Effect on plaque of a xylitol-containing chewing-gum

A clinical and biochemical study

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The effects on plaque induced by single or repeated use of xylitol- and sucrose-containing chewing-gums were studied on twenty subjects refraining from tooth-brushing for three days. Mechanical removal of plaque by chewing during a single period was shown to yield no significant differences between gravimetric values obtained before and after chewing. Daily use of a xylitol-containing chewing-gum reduced by 47% the weight of plaque formed in comparison to a sucrose-containing product, and by 20—41% in comparison to conditions when no chewing-gum was used. The use of a xylitol chewing-gum induced a lower invertase-like activity in plaque extra cellular phase. Results at variance with the gravimetric data were obtained by using a stained plaque scoring system; the eventual inadequacy of such plaque scoring systems was discussed.

Key-words: Dental plaque; xylitol; sucrose; chewing-gum; invertase; plaque index

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It has been established that a partial substitution of sucrose by xylitol in the diet during four or five days reduced the amount of dental plaque and caused certain changes in its chemical composition (*Scheinin & Mäkinen*, 1971, 1972; *Mäkinen & Scheinin*, 1971, 1972). On the other hand, it appears that complete cleansing of the dentition cannot be expected from food mastication (*Wilcox & Everett*, 1963; *Löe*, 1971), or from the use of chewing-gum (*Möller & Poulsen*, 1973; *Bratthall et al.*, 1973). However, *Finn & Jamison* (1967) have observed that the surfaces of the posterior teeth appeared unusually clean after the use of a chewing-gum. Furthermore, chewing-gum can be considered as a good vehicle for an active agent as it allows a sufficient contact time for this one to be effective (*Ennever & Sturzenberger*, 1961; *Packman et al.*, 1963). In a recent short-term pilot study chewing a xylitol-containing product resulted in a reduction of the dental plaque formation when compared to the use of a corresponding sucrose-containing chewing-gum (*Mouton, Scheinin & Mäkinen*, 1974). The purpose of this paper was to further compare in a short-term experiment the effects of xylitol- and sucrose-containing chewing-gums on the formation and composition of dental plaque. The eventual effect of a single chewing period was also studied.

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MATERIALS AND METHODS

Twenty dental students, 5 males and 15 females, aged 20—25 years, acted as voluntary test-persons in a four-week investigation. Each test week, started by a two min tooth-brushing and was scheduled as follows:

- a test-period of three days during which oral hygiene was neglected and chewing-gums were used
- a clinical registration on the morning of the fourth test day
- a normalization period lasting for the three and half remaining days of the week during which the subjects had to resume their oral hygiene.

The arrangement of the study is described in Tables I and II. In each clinical registration two procedures were carried out: plaque collection and photography.

Table I. *The general arrangement of the study*

Day(s)	Procedure	Week			
		1	2	3	4
1	Tooth-brushing	+	+	+	+
1—3	No oral hygiene	+	+	+	—
	Chewing 6 pieces/ day	— Sucrose		— Xylitol	
4	Clinical registration	+	+	+	+
4—7	Normalization period	+	+	+	+

Table II. *Chronology of the clinical registration procedures*

Group I	*Group II*
Collection of plaque samples	Staining — Photographing
Staining — Photographing	Chewing (2 × 10 min)
Chewing (2 × 10 min)	Collection of plaque samples
Staining — Photographing	Staining — Photographing

Plaque collection. Plaque was meticulously collected within a period of 2.5 min, starting from the central incisor from the buccal aspects of teeth 18—17—16—15—14—13—12—11. The fresh weight of the plaque material (PFW) was immediately determined. The material obtained from each test-person was then suspended into cold (4° C) 0.9% sodium chloride solution.

Since one of the purposes of this study was to investigate whether one single chewing-period could yield any immediate change in the amount or composition of plaque, it was necessary to arrange for two kinds of plaque samples: samples obtained immediately before chewing, and samples obtained after a chewing period. A chewing period of 20 min, comprising two pieces of chewing-gum and two separate 10 min phases, was arranged in connection with the clinical registration. Consequently, the subjects were divided into two groups:

- Group I; 10 randomly selected persons, plaque being collected prior to chewing (Table II)
- Group II; 10 randomly selected persons, plaque being collected after chewing (Table II).

Photography. A disclosing solution (0.1% basic fuchsin in water was applied on the teeth 23—26, 33—36, using a pipette, followed by a mouth rinse with water. Both procedures were repeated once. A photograph was taken of the left side of the dentition of each subject, using a 35 mm camera (with focusing bellows and a f = 9, 1 : 4 Leitz lens, enlargement × 0.6), equipped with two Mecablitz 212 flashes. For photography, the head of the subject was in a fixed position using a modified head-rest also immobilizing the camera. A 35 Agfachrome 50 S Professional film was used. The method was

similar to that described by *Carlsson & Egelberg* (1965).

For each subject, during each clinical registration, the first picture was taken of the stained left side teeth before the 20 min chewing period; the second picture, preceded by a repeated staining, was taken after the 20 min chewing period. In order to avoid interference of the disclosing solution with the right side where plaque was to be collected, plaque samples were collected before the pictures were taken. In subjects of group II in which plaque was collected after the 20 min chewing period, some disclosing solution applied on the left side for the first picture, may have been transferred onto the right side, but no such cases were, however, visually detected.

Each picture (8 for each subject in the whole study) was assessed by the plaque index of *Quigley & Hein* (1962), as modified by *Bay*, *Kardel* and *Skougaard* (1967) with regard to the extent of plaque on the buccal aspects of teeth 23—26, 33—36. The sum of the scores thus obtained for each picture was divided by the number of teeth examined (a few subjects had missing teeth or crowns), giving a plaque score per tooth. Examination of the pictures was performed against a light background. A magnifying glass was used in all cases involving difficulties in the evaluation.

The activity of invertase-like enzymes of the water-soluble fraction of plaque was determined for each plaque sample as described earlier (*Mäkinen & Scheinin*, 1972) and expressed as specific activity (in μmole of liberated reducing sugars per min and per mg protein).

The chewing-gum pieces used had a weight of ca. 4 g and a volume of ca. 3 cm^3. The sugar content of each piece was 3 g sucrose (72%) or 2 g xylitol (50%).

Table III. *The chewing schedule for test periods 2 and 4; 6 pieces per day to be chewed*

At awakening	1 p. for 10 min.
After 1st meal	1st p. for 10 min.
	2nd p. for 10 min.
After 2nd meal	1st p. for 10 min.
	2nd p. for 10 min.
At bedtime	1 p. for 10 min.

During the testperiods 2 and 4, six pieces of chewing-gum were to be chewed each day according to the schedule of Table III.

RESULTS

The values obtained for the different data under investigation at the end of each test-period and for each group are shown in Table IV.

Gravimetric data. The means and standard deviations of plaque fresh weight at the end of each test-period are shown in Fig. 1. A single chewing period of 20 min was proved not to affect the amount of plaque, as established by performing a Kruskal-Wallis test for each test-period comparing the groups I and II. This allows for pooling the groups I and II in order to compare the amount of plaque formed during a three-day period without oral hygiene with or without the use of chewing-gum. A Friedman-test was thus applied to the pooled groups for each week. The test showed significant differences at the 0.05 level, meaning that there were differences between the weeks.

A pairwise comparison was then made. The amount of plaque formed during the test-period 1 (no oral hygiene for 3 days) and the test-period 2 (no oral hygiene for 3 days, chewing a sucrose-containing chewing-gum) was significantly higher at the 0.05 level (test of Wilcoxon and Wilcox) than the amount of plaque formed during the test-period 4 (no oral hygiene

Table IV. *Results from group I and group II obtained for the different parameters under investigation. PFW = plaque fresh weight in mg; QHI = Quigley & Hein plaque index; INA = invertase-like activity in μmole/(min × mg protein)*

	GROUP I			GROUP II		
TRIAL 1	PFW	QHI	INA	PFW	QHI	INA
Minimum	2.7	2.33	0.123	5.3	2.50	0.305
Maximum	18.8	4.25	0.505	22.5	3.75	0.578
Median	8.75	2.87	0.156	8.70	3.38	0.510
$\bar{x}$	10.23	3.04	0.224	10.66	3.30	0.477
S.D.	5.44	0.66	0.159	5.27	0.41	0.104
TRIAL 2						
Minimum	2.9	2.33	0.114	4.2	2.62	0.197
Maximum	22.6	4.14	0.347	19.6	4.00	1.004
Median	11.40	3.25	0.183	12.70	3.43	0.302
$\bar{x}$	11.24	3.25	0.204	12.09	3.40	0.406
S.D.	6.08	0.61	0.096	5.36	0.51	0.339
TRIAL 3						
Minimum	2.8	2.50	0.055	2.5	2.87	0.000
Maximum	25.1	4.42	0.306	13.9	4.33	0.375
Median	5.55	3.25	0.143	5.00	3.52	0.069
$\bar{x}$	8.56	3.36	0.152	7.02	3.54	0.116
S D.	7.41	0.64	0.106	4.52	0.50	0.148
TRIAL 4						
Minimum	1.9	2.67	0.010	1.3	2.75	0.039
Maximum	10.2	2.67	0.127	13.1	4.33	0.436
Median	5.45	3.18	0.078	6.05	3.75	0.114
$\bar{x}$	5.73	3.31	0.072	6.60	3.57	0.165
S.D.	2.86	0.58	0.045	3.71	0.55	0.158

for three days, chewing a xylitol-containing chewing-gum).

Table V gives the comparative results expressed as means and percentage differences of the PFW for the different test-periods. There was 47% less plaque formed when chewing xylitol than when chewing sucrose (significant) at the 0.05 level) and from 20% (not significant) to 41% (significant at the 0.05 level) less plaque when chewing xylitol than when using no chewing-gum. On the other hand,

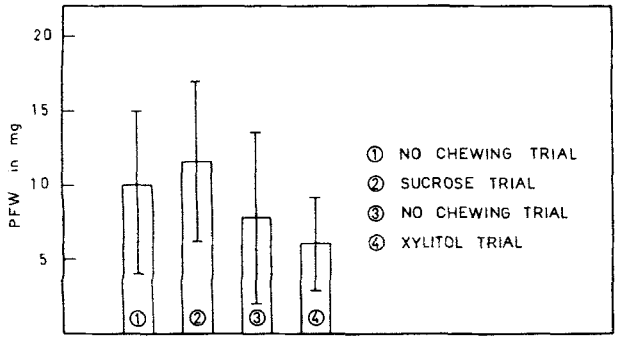

Fig. 1. The means and standard deviations of plaque fresh weight (PFW). Comparison between trial 1 and trial 4: $p < 0.05$; between trial 2 and trial 4: $p < 0.05$.

Table V. *Comparative results expressed as means and percentage differences of the plaque fresh weights (PFW)*

Trials	Mean PFW in mg		Effect in %	Significance
Xylitol versus Sucrose	Sucrose	11.67	47 % decrease	$P < 0.05$
	Xylitol	6.17		
Xylitol versus No chewing	No chewing (1)	10.45	41 % decrease	$P < 0.05$
	Xylitol	6.17	20 % decrease	Ø
	No chewing (3)	7.79		
Sucrose versus No chewing	No chewing (1)	10.45	11.5% increase	Ø
	Sucrose	11.67	49 % increase	$P < 0.05$
	No chewing (3)	7.79		

there was from 11.5% (not significant) to 49% ($p < 0.05$) more plaque formed when chewing sucrose than when using no chewing-gum.

A 25% decrease in the mean PFW between the test-period 1 (1st baseline) and the test-period 3 (2nd baseline) from 10.45 mg to 7.79 mg (significant difference at the 0.05 level) was also noticed in this investigation.

The plaque index. Table VI shows the range, medians and means of the values obtained when assessing the Quigley & Hein index at the pictures taken at the end of each clinical registration. An unexpected finding was that the highest values were obtained for the xylitol-chewing test period (trial 4), the lowest values for the first no-chewing period (trial 1), and intermediate values for the sucrose-chewing period (trial 2) and the second no-chewing period (trial 3). The differences were found significant with regard to the trials 1 and 3, and the trials 1 and 4, respectively (test of Wilcoxon and Wilcox, $P < 0.05$).

Table VI. *Range, medians and means of the Quigley & Hein plaque index values*

Trial	1 No chewing	2 Sucrose chewing-gum	3 No chewing	4 Xylitol chewing-gum
Range	2.33	2.33	2.50	2.75
	4.25	4.14	4.42	4.33
Md	3.19	3.25	3.37	3.43
$\bar{x}$	3.17	3.33	3.45	3.44

The invertase-like activity. For practical reasons, the invertase assay was possible for ten (out of 20) subjects only; five belonged to the group I (plaque collected before the 20 min chewing period), and five belonged to the group II (plaque collected after the 20 min chewing period). Table VII shows the results, expressed in liberated μmole of reducing sugars per min and per mg protein. The highest values were obtained when chewing sucrose (trials 1 and 2) and the lowest values when chewing xylitol (trials 3 and 4). Chewing sucrose chewing-gum for 20 min after a period of three days with restricted oral hygiene (trial 1, group II) yielded a higher invertase activity [0.447 μmole/(min $\times$ mg protein)] than no-chewing after the same period of restricted oral hygiene [trial 1, group I: 0.224 μmole/(min $\times$ mg protein)]. The difference was significant at the 0.05 level (two-tailed Mann-Whitney U-test).

The daily use of the xylitol-containing chewing-gum during a test-period of three days without oral hygiene (trial 4) yielded

Table VII. *Mean specific invertase-like activity in μmole/(min. × mg protein) for group I and group II*

Trial	1 No chewing		2 Sucrose chewing-gum		3 No chewing		4 Xylitol chewing-gum	
Group	I	II	I	II	I	II	I	II
	(*)	(*)	(**)	(**)			(**)	(**)
$\bar{x}$	0.224	0.477	0.204	0.406	0.152	0.116	0.072	0.165
S.D.	0.159	0.104	0.098	0.339	0.106	0.148	0.045	0.158

(*) significant difference at the 0.05 level between group I and group II (II > I)
(**) significant difference at the 0.05 level between trial 2 (sucrose chewing-gum) and trial 4 (xylitol chewing-gum)
($I_{Sucrose} > I_{Xylitol}$; $II_{Sucrose} > II_{Xylitol}$)

a substantially lower invertase activity than the use of the sucrose-containing chewing-gum in the same conditions (trial 2): 0.072 versus 0.204 μmole/(min × mg protein) for group I, and 0.165 versus 0.406 μmole/(min × mg protein) for group II, respectively (significant at the 0.05 level, test of Wilcoxon and Wilcox).

DISCUSSION

Concerning the gravimetric data, the differences recorded between the four trials, distributed in four subsequent weeks, deserves consideration. Comparisons between the trials on the whole indicate that least plaque was formed when a xylitol-containing product was used, most plaque was formed when a sucrose-containing chewing-gum was used, and intermediate plaque gravimetric values were obtained when no chewing-gum was used. However, the comparison between the two no-chewing trials, considered as baselines at the end of which one might have expected similar values in the PFW, showed a significant difference, consisting of a 25% decrease from the first baseline to the second. A possible explanation might be formed when considering the psychological aspect of such an experiment, extending over four weeks and affecting the behaviour of the test persons. These, consciously or not, may have reduced their carbohydrate intake, by restricting their consumption of pastry, sweets and other sucrose-containing products. Designing an experimental investigation should take in to consideration such as possibility.

The comparison between the PFW of groups I and II, i.e. comparison of PFW before and after a single chewing period (20 min duration) showed no significant difference. This substantiates the inability of chewing-gum *per se* to remove plaque from the tooth surfaces, or at least, not for sufficient oral hygiene purposes. On the contrary, the frequent use of a sucrose-containing chewing-gum will favour dental plaque accumulation as it involves an increased consumption of sucrose. Its harmfulness in habitual excessive use, owing to its sugar content has already been emphasized (*Fleisch*, 1942).

If the mechanical removal of plaque was not caused by gum chewing, the

decrease in plaque formation noticed when using a xylitol-containing product must have been due to some chemical property affecting, the bacterial growth or colonization. A significantly lower incidence of *Streptococcus mutans* (*Gehring et al.*, 1974); and a decrease of the acidophilic flora (*Larmas, Mäkinen & Scheinin*, 1974), corresponding to a lower plaque formation has been observed when substituting sucrose by xylitol in the diet (*Mäkinen & Scheinin*, 1974). The decreased plaque formation noticed in the present investigation, associated with the daily use of a xylitol-containing chewing-gum, may be related to the above microbiological background, therefore giving an explanation for a possible topical effect of xylitol on the growth of *Streptococcus mutans*. This would affect the synthesis of extracellular polysaccharides of the glucan-type, yielding a lower amount of plaque.

As earlier established (*Mouton, Scheinin & Mäkinen*, 1974), the inclusion of xylitol in a chewing-gum yielded a lower invertase-like activity in the plaque extracellular phase than a sucrose-containing product, or than the use of no chewing-gum. Furthermore, it was noticed in the present study that a single phase of chewing (20 min duration) yielded an immediate effect on plaque: the sucrose content of the chewing-gum significantly increased the invertase-like activity of the plaque extracellular phase.

The unexpected plaque index values in the present study (high score of stained plaque for the xylitol chewing trial; low score of stained plaque for the sucrose chewing trial), controversial with the gravimetric findings, must be seen against the background of the former being subjective and nonlinear. Discrepancies between a recording system recording the extent of stained plaque and assessment of the amount of plaque by collecting it and weighing it, have already been revealed and discussed by *Loesche & Green* (1972). These authors drew the conclusion that »ideally, in the assessment of plaque mass, both a weight measurement and an unstained plaque score should be obtained». Plaque indices such as this one used in the present study, designed for evaluating oral hygiene or for epidemiological studies, mainly deal with the area coverage of the tooth-surface by plaque material. If, according to *Carlsson & Egelberg* (1965), *Saxton* (1973) and *Gibbons & van Houte* (1973), the dynamics of dental plaque formation is related to 1) a colonization of the tooth-surface by specific microorganisms, 2) an increase in thickness of the bacterial mass by adhesion of new microorganisms, as well as by growth of the microorganisms already present, it is easily seen that any investigation on the formation of dental plaque should take into consideration, separately, the above two parameters. Quantitation using the Quigley & Hein plaque index, may provide precise information of the tooth-surface covered by stained plaque material, but must be considered as inadequate for assessing plaque mass. When an active agent, supposed to be effective on the second step of plaque formation is to be studied, an assessment of the tooth-surface area covered by plaque seems to be inappropriate. The method of choice will then be the method of assessing the plaque mass, e.g. a gravimetric recording, rather than any two-dimensional representation of a three-dimensional parameter.

In instances where plaque cannot be collected and weighed, the need for an assessment of the amount of plaque by a plaque index scoring system still remains. It appears that a scoring system taking the

aforementioned twofold dynamics of dental plaque formation into consideration should be developed.

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