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Saliva composition in three selected groups with normal stimulated salivary flow rates, but yet major differences in caries experience and dental erosion

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Abstract

Objective. It was hypothesized that, by comparing matched subjects with major differences in these dental diseases, but yet normal saliva flow rates, it would be possible to obtain data on the effect of saliva composition on dental disease isolated from the effect of the flow rate. Thus, the aim of the study was to compare the major physicochemical characteristics of stimulated whole saliva in three groups of 85 subjects, each with normal saliva flow rates and at least 24 remaining teeth. ***Materials and methods.*** A group with very little dental disease (healthy), a group with dental erosion (erosion) and a group with very high caries experience (caries) were chosen. Furthermore, the aim was to determine whether differences among groups could also be found on an individual level. ***Results.*** Although it was not possible to retrieve three groups whose members were completely identical, the present study points in the direction that, on a group level, subjects with very little dental disease seemed to have a more favorable physicochemical saliva composition with respect to higher calcium, phosphate, bicarbonate, pH, degree of saturation with respect to hydroxyapatite and a lower critical pH ($p < 0.05$ or less). However, on an individual level the explanatory power for the saliva composition was only 10% for caries experience and only 11% for dental erosion ($p < 0.001$). ***Conclusion.*** The compositional analyses performed in this study on stimulated whole saliva, including major physicochemical characteristics of saliva, will most likely have little predictive value for future dental caries and erosion in single individuals.

Key Words: *dental caries, dental erosion, chewing-stimulated whole saliva composition, calcium, phosphate, bicarbonate*

Introduction

Saliva is the fluid that constantly flows through the oral cavity while awake and, thereby, saliva affects nearly all processes between the enamel and the environment surrounding it. Therefore, much dental research has tried to determine whether saliva composition has an effect on the way that conditions and diseases of the enamel develop. Thus, many studies have tried to relate the saliva composition to dental erosion [1] as well as numerous studies relating saliva composition to dental caries [2]. However, most studies have not been able to pinpoint compositional salivary variables that clearly have an explanatory power on dental erosion and caries. This is most likely due to a high degree of functional redundancy for saliva, where one function often is supported by a concerted action from many different components within the saliva [2]. Therefore, most compositional variables probably will have to be determined simultaneously to obtain a more composite measure of the saliva capacity to avoid dental disease. Because the effect of the salivary flow rate is a major factor for dental disease [1,3] this parameter often becomes the primary determining factor for dental disease at the expense of weaker compositional variables of the saliva [4]. Nevertheless, reviewing a large number of studies has indicated that low salivary buffering capacity, calcium and phosphate concentrations and levels of secretory IgA (sIgA) show some link to increased caries [2].

For the present study we hypothesized that, by comparing age, gender and geographically matched dentate subjects with major differences in dental diseases, but yet normal stimulated salivary flow rates, it would be possible to obtain more valid data on the

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effect of saliva composition on dental disease. Thus, the aim of the study was only to focus on the saliva composition in healthy subjects with normal paraffin chewing stimulated whole saliva flow rates and to compare the major physicochemical characteristics of this saliva among groups that distinctively differed with respect to the two main disease of the dental enamel, namely dental caries and erosion. Furthermore, the aim was to determine whether differences obtained on a group level could also be found on an individual level.

Materials and methods

Study groups

The study groups were selected among a total of 4402 subjects participating in the oral part of the Danish Health Examination Survey (DANHES) that was performed in 13 municipalities in Denmark in the years 2007 and 2008 [5] and all data and samples for the present study were retrieved entirely from DANHES in this post-hoc analysis. The overall inclusion criteria were: (1) normal paraffin chewing stimulated whole saliva flow rate of 1 mL/min or more, (2) at least 24 remaining teeth and (3) a valid identification of the saliva sample in the biobank (DANHES) as well as a sufficient amount and quality of the saliva sample for analyses. Among 3122 subjects fulfilling the overall inclusion criteria, three groups were selected according to: (1) a group without dental erosion and with a very low caries experience (healthy group), (2) a group with dental erosion regardless of caries experience (erosion group) and (3) a group with a very high caries experience and no dental erosion (caries group).

The methods used for determination of decayed and filled surfaces as well as dental erosion have been described previously [5]. With respect to dental erosion, only subjects with clearly visible signs of the disease were diagnosed. Stimulated whole saliva flow rates were determined in response to chewing on 1 g of paraffin wax at a regular pace for 4 min. Saliva collected in the first minute was discarded and only saliva collected in the last 3 min was stored at 80°C. The saliva flow rate in mL/min was determined by weighing the disposable plastic cup before and after the saliva collection, presuming that 1 g of saliva was equivalent to 1 mL [6].

Among the 3122 subjects who fulfilled the overall inclusion criteria, only 85 subjects were identified with clearly visible dental erosion. Therefore, the

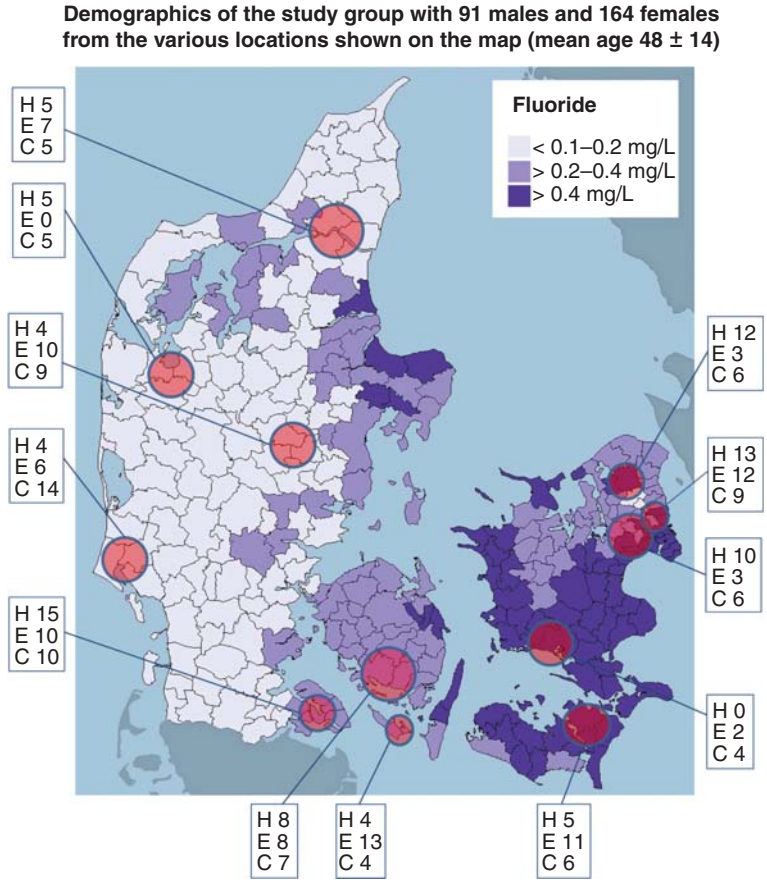

Figure 1. Demographics of the study population showing the distribution of subjects who were dentally healthy (H), subjects with dental erosion (E) and subjects with a very high caries experience (C) across the land of Denmark. Two municipalities were lying side-by-side and are shown as one in the figure. Geochemical differences in the drinking water fluoride content (mg/L) are illustrated by different shades of blue.

erosion group became the determining group for selection of subjects for the other two groups. When selecting subjects for the healthy and caries groups, great care was taken to match the subjects in these groups with the erosion group according to age, gender and geographic location of the subjects, so that geochemical differences in drinking water composition did not become a determining factor for dental disease (Figure 1). The groups were selected by hand by ranking all subjects that matched a specific subject in the erosion group on their caries experience. Subjects for the caries group were selected among those with the highest caries experience and subjects for the healthy group among those with the lowest caries experience. By these means three groups of 85 subjects in each were identified, comprising 255 subjects in total (Table I). Data on drinking water composition were retrieved from the Geological Survey of Denmark and Greenland (GEUS) as weighted averages according to the production and composition in each waterworks in each municipality [7].

The study was conducted in accordance with the Helsinki Declaration and approved by the Ethical Committees for the Region of Copenhagen (H-C-2007-0118). Written informed consent was obtained from all participants and the establishment of a biobank was approved by the Danish Data Protection Authority (2007-41-1567).

Compositional analyses of whole saliva

The salivary concentrations of sodium and potassium were determined by atomic absorption spectroscopy (AAS) in the emission mode at 589.0 nm and 766.5 nm, respectively. The salivary calcium concentration was determined by AAS in the absorption mode at 422.7 nm with KCl and $SrCl_2$ in the matrix for reduction of oxysalts. For all AAS analyses, samples were diluted in Millipore water containing 1% analytical grade nitric acid. For sodium and potassium, samples were diluted 1000-fold and, for calcium, samples were diluted 100-fold. Salivary chloride, total phosphate, total protein and amylase activity were determined by colorimetric methods. Briefly, chloride was determined by the mercury-chloride/iron-TPTZ reaction at 610 nm [8], total phosphate by the molybdenum reaction at 700 nm and total protein by the Lowry method at 750 nm (μg/mL). For total protein, the standard curve was obtained from lyophilized unstimulated and paraffin chewing stimulated whole saliva proteins from multiple ($n > 100$) human subjects [9]. Amylase activity, which is the catalytic activity of the enzyme, was determined with the Phadebas amylase test kit as U/mL. All samples were measured at least twice. When a sample exceeded the calibrations in any of the analyses, dilution of the sample was performed until a measurable value was obtained.

The concentration of bicarbonate was estimated by ionic balance calculations assuming bicarbonate as the missing anion when the equivalents of all other major salivary ions, i.e. sodium, potassium, calcium, chloride and total phosphate, were known [10]. The saliva pH was calculated by the Henderson-Hasselbalch equation using the bicarbonate concentration and assuming an average P_{CO2} for whole saliva of 4.4 kPa [11,12]. From these measures the degree of saturation (DS) with respect to hydroxyapatite (HAp) was determined for conditions at 37°C [13]. The solubility product for HAp [$Ca_{10}(PO_4)_6(OH)_2$] was set at 117.3 (pK) [14], pKw at 13.6, pK for H_3PO_4/

Table I. Characteristics of the three study groups (mean ± SD or ratio).

	Healthy (H) (n = 85)	Erosion (E) (n = 85)	Caries (C) (n = 85)	H vs E	H vs C	E vs C
Age (years)	47 ± 15	48 ± 15	48 ± 14	NS	NS	NS
Gender (male/female)	35/50	29/56	27/58	NS	NS	NS
Teeth (number)	28.9 ± 2.2	28.1 ± 1.9	27.2 ± 2.3	0.012	<0.001	0.017
DFS (% of all)	5.5 ± 5.1	24.6 ± 14.6	56.0 ± 21.5	<0.001	<0.001	<0.001
Erosion (yes/no)	0/85	85/0	0/85	<0.001	NS	<0.001
Surfaces with erosion	0.0 ± 0.0	9.3 ± 14.4	0.0 ± 0.0	<0.001	NS	<0.001
Dry mouth (yes/no)*	4/81	11/74	13/72	NS	0.041	NS
Saliva flow (mL/min)	2.19 ± 0.82	2.05 ± 0.75	1.87 ± 0.60	NS	0.004	NS
Subnormal flow (yes/no)**	0/85	0/85	0/85	NS	NS	NS
Water hardness (dH)	17.3 ± 5.3	16.8 ± 5.3	15.1 ± 5.9	NS	0.011	0.042
Water calcium (mg/L)	98.7 ± 28.9	97.5 ± 30.5	86.2 ± 32.9	NS	0.009	0.021
Water fluoride (mg/L)	0.34 ± 0.15	0.32 ± 0.19	0.30 ± 0.20	NS	NS	NS

Comparisons were made by a two-sample t-test or the Chi-squared test for tabular data and only p-Values with $p \leq 0.050$ are shown. The broken horizontal line indicates the drinking water data were obtained on a municipality level. * Indicates subjective complaints of dry mouth [25] and ** denotes a stimulated whole saliva flow rate <1.0 mL/min. NS denotes non-significant differences (i.e. $p > 0.050$).

Table II. Saliva composition in the three study groups (mean ± SD).

	Healthy (H) (n = 85)	Erosion (E) (n = 85)	Caries (C) (n = 85)	H vs E	H vs C	E vs C
Sodium (mmol/L)	19 ± 9	15 ± 9	14 ± 9	0.013	0.001	NS
Potassium (mmol/L)	18 ± 3	19 ± 3	19 ± 3	NS	NS	NS
Chloride (mmol/L)	16 ± 8	15 ± 6	16 ± 7	NS	NS	NS
Total calcium (mmol/L)	1.6 ± 0.7	1.4 ± 0.6	1.3 ± 0.4	0.065	0.003	NS
Total phosphate (mmol/L)	4.1 ± 1.4	3.7 ± 1.2	3.6 ± 0.9	0.088	0.010	NS
Bicarbonate (mmol/L)	17.0 ± 6.5	15.5 ± 6.1	13.3 ± 6.0	NS	<0.001	0.017
pH	7.29 ± 0.18	7.25 ± 0.19	7.17 ± 0.23	NS	<0.001	0.017
log DS_{HAp}	1.39 ± 0.15	1.33 ± 0.17	1.26 ± 0.28	0.019	<0.001	0.035
Critical pH	5.29 ± 0.15	5.34 ± 0.16	5.34 ± 0.13	0.072	0.020	NS
Total protein (μg/mL)	3241 ± 1148	3223 ± 1255	3358 ± 983	NS	NS	NS
Amylase activity (U/mL)	169 ± 84	176 ± 75	184 ± 89	NS	NS	NS

Comparisons were made by a two-sample t-test and all p-Values with $p \leq 0.100$ are shown. The broken horizontal line indicates inorganic variables (top) and organic variables (bottom). NS denotes non-significant differences (i.e. $p > 0.100$).

$H_2PO_4^-$ at 2.2, for $H_2PO_4^-/HPO_4^{2-}$ at 7.2 and for HPO_4^{2-}/PO_4^{3-} at 12.2, with all dissociation constants corrected for the ionic strength in each sample [11,15]. The critical pH was iteratively estimated as the pH at which the ionic product for HAp equaled the solubility product for HAp at 37°C (i.e. pK 117.3). Iterations were repeated until the pH used for determination of phosphate differed no more than 0.5‰ from the estimated critical pH [16]. All calculations were processed as a script in a computer program [17], allowing for process of multiple samples simultaneously [13].

Statistical analyses

Statistical analyses were done with Excel and with the R statistical program [17]. Linear data from all three groups were compared using a two-sample t-test and when transformed to an individual level analyzed for further exploration by multiple regression analysis with R and p-Values given in the tables. Differences in tabular data among groups were determined by the Chi-squared test. During statistical analyses pH and critical pH were expressed in concentrations and then averaged for mean results as $[\log((\sum 10\text{^-pH}_X)/n]$. For the demographic data the level of significance was set at $p \leq 0.05$ and values above the significance levels are not shown. However, for the salivary data also p-Values ≤ 0.10 are shown in order also to reveal results that were borderline significant and, thus, to clarify these results further.

Results

The groups

Table I shows the main characteristics of the three study groups. No significant differences were present between the age and gender distributions in the three study groups. All the groups differed in number of remaining teeth which was highest in the healthy group and lowest in the caries group. Nonetheless, the maximum numerical difference in number of teeth amounted to less than two teeth in total and was obtained between the healthy and the caries group. DFS calculated as a percentage of all

Table III. Multiple regression analysis on %DFS without the compositional salivary variables (n = 255).

Variable	Estimate	SD	t-value	p-Value
Age (years)	0.61	0.10	6.00	< 0.001
Remaining teeth (n)	3.48	0.67	5.23	< 0.001
Salivary flow rate (mL/min)	6.02	1.93	3.13	< 0.01
Complaints of dry mouth	8.64	3.48	2.48	< 0.05
Multiple R^2 value				0.334
p-Value for model				< 0.001

All subjects and variables from Table I were included in the analyses. The model was identified using iterative search and testing and backwards elimination.

Table IV. Multiple regression analysis of the annual increment in %DFS, saliva composition and demographic variables in a selected group of subjects without complaints of dry mouth ($n = 227$).

Variable	Estimate	SD	*t*-value	*p*-Value
Bicarbonate (mmol/L)	0.006	0.002	2.83	< 0.01
Phosphate (mmol/L)	0.040	0.010	3.98	< 0.001
Calcium in water (mg/L)	0.001	0.000	2.65	< 0.01
Salivary flow (mL/min)	0.042	0.018	2.40	< 0.05
Number of remaining teeth	0.016	0.005	2.90	< 0.01
Multiple R^2 value				0.190
p-Value for model				< 0.001

The annual increment in %DFS was calculated as %DFS divided by the age of the subject minus 10. The model was identified using iterative search and testing and backwards elimination.

remaining surfaces in the dentition showed that, on average in the healthy group, about one in 20 surfaces were affected by caries or filled compared with the caries group where more than half of all surfaces were affected by caries or filled. The erosion group had a % DFS that was nearly in the middle between the healthy and the caries group. No subjects in the healthy and caries groups showed signs of dental erosion, whereas all subjects in the erosion group showed clearly visible signs of dental erosion, although with a varying number of affected teeth (Table I). With respect to the sensation of dry mouth, the healthy group differed from the caries group by having fewer subjects complaining of dry mouth. Also the salivary flow rate was slightly higher in the healthy group compared with the caries group, although no subjects in any of the groups had sub-normal chewing-stimulated whole saliva flow rates (i.e. <1.0 mL/min).

With respect to the geochemical composition of the drinking water, subjects from the healthy and the erosion groups came from areas with slightly higher calcium content in the drinking water and also slightly higher drinking water hardness ($p < 0.05$ or less). Nonetheless, no significant differences were obtained in the measure for fluoride in the drinking water among groups (Table I) and in general all municipalities were represented with subjects from the three groups, although, in a slightly varying manner (Figure 1).

Saliva composition in the three groups

Table II shows the saliva composition in the three study groups. Compared with the caries group the healthy group had more sodium, calcium, phosphate and bicarbonate as well as a higher pH and degree of saturation with respect to HAp and a lower critical pH ($p < 0.05$ or less). Compared with the erosion group, the healthy group also had more sodium, calcium and phosphate as well as a higher degree of saturation and a slightly lower critical pH ($p < 0.05$ or less). Comparing the erosion group with the caries group, fewer differences were obtained; here only the pH, bicarbonate concentration and the degree of saturation were higher in the erosion group compared with the caries group ($p < 0.05$). No differences were obtained in the concentration of total protein or amylase activity among the three groups, although the caries group had the highest amylase activity and protein concentration among the three groups. Overall the healthy group differed the most from the other two groups, generally in having a better physicochemical saliva composition with respect to higher concentrations of calcium, phosphate and bicarbonate. Fewer differences were obtained between the caries and erosion groups.

Saliva composition and dental caries on an individual level

Table III shows the effect of significant demographic and individual variables from Table I on %DFS on an individual level using multiple regression analysis. As shown, the explanatory power on %DFS for a model containing the age of the subjects, the number of remaining teeth, the stimulated whole saliva flow rate and complaints of oral dryness was 33% ($p < 0.001$). %DFS increased with age, as did complaints of dry mouth, and decreased with higher saliva flow rates and higher number of remaining teeth. To eliminate these effects in further analyses it was decided to exclude subjects with dry mouth ($n = 28$) and to calculate a composite measure for % DFS, which was also based on the age of the subject. Thus, assuming a linear increase in %DFS, the annual increment in caries experience was calculated using the per cent decayed and filled variable divided by the age of the subjects minus 10. Thereby a composite estimate of the annual increment in % DFS in each of the 255 subjects could be determined and age could be eliminated as a covariate in the analyses, focusing on the effect of the saliva composition on %DFS.

Using this approach the highest explanatory power was obtained with the saliva bicarbonate and

phosphate concentrations combined. The explanatory power on the annual %DFS variable for a model containing the saliva bicarbonate and phosphate concentrations was 10% ($p < 0.001$). High concentrations of both ions decreased the annual %DFS. Significant relations could also be obtained with calcium, pH and the degree of saturation with respect to HAp ($p < 0.05$), but none as significant as for bicarbonate and phosphate combined. Furthermore, a final model for annual %DFS was also developed using all available data (Table IV). This model had an explanatory power of 19% ($p < 0.001$) and contained salivary bicarbonate and phosphate as well as the drinking water calcium level, the stimulated saliva flow rate and the number of remaining teeth. High levels or numbers of all five variables decreased annual %DFS and vice versa.

Saliva composition and dental erosion on an individual level

In general, the numbers of eroded surfaces decreased with increasing age, although not significantly and as a reflection of fewer cases of dental erosion among the older age groups. To determine the effect of saliva composition, similar analyses were applied to the number of eroded surfaces, without the dry mouth subjects and only among subjects having eroded surfaces. The highest explanatory power was obtained from the saliva degree of saturation with respect to HAp and the drinking water level of chloride ($R = 0.18$; $p < 0.01$). Without the drinking water variable and only the saliva degree of saturation, the explanatory power was 11% ($p < 0.01$) and, thus, close to that obtained for caries and saliva composition.

Discussion

The effect of saliva on dental health has been of major interest in dental science for more than 100 years. Many studies have clearly pinpointed the saliva flow rate [2] and especially the unstimulated saliva flow rate [3] as a major determinant of caries lesion formation and caries experience as well as a main factor in dental erosion [1]. However, also the effect of saliva composition on dental caries and erosion has been studied intensively. Nevertheless, no clear results on unique compositional salivary variables as predictors of dental caries as well as dental erosion have been identified, although it is indicated that low salivary buffering capacity, calcium and phosphate and sIgA show some relation to increased caries [2]. One problem with many studies on the effect of saliva composition on dental disease is that subjects with very different saliva flow rates are often included and compared with others. Including subjects with different flow rates makes comparisons on a compositional level nearly impossible because of the pronounced effect of the flow rate on both dental caries [3] and dental erosion [1]. Thus, preferably only subjects with equal saliva flow rates should be compared with one another with respect to the saliva composition and diseases of the enamel and dental hard tissues [4].

In the present study great care was taken to include only subjects with normal salivary flow rates and preferably with similar flow rates in all three groups. In spite of this effort, the subjects in the healthy group still came out with a slightly higher salivary flow rate than the subjects in the two groups with diseases of the enamel, especially the caries group, and more subjects in the caries group complained of dry mouth compared with the healthy group. The drinking water calcium concentration and hardness also differed slightly among the groups. However, the major environmental factor for dental diseases, namely the fluoride level in the drinking water [7], did not differ significantly among the groups. Thus, especially the drinking water fluoride content was a major factor when the selection of the groups was performed and, therefore, this measure was nearly equal among groups (Table II and Figure 1). With a relatively low geographic mobility in Denmark, which decreases with age, most subjects would probably have lived in the same municipality for the majority of their lives [18]. Thus, the differences in disease pattern and of the magnitude described seem unlikely to have been strongly influenced by geographic mobility.

With respect to the demographic variables described, the caries group differed the most from the other two groups, whereas fewer differences were obtained between the healthy and erosion groups. Still, the pattern of dental diseases was highly different among the groups, with the caries group having 10-times higher caries experience compared with the healthy group and the erosion group differing distinctively in this parameter compared with the two other groups. Assuming that the general differences among the groups were minor, then the differences obtained in dental disease pattern could be related to the saliva composition. For this parameter the healthy group seemed to be different from the other two groups with respect to generally having a better physicochemical and more tooth protective saliva composition. Thus, with more calcium, phosphate and bicarbonate and a higher pH and DS_{HAp}, this group would have a better re-mineralizing potential than the other two groups. Also the finding of a lower critical pH in the healthy group shows that plaque pH most likely has to become more acidic for enamel to dissolve in this group compared with the two other groups. Thus, on a group level the healthy group seemed to be the one that differed the most compared with the two other groups. In this perspective one may say that, on a group level, some subjects were better protected against dental caries and erosion due to a more favorable physicochemical saliva

composition. Thus, all the findings obtained for the healthy group were synonymous with a higher salivary protection against enamel dissolution and a higher remineralizing potential.

Nonetheless, relations obtained on an individual level using the annual increment in percentage caries experience and without subjects complaining of dry mouth showed a very poor explanatory power. Thus, only 10% of the variation in annual %DFS could be explained by the saliva composition when data were analysed on an individual level. These findings reveal that the relations for saliva composition were primarily related to a group level and that many subjects with little caries experience had saliva compositions that were worse from a physicochemical perspective than some of the subjects with high caries experience. This finding was in spite of a considerable variation in the variable for annual increment in %DFS, which ranged from 0% up to 5% per year and around a mean of 0.8% per year. However, the assumption of a linear increase in %DFS most likely does not reflect reality and, therefore, this measure could also have had an effect on the results. Thus, many subjects would probably have had periods with high caries activity and development of dental erosion and other periods, especially later in their life, with a lower activity. Nonetheless, according to the data in the present study, a measurement of the compositional salivary variables studied seems not to be predictive of the caries experience on an individual level. Expanding the model with the salivary flow rate, the drinking water calcium level and the number of remaining teeth increased the explanatory power slightly to 19% on annual %DFS, although no clear relations were obtained (Table IV). Thus, it is more than likely that other factors have had greater effect on %DFS than the saliva composition and the drinking water calcium. Among the factors that could have had an effect are oral hygiene habits are daily exposure to fluoride-containing foodstuffs and oral hygiene products [19], as well as the intake of sucrose [20]. Most likely, these behavioral factors would have shown a higher explanatory power on annual %DFS than the stimulated whole saliva composition.

Similar analyses were also performed for dental erosion, with only the erosion group in the analyses and without subjects complaining of dry mouth. It is possible that a few of the lesions that were in the erosion group could have had a component of other wear conditions, especially in the older age groups. The findings for the erosion group may, therefore, not complete represent chemical wear, although great care was taken to instruct the investigators to look specifically for erosive wear. With this in mind, then the saliva variable that had the highest explanatory power on erosive wear was the saliva DS_{HAp}. By itself this variable could explain 11% of the variation in dental erosion ($p < 0.01$). This is very near the explanatory power of the saliva composition on % DFS and it, therefore, seems that the stimulated whole saliva composition among healthy subjects with a normal salivary flow rate only to a small degree determines the level of disease in the dental hard tissues. The explanatory power on the number of eroded surfaces could be increased further by adding the drinking water chloride levels to the model. However, this relation was most likely an artifact and does not reflect any likely physicochemical effects on enamel dissolution. Thus, most likely the intake of acidic beverages would have had a much higher explanatory power on the number of eroded surfaces [21,22].

With respect to the protein content and amylase activity in whole saliva, no differences were obtained among the groups. This could have been anticipated, because many salivary proteins have antimicrobial effects and because the salivary proteins are the source of material for the acquired pellicle, which seems also to offer considerable protection against dental erosion [23]. Theoretically, the finding of a slightly higher amylase activity among subjects with high caries experience could be related to the starch degrading activity of this enzyme, making fermentable sugars more available for the oral micro-organisms [24]. However, the difference was not significant and no relations between the amylase activity and %DFS were found on an individual level using regression analysis (R^2 less than 0.001). However, because individual protein profiles were not analyzed for each subject, it was not possible to determine differences in individual salivary proteins among the groups. It is not unlikely that such differences could be more pronounced than differences in total protein and amylase activity.

The present study focused on the value of only the paraffin-chewing stimulated whole saliva composition as a tool in clinical decision-making. Thus, unstimulated whole saliva was not collected and, therefore, no data are available on the flow and composition of this secretion in the present study groups. It is possible that more significant and clear relations could have been obtained with unstimulated whole saliva [1–3]. Thus, unstimulated whole saliva is the physiological background for most processes that occur between the enamel and the environment surrounding it. In contrast, stimulated whole saliva is only present for a short period of time during meals and this may be the reason why such poor relations were obtained in the present study. Thus, studies focusing on the composition of unstimulated whole saliva with a set-up similar to the present would seem to be worthwhile.

Conclusions

The present study points in the direction that, on a group level, subjects with very little past and present diseases of the enamel seem to have a more favorable physicochemical saliva composition with higher

concentrations of calcium, phosphate and bicarbonate, compared with subjects with dental diseases. However, on an individual level the explanatory power even for the most significant salivary findings among the groups was poor. Thus, it is concluded that the compositional analyses performed in the present study on stimulated whole saliva, including major physicochemical characteristics of saliva, most likely will have little predictive value for future dental caries and erosion in single individuals.

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