysis

A one-way analysis of variance with a Tukey HSD multiple range test was used to evaluate statistically significant differences between the tablets for the adhesion force and energy and t_d measured in-vitro. For the in-vivo experiment, the same test was used to determine significant differences in adhesion time, the values for AUC and the salivary fluoride concentrations at each sampling. Multiple regression analysis was performed to evaluate the influence of variables related to the volunteers (age, sex and salivary flow) in-vivo adhesion and release study.

Results and Discussion

In-vitro bioadhesion

The force and energy of adhesion determined in-vitro are shown in Table 1. For the modified starch formulations, SDWM had the lowest adhesion force and energy, followed by DDWM, whereas the formulation containing 5% (w/w) polyacrylic acid (DDWMC) showed the highest adhesion force and energy values. The difference in adhesion force and energy between SDWM and DDWMC was significant ($P < 0·05$). The polyacrylic acid (PAA) formulations had high adhesion force and energy, with PAA100 showing significantly higher values than all other formulations in this study. Force and energy of adhesion gradually decreased with higher proportions of HPMC in the formulation. There was no significant difference in adhesion between PAA90 and PAA50. Considering the surface area of the tablets, the adhesion force and energy values were comparable with those reported by Ponchel et al (1987).

Among the polyethylene glycol formulations, PEG750 and PEG3000 had the highest adhesion force and energy, followed by the somewhat lower mol. wt PEG80. The forces and energies of adhesion of these three formulations differed significantly from those of the high mol. wt formulations PEG301 and PEGCoa. Optimal adhesion seemed to be reached at a mol. wt of 400 000, after which the adhesion force did not increase and adhesion energy even decreased. This finding is in agreement with the theoretical considerations reported by Peppas & Bury (1985). Both

Table 1. Characteristics of the formulations used in the in-vitro study. Bioadhesive force and energy are the results of 6 replicates ± s.d., t_d (time needed to release 62·3% of the total fluoride) was calculated with the Weibull distribution and is the mean value from 4 replicates ± s.d.

Formulation		Relative swelling rate	Adhesion force (N)	Adhesion energy (mJ)	t_d (min)
A. Thermally modified maize starch					
Drum-dried waxy maize	without F	2·0 ± 0·2	0·92 ± 0·28	0·30 ± 0·10	—
(DDWM)	with F	1·8 ± 0·2	1·17 ± 0·76	0·24 ± 0·14	138 ± 33
Spray-dried waxy maize (SDWM)	without F	2·3 ± 0·2	0·51 ± 0·25	0·18 ± 0·09	—
DDWM + 5% (w/w) Carbopol (w/w)	without F	2·7 ± 0·2	1·31 ± 0·44	0·44 ± 0·09	—
(DDWMC)	with F	3·2 ± 0·2	1·44 ± 0·26	0·33 ± 0·09	101 ± 13
B. Polyacrylic acid (Carbopol 934)					
Carbopol 100% (PAA100)	without F	5·9 ± 0·4	1·92 ± 0·52	1·11 ± 0·19	—
Carbopol 90% (w/w) HPMC 10%	without F	4·0 ± 0·3	1·10 ± 0·22	0·41 ± 0·09	—
(PAA90)	with F	5·2 ± 0·2	1·29 ± 0·41	0·35 ± 0·07	70 ± 2
Carbopol 50% (w/w) HPMC 50%	without F	3·1 ± 0·2	1·17 ± 0·28	0·24 ± 0·11	—
(PAA50)	with F	2·9 ± 0·3	1·47 ± 0·46	0·28 ± 0·14	104 ± 11
C. Polyethylene glycol (Polyox)					
Polyox WSR-N-80	without F	1 ± 0·1	1·15 ± 0·14	0·39 ± 0·11	—
(PEG80)	with F	1·6 ± 0·2	1·02 ± 0·24	0·29 ± 0·9	135 ± 14
Polyox WSR-N-750	without F	1·9 ± 0·2	1·29 ± 0·31	0·33 ± 0·18	—
(PEG750)	with F	1·7 ± 0·2	1·85 ± 0·58	0·44 ± 0·26	166 ± 45
Polyox WSR-N-3000	without F	2·5 ± 0·2	1·73 ± 0·34	0·23 ± 0·08	—
(PEG3000)	with F	1·7 ± 0·2	1·81 ± 0·17	0·26 ± 0·03	210 ± 32
Polyox WHR 301 (PEG301)	without F	3·7 ± 0·3	0·83 ± 0·27	0·09 ± 0·04	—
Polyox Coagulant (PEGCoa)	without F	3·3 ± 0·3	1·02 ± 0·24	0·17 ± 0·07	—
D. Sodium carboxymethylcellulose (Tylose)					
Tylose 1000	without F	6·2 ± 0·5	0·35 ± 0·15	0·11 ± 0·06	—
(CMC1000)	with F	5·8 ± 0·5	0·46 ± 0·14	0·13 ± 0·06	59 ± 4
Tylose 10000	without F	8·7 ± 0·5	1·18 ± 0·34	0·17 ± 0·05	—
(CMC10000)	with F	9·1 ± 0·5	0·93 ± 0·28	0·17 ± 0·06	75 ± 8

carboxymethylcellulose formulations had low values for adhesion force and energy, due mainly to fractures which could be observed within the tablets which occurred before the rupture of tablet-mucosa bond. Because of the possibly higher bioadhesive potential of CMC tablets, a statistical comparison with the other formulations would be misleading. CMC1000 had a lower adhesion force and energy than CMC10000. The latter polymer was easier to compress and tablets with a smooth surface and higher mechanical strength could be obtained. The adhesion force and energy were not significantly different after the addition of 0·1 mg sodium fluoride to the tablets, although some of the formulations showed a somewhat higher adhesion force (Table 1). The overall effect of a small amount of fluoride on the bioadhesive characteristics seems therefore negligible.

Although swelling ability of the bioadhesive formulation was said to be advantageous in obtaining a good polymer-mucosa interaction, no relationship could be found between a high swelling rate and bioadhesion in this study. The modified starch and polyethylene glycol formulations achieved high values of adhesion force and energy with a low swelling rate. The carboxymethylcellulose formulations had the highest swelling rates but achieved rather low values of adhesion force and energy. Swelling of the polymer contributed to the interpenetration of mucus and polymer and made bioadhesion possible (Peppas & Bury 1985). Too much water absorption in the polymer could lead to the formation of a low viscosity interface at the mucosal side of the tablet or within the tablet, and cause detachment (Chen & Cyr 1970).

Since all polymers evaluated in this study met the theoretical requirements for bioadhesion, being water-swellable long-chain polymers with polar components (Longer & Robinson 1987), it was not surprising that all formulations, except SDWM with a low adhesion force and energy and PEG301 and PEGCoa with a low adhesion energy, were effective bioadhesives in-vitro.

In-vivo bioadhesion

The mean adhesion times as well as the irritation reported by the volunteers are given in Table 2. Of the thermally modified starches, SDWM had the shortest adhesion time. A high proportion of the tablets were lost by detachment (7 out of 18 volunteers) and rapid erosion of the tablet surface by soft tissue (lip and cheek) friction was reported. The adhesion time of DDWM was not significantly longer. However, the DDWMC formulation had a significantly longer adhesion time than the other starch formulations. This was due to a higher erosion resistance and fewer cases of detachment (3 out of 14 volunteers). Irritation was not reported for the thermally modified starch formulations.

The PAA or PAA/HPMC formulations were able to adhere for longer than 9 h but caused irritation which resulted in a small mucosal lesion in some of the volunteers. Therefore these formulations were judged clinically unusable. Possibly the low surface pH of these formulations caused the irritation (Table 2). No significant differences in adhesion time could be found between the PAA or PAA/HPMC formulations. The formulations containing HPMC (PAA90 and PAA50) caused somewhat less frequent irritation. PAA-containing formulations had been studied extensively in-vitro and in animals (Ishida et al 1981; Ponchel et al 1987). One study with volunteers was reported for polyacrylic acid containing tablets (Ishida et al 1982), but these tablets contained lignocaine which might have masked

Table 2. Adhesion time of bioadhesive formulations determined in-vivo. The number of volunteers (n) who did not remove the tablet due to irritation is given in column 'valid'. The number of volunteers reporting no irritation (1) or some form of irritation (2 to 5) is also given.

Formulations	Surface pH	Adhesion time (min ± s.d.)	Total n	Valid n	Index of irritation (number of cases)				
					1	2	3	4	5
DDWM	4·6	320 ± 110	18	18	17	1	—	—	—
SDWM	4·7	263 ± 148	18	18	18	—	—	—	—
DDWMC	3·3	578 ± 213	14	14	13	1	—	—	—
PAA100	2·0	548 ± 173	16	9	4	2	3	3	4
PAA90	1·8	551 ± 172	17	9	4	5	—	5	3
PAA50	2·9	594 ± 182	18	13	5	4	4	4	1
PEG80	8·2	185 ± 81	18	18	14	4	—	—	
PEG750	7·8	344 ± 142	14	14	11	3	—	—	—
PEG3000	8·1	444 ± 138	16	16	9	6	1	—	—
PEG301	7·9	474 ± 241	18	18	8	7	3	—	—
PEGCoa	7·0	478 ± 236	13	13	7	4	2	—	—
CMC1000	5·3	312 ± 213	14	14	11	3	—	—	—
CMC10000	4·7	394 ± 211	14	14	6	6	2	—	—

pain and irritation. Despite its long adhesion time in-vivo, clinical application of polyacrylic acid in the oral cavity is not recommended based on the present findings. When used in small amounts (5% w/w) together with a non-irritating polymer such as DDWM, polyacrylic acid improves the bioadhesive qualities of the formulation.

Among the polyethylene glycol formulations, adhesion time increased with increasing mol. wt. The adhesion time of PEG750, PEG3000, PEG301 and PEGCoa were not significantly different from each other, but significantly longer than for PEG80 ($P < 0.05$). The PEG80 tablets adhered well (2 cases of detachment out of 18) but eroded rapidly in the mouth. This study could not confirm the description of Longer & Robinson (1987) of polyethylene glycol as a poor bioadhesive; however, these authors did not mention the mol. wt of the polymer. It should also be noted that no standardized test for bioadhesion measurements is available, which makes the comparison of the results of different studies difficult. Some irritation was reported for the PEG301 and PEGCoa formulations. The volunteers did not experience the same painful irritation as in the case of the PAA formulations but reported the feeling of a dry mouthed increased salivary viscosity and that the tablets turned into a viscous slime after several hours. For the CMC1000 tablets several volunteers reported a fracture in the tablet as observed in the in-vitro experiment and disintegration of the tablets during the manipulation. Nevertheless, a mean adhesion time of about 5 h could be obtained which was not significantly different from the higher mol. wt PEG formulations. The adhesion time of CMC10000 was not significantly longer than that of CMC1000 but the former formulation caused some irritation mainly by its high swelling which impaired the lip movements of the volunteers. The volunteers' stimulated and unstimulated salivary flow rate made no significant contribution to the adhesion times in the regression analysis. Furthermore, no significant regression coefficients were found for in-vitro adhesion force ($P = 0.07$) and energy ($P = 0.87$). Therefore the eventual in-vivo adhesion characteristics seemed not to be well related to the in-vitro adhesion measurements. Possible reasons for this finding were the erosion resistance of the tablets which was not tested in-vitro. For example PEG80 had an adhesion force and energy comparable with PEG750 but a significantly shorter adhesion time in-vivo.

It may be concluded that the in-vivo adhesion times and behaviour were satisfactory for the PEG750 and PEG3000 formulations and DDWMC. These values were higher than those of the drug-free gelatine/PAA tablets for oral use reported by Jacques et al (1989). A difference in tablet formulations and location (on the inner side of the cheek) may account for the observed difference. The authors reported, as in the present study, a high variance of adhesion times.

In-vitro fluoride release

The release curves in-vitro are shown in Fig. 1 and the parameters obtained with the Weibull transformation are presented in Table 1. The CMC and PAA-containing formulations had a release rate that was significantly higher than that of the other formulations. It seems reasonable to

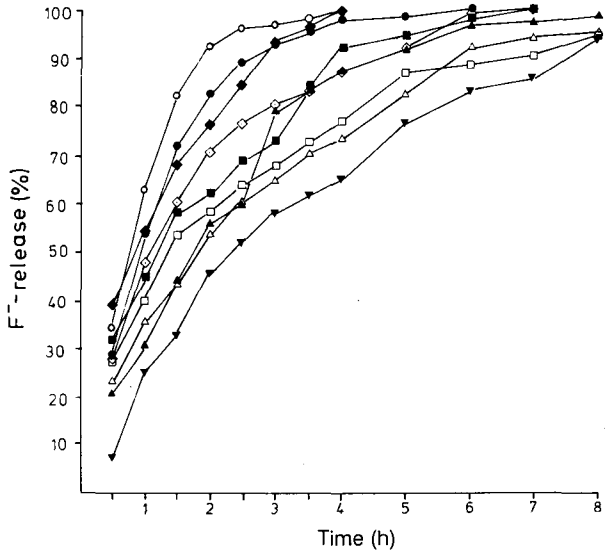

FIG. 1. Percent release of fluoride in-vitro (n = 4). □——□ DDWM; ■——■ DDWMC; ▲——▲ PEG80; △——△ PEG750; ▼——▼ PEG3000; ◇——◇ Carbopol 50%; ○——○ CMC1000; ●——● CMC10000.

assume that the release rate in-vitro was related to the swelling rate of the tablets. Once hydration of the polymer matrix is achieved, the fluoride is dissolved and can diffuse out of the polymer. Diffusion of fluoride through polymer hydrogels is reported to be very fast (Nelson & Farng 1972; Hattab & Lindén 1985). From all formulations tested, PEG3000 had the longest t_d, followed by the other PEG formulations in order of decreasing mol. wt, and the thermally modified corn starch. These formulations were all characterized by a low swelling rate. It can be concluded from these in-vitro results that PAA-containing formulations, because of their irritation and the fast release rate, are not suitable as a slow-release carrier for oral fluoride application and the CMC formulations are not suitable because of their porosity and consequently low mechanical strength. No differences between the different formulations could be obtained for the form parameter β.

In-vivo fluoride release

In-vivo fluoride concentration-time curves are shown in Fig. 2. Both bioadhesive tablets, DDWMC and PEG750 showed a minimum sustained fluoride concentration of 160 μg mL^{-1} and a C_{max} of 600 to 800 μg L^{-1}. The values of C_{max}, t_{max} and AUC are given in Table 3. The fluoride levels in the DDWMC group remained significantly higher than the initial level (40 ± 28 μg L^{-1}) from 1 to 7 h after tablet insertion. The fluoride levels after insertion of the PEG750 tablet were significantly higher than the baseline level throughout 8 h observation. No elevation of the salivary fluoride levels was observed with the DDWMC or the PEG750 control tablets (without fluoride). The t_{max} values of the bioadhesive tablets were ranked in the same order as the t_d values in the in-vitro experiment, with a somewhat faster release from the DDWMC formulation. No significant difference could be found for t_{max} and C_{max} between these bioadhesive formulations.

The toothpaste had the highest fluoride level during and directly after the 2 min toothbrushing: 105 ± 39 mg L^{-1} after 1 min of toothbrushing, $8{\cdot}4 \pm 6{\cdot}5$ mg L^{-1} after the rinsing and $2{\cdot}1 \pm 2{\cdot}0$ mg L^{-1} after 10 min. About half of the fluoride applied was lost with the rinsing ($0{\cdot}18 \pm 0{\cdot}05$ mg) and after half an hour the fluoride levels were decreased to 550 μg mL^{-1} (Fig. 2). The fluoride was cleared further and 2 h after application the salivary fluoride levels were significantly lower than the fluoride levels obtained with the slow-release tablets. After 6 h there were no significant differences from the baseline levels. The AUC from toothpaste was the highest because of the administration of the 4-fold amount of fluoride. The major part of the AUC (88%), however, was found in saliva between the start of the experiment and 0·5 h after administration. Fluoride was cleared rapidly; only a small part of the fluoride in interdental spaces or adsorbed to the mucosa might have formed a reservoir. The amounts of fluoride after toothbrushing were similar to the values reported by Bruun et al (1982). When compared with the toothpaste, both bioadhesive tablets had a greater ability to sustain an elevated fluoride concentration in saliva despite the administration of a smaller dose. Other topical application methods achieved higher salivary fluoride levels, but the dose could be up to 100 mg per application, for example with a fluoride gel (Ripa 1990). Furthermore, the fluoride levels are shown to decrease rapidly (Bruun et al 1982) despite the initially high concentrations.

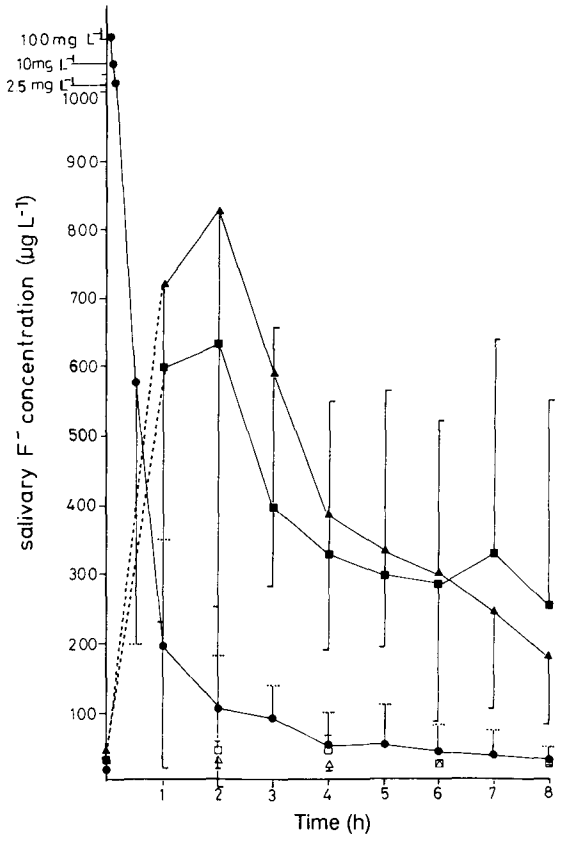

FIG. 2. Salivary fluoride concentrations obtained in-vivo with bioadhesive slow-release tablets (±s.d., n = 16), toothpaste (±s.d., n = 11) and fluoride-free bioadhesive tablets (±s.d., n = 6). ▲——▲ DDWMC (with fluoride); ■——■ PEG750 (with fluoride); ●——● toothpaste; △——△ DDWMC (fluoride-free); □——□ PEG750 (fluoride-free). ⌋ s.d. bars relating to DDWMC, ⌊ for PEG750, and ⌋⌊ for toothpaste. The upper part of the scale is logarithmic and refers to the salivary fluoride concentration obtained with the toothpaste.

The in-vivo results for the adhesion and release experiments varied considerably in contrast to the in-vitro experiments. This may have been caused by erosion of the tablets

Table 3. Results of the in-vivo fluoride release study, n is the number of participating persons.

Formulation	Amount of F mg	n	AUC μg h L^{-1} ±s.d.	t_{max} min ±s.d.	C_{max} μg L^{-1} ±s.d.
DDWMC tablet	0·1	16	1425 ± 618	139 ± 71	908 ± 583
PEG750 tablet	0·1	16	1262 ± 593	247 ± 154	840 ± 556
Toothpaste	0·4	11	2634 ± 1211	1	104550 ± 39010

by lip or cheek movements, since every individual had a different movement pattern and a different tendency to 'play' with the tablet. The erosion could also have contributed to the release by dislodging particles from the tablet surface, which because of their smaller diameter hydrated faster and released fluoride earlier. Other in-vivo studies with slow release devices also reported high interindividual variability: Ekstrand et al (1990) reported a high variance for the AUC after administration of a slow-release lozenge and Kula et al (1987) described a high variation in the salivary fluoride levels obtained with a slow-release device.

Compared with other experimental fluoride releasing devices in the form of a polymethylmethacrylate pellet cemented to a tooth surface (Mirth et al 1985), bioadhesive tablets had a shorter period in which the salivary fluoride level is elevated. The polymethylmethacrylate device was designed to release fluoride for several weeks but the application of bioadhesive tablets was easy and did not require sophisticated equipment or the preparation of the tooth-surface by acid etching. Furthermore, since the bioadhesive tablets eventually eroded totally, the removal of exhausted application devices was not needed. Both systems were able to maintain similar fluoride concentrations in saliva and bioadhesive tablets can be inserted repeatedly if a prolonged fluoride application is desired.

Some aspects of the possible caries-preventive action of these tablets need to be elucidated. For example, the fluoride was measured in whole saliva and did not reflect local variations of salivary fluoride concentrations in the mouth as described by Weatherell et al (1984) for the application of a conventional fluoride tablet. Consequently, the fluoride concentration may be much higher near the tablet than in other parts of the oral cavity. Also the efficiency of the fluoride-containing tablet in inhibiting or reversing initial carious lesions has yet to be tested. The salivary levels remained above 150 ng mL^{-1} during the 8 h period. To obtain a higher level of enamel protection, a permanent fluoride concentration of 500 ng mL^{-1} is desirable (Margolis et al 1986) and the fluoride dose and the tablet formulation would need to be adapted. The efficiency of long-term release devices has been shown experimentally (Corpron et al 1985; Mirth et al 1982) and a similar experiment using bioadhesive tablets will need to be performed.

We conclude that bioadhesive polymers such as thermally modified corn starch with 5% polyacrylic acid or polyethylene glycol (mol. wt 300 000) can be used as a slow-release device for fluoride. The system we describe is a positive step in the development of dose-efficient fluoride administration in the oral cavity.

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Patent

A patent for the application of modified starch as a bioadhesive slow-release drug carrier (E.P. Nr. 9087005.2-) has been applied for.

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