LICENTIATAVHANDLING

ODONTOLOGISKA FAKULTETEN

GÖTEBORGS UNIVERSITET

On Minor Salivary Glands in Relation to Medication, Life-style and Use of Dentures

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Contents

Original Papers

This thesis is based on the following papers, which are referred to in the text by their Roman numerals:

I. Eliasson L, Heyden G, Landahl S, Steen B. Effects of tobacco and diuretics on human palatal salivary glands. J Oral Pathol Med 1991; 20: 126-9.

II. Eliasson L, Birkhed D, Heyden G, Strömberg N. Studies on human minor salivary gland secretions using the Periotron® method. Archs Oral Biol 1996; accepted for publication.

III. Eliasson L, Dahlén G, Heyden G, Möller Å. The predominant microflora of the palatal mucosa in an elderly island population. Acta Odontol Scand 1992; 50: 163-9.

Abstract

Minor salivary gland pathology and function were studied among participants of the Koster Health Project. It was confirmed by comparing colour macro-photographs of the palatal mucosa with histological preparations that clinical photographs could be used as non-invasive tools for demonstration of palatal sialadenitis. Use of tobacco and diuretics, as well as the wearing of upper dentures, were identified as risk factors for the development of sialadenitis in palatal salivary glands.

Mean secretion rates for palatal, buccal and lower labial mucosal salivary glands were calculated in a population of 127 individuals from measurements with the Periotron® method. The buccal glands showed the highest, and palatal the lowest values. Women had lower mean secretion rates in all mucosal sites investigated. Among individuals with risk factors for sialadenitis, the tobacco users had a 27% higher mean palatal secretion rate, the diuretics users a 15% lower buccal secretion rate, and the upper denture wearers a 300% higher palatal secretion rate, than the controls. Persons with subjective oral dryness had a 21% lower output from lower labial mucosal glands as compared with others.

Palatal mucosal colonization of microorganisms was investigated in wearers of upper dentures. Non-oral, gram-negative species and fungi were found among individuals with denture stomatitis. It is suggested that immunological factors, associated with a changed salivary environment under upper dentures, should be included among pre-disposing factors for shift of the microbial flora, and the development of denture stomatitis.

Introduction

It is generally believed that a deteriorating general health, medications against diseases and negative life-styles may increase the risk for caries, periodontitis, and pathological oral mucosal changes. Early symptoms of disease could also cause unspecified alterations in oral mucous membranes. It has thus been proposed that dentists should use their judgment of oral health changes to monitor general health alterations, especially among elderly people (1).

Saliva plays an important role in maintaining oral health by its host-defense functions, established by specific salivary proteins (2, 3). Salivary glands produce their secretions by transforming capillary blood to interstitial fluid, which is secreted as acinar fluid in the terminal end pieces of the gland parenchyma, and finally modulated in the ductal systems (4). Factors negatively influencing the blood circulation in the tissues of the human body must therefore, logically, be regarded as potential risk factors for oral health by altering the salivary secretion.

Influence of tobacco use

Tobacco habits are known to be hazardous to oral health. The relationship between tobacco use and a significant lower number of remaining teeth (5, 6), higher prevalence for periodontitis, aggravation of the disease by increased pocket depth (7-10), higher plaque index (11), lower salivary buffer effects, higher amount of lactobacilli and *Streptococcus mutans* (12), and increased caries incidence (13) among tobacco users have been reported. These effects of tobacco could, to some extent, directly depend on local or vaso-active effects in the oral cavity of noxious components from tobacco products (14), but effects on salivary glands with adverse effects on the gland secretions must also be considered as possible ways of threat to oral health (12, 15, 16).

Influence of disease and medication

Medication, especially with drugs causing oral dryness symptoms, lowering the salivary secretion rates or altering the composition of saliva, has long been regarded as a potential risk factor for caries development (17-21). These reports are based on findings of whole saliva secretions, but lowered palatal gland secretion rate among medicated persons has also been shown (22).

However, it has also been discussed that diseases, such as cystic fibrosis, Chron's disease and Sjögren's syndrome, regardless of treatment, could have influences on the salivary secretions (23-27).

Influence of age and gender

Conflicting data about the influence of age and gender on major salivary gland secretion rates have been reported (for a review, see (28)). Various investigators have found an age related lower salivary flow rate or degenerative changes in labial glands, suggesting reduced function with increasing age (29-31). Smith *et al.* found an age related decline but no gender based differences (32), while Niedermeier *et al.* found no correlation between age or sex on palatal gland secretion rates (22, 33, 34). Östlund observed that the secretory capacity decreases with increasing age, and that the secretory rate of the palatal glands is higher in men than in women (35). Two recently published investigations claim that there is no influence of gender or age on minor gland secretory rates (36, 37). The reports indicate a lower minor gland flow output among women than among men. However, in the populations analyzed this difference was not statistically significant.

Influence of upper denture wearing

Niedermeier *et al.* (22) have measured palatal gland function and found increased salivary flow rates among wearers of upper dentures, a finding which had been pointed out some years earlier (34).

Co-variation between major and minor salivary gland secretion rates

In studies, comparing major and minor gland flow rates, the absence of co-variation between these glands has been demonstrated (33, 36, 38). It has also been shown that the minor glands respond less to stimulation than the major glands (39, 40).

Oral dryness and salivary secretion rate

Total salivary secretion rates among persons complaining of oral dryness was found to be low in a study by Johnson *et al.* (19), while others report on poor correlation (20, 41). Minor gland secretion in individuals with feelings of oral dryness has also gathered interest from some authors, and palatal and labial gland secretion rates have been found lowered among these persons compared to others (33, 34, 42).

Minor salivary gland secretions

Compared to major salivary gland secretions and whole saliva, less data has been published on minor gland function. Even though Schneyer (43) expressed the opinion that minor salivary glands make no significant contribution to the total unstimulated volume of saliva, the findings of Dawes and Wood (44) that 7-10 % of the total volume of whole saliva is minor salivary gland secretions, with high concentrations of IgA (45-47) and glycoproteins (48), with considerable contribution to the formation of the acquired pellicle (49) and high biological activities (50, 51) is mostly embraced today.

Methods for studying minor salivary gland functions

Some of the above mentioned investigations of minor salivary glands are based on histomorphologic studies (24, 25, 29, 30, 35, 52), but the difficulties in the collection of this kind of saliva have hampered studies on minor salivary gland secretions. Micro-pipettes, capillary tubes, sponges, synthetic discs and filter-papers have been used in collecting minor salivary gland secretions for qualitative research (39, 40, 45-47, 49, 51, 53-56). Semi-quantitative estimations, weighing methods or measurements of the secreted droplets, have been applied to estimate minor gland flow rates (27, 31, 32, 35, 40, 42, 43, 46, 55, 57-59).

The Periotron® method

Recently, an electronic device (Periotron®, Pro-Flow™, Inc., Amityville, NY, USA), originally designed for gingival fluid measurements (60), has been commercially available in a modified version for quantitative estimations of minor gland secretions.

This new device seems to have greatly improved precision and versatility in estimating minor gland function, expressed as secretion rate, and some of the published investigations of mucosal salivary flow rates have been performed with this technique (22, 33, 34, 36-38, 61). The mean values of mucosal gland secretion rates presented in these publications are summarized in Table 1.

Theoretical principles of the Periotron® method

Moisture from the oral mucosa is harvested by applying a piece of blotting paper against the mucosa. Volume measurements of the moisture are carried out by determining the *dielectric properties* of the blotting paper. These properties change depending on how much saliva has been absorbed by the paper.

Table 1. *Reported means (µl/cm²/min) for secretion rates from oral mucosal glands in published works using the Periotron® method.*

		Niedermeier & Huber 1989 (34)	Niedermeier, Schaller & Fischer 1989 (33)	Niedermeier & Krämer 1992 (22)	Shern, Guckes & Li 1993 (38)	Shern *et al.* 1990 (61)	Shern, Fox & Li 1993 (36)	Sivarajasingam & Drummond 1995 (37)	Eliasson *et al.* 1996 (Paper II)
Number of investigated individuals		134	121	86	12	14	51	99	127
Palatal gland estimations	Males	1.36*	1.06	-	0.31*	0.68*	0.58	0.59	1.02
	Females		0.99	-			0.53	0.53	0.78
	Denture wearers	+	-	3.69*	-		-	-	3.60*
Buccal gland estimations	Males	-	-	-	-	2.60*	2.36	3.02	16.7
	Females	-	-	-	-		2.09	2.94	15.2
Lower labial gland estimations	Males	-	-	-	-	0.92*	0.86	2.38	5.12
	Females	-	-	-	-		0.78	2.00	4.38

* no gender difference shown. + higher mean compared to non-denture wearers reported.

The instrument is equipped with a *capacitor,* which is a device capable of storing electric charge. This capacitor consists of two arbitrarily shaped parallel plates separated by a distance. When applying a voltage across the plates, an excess of electrons will build up on the plate connected to the negative terminal and a corresponding depletion of electrons will occur on the plate connected to the positive terminal, giving an excess of positive charge. The relation between plate charge and voltage is linear, i.e. the charge on the plates is equal to the voltage applied times a proportionality constant. This proportionality constant is called the *capacitance* of the system. If a slab of some electrically non-conducting material is introduced between the plates, the charge of the capacitor will increase by a factor, called the *dielectric constant* of the material in the slab. This behavior can be understood by looking at the slab material at the molecular level. The substance contains a large number of *dipoles,* i.e. two particles of opposite charge but with the same magnitude separated by some distance. When a voltage is applied across the system, the dipoles will align themselves in such a way as to point in the direction of the electric field imposed by the voltage. This will result in an excess of charge at the interface between the slab and the electrode. Since the plus sides of the dipoles are attracted by the negatively charged electrode and the minus sides are attracted to the positively charged electrode, the total charge in the system made up of one electrode and the interface will be smaller than the charge the plate had before the dielectric material was introduced. To compensate for this, the charge on the plates will increase so that the net charge will be the same as it was before the slab came into place. Thus, the introduction of the slab will lead to an increase in the capacitance.

The capacitance can be determined by applying an in time alternating voltage across the capacitor. As the frequency of the applied field is increased, it will be more difficult for the dipoles to align themselves with the field. The motion of the dipoles will thus be delayed to some extent with respect to the field frequency. Mathematically this is described by splitting the *dielectric constant* into two parts; one in phase with the field (real component), and one 90° out of phase (imaginary component). It can be shown that each of these parts is dependent upon the moisture content in the blotting paper. Since the capacitance is proportional to the dielectric constant, it can also be described using this two-component representation. By measuring the capacitance at a fixed frequency, and calculating its real and imaginary components, it is possible to determine the moisture content of the paper.

Denture stomatitis

Since Newton published his article in 1962 (62), denture stomatitis has been classified into three different stages, and this has later been supported by others (63-65). Newton (62) suggested that type I should be regarded as a response to mechanical occlusion of the excretory ducts of the palatal salivary glands. Increased growth of microorganisms has been found in the types II and III denture stomatitis (64, 66-72), and it has been suggested that the local environment on the palatal mucosa in denture wearers might predispose to fungal growth (73).

Microbiology of denture stomatitis

As fungal infection (i. e. *Candida albicans*) has been regarded as the main ethiological factor for the development of denture stomatitis, the investigations of the microbiota of the palatal mucosa in denture wearers have been directed mostly towards the detection of fungi (62, 64-66, 70-79). However, papers supporting the opinion of a more complex microbiological ethiology have been published as well (63, 67, 68, 80, 81). Holbrook and Russell (82) have emphasized that *Klebsiella spp.* may serve as pathogens in denture stomatitis. Koopmans *et al.* found shift to more anaerobic bacteria (69) and regarded *Candida spp.* as a super-infection over a changed microbial flora in denture stomatitis (83).

Aims

As saliva, and particularly the minor salivary gland secretions, play an important role in maintaining oral health, it is important to clarify how certain factors can influence their secretion ability, and how changes in the secretory patterns could affect bacterial activities in the oral cavity.

The aims of the investigations in this thesis were therefore

- to relate histomorphological changes in minor mucosal glands to macrophotographically registered sialadenitis
- to investigate influences of denture wearing, tobacco habits and diuretics use on sialadenitis in palatal minor salivary glands
- to record the oral minor salivary gland flow output in an adult population
- to investigate influences of denture wearing, tobacco habits and diuretics use on oral minor salivary gland flow output, and
- to describe the predominant microflora of an oral mucosal area, dominated by one distinctive saliva.

A

B

Fig. 1. Macrophotographs of the palatal mucosa from a person, who did not use tobacco at first examination (A), and the next year (B) had started smoking cigarettes with a coincidental development of sialadenitis.

Own investigations

I. EFFECTS OF TOBACCO AND DIURETICS ON HUMAN PALATAL SALIVARY GLANDS

The aim of this study was to confirm histologically the photographically registered signs of sialadenitis in the palatal salivary glands, and to study the prevalence of palatal sialadenitis with special reference to use of tobacco and medications.

Material and methods

The prevalence studies were performed among 184 persons, 102 women and 82 men, aged 23-90 years (mean age 56 years). They were all permanent inhabitants of the Koster Islands, Strömstad, Sweden, and participated in 1989 in the annual health monitoring procedures of a longitudinal, interdisciplinary project called the Koster Health Project (84). Detailed social, psychological, nutritional, neighbouring environmental, medical, and oral medical data were recorded in a team work between physicians and dentists. In the oral medical part, a series of photographic 35 mm colour slides (magnification 1: 1 or 1: 1.5) was taken. The camera set-up and the technique has been described in detail (85).

Signs of sialadenitis were registered from the photographs of the palatal mucosa (Fig. 1) without any simultaneous access to the corresponding medical records. Code numbers printed on each slide disguised the identity of each case. For the statistical testing of the differences of prevalence of palatal sialadenitis a technique with matched pairs was chosen, and *p*-values below 5% were considered statistically significant.

Ten consecutive islanders with clinical signs of palatal sialadenitis were chosen for palatal biopsies when they visited a dental surgery for routine dental check-ups. Care was taken to include both photographically documented normal and inflamed orifices in the same biopsies for comparison.

Results

The histomorphologic analyses showed a close relationship between the photographically visible changes and the tissue reactions. All palatal gland orifices appearing inflamed on the slides showed various degrees of inflammatory reactions with lymphocytes and plasma cells in the connective tissue adjacent to their terminal ducts. Squamous metaplasia of the duct epithelium was also seen in some of the most inflamed areas. Additionally, all affected ducts were accompanied by pathological changes in their deeper gland parenchyma. These changes could be seen as accumulations of inflammatory cells and mucous in the stroma, duct dilatation with mucous retention, and acinar atrophy. It was noted that the atrophy was followed by adipose tissue in growth. Glands with apparently normal orifices, included in the same biopsies did not show any histopathologic changes.

The overall prevalence of photographically documented palatal sialadenitis in this population was 41%. Women had a slightly lower prevalence than men (36 and 46% resp.) but the difference was not statistically significant. Denture wearers were heavily represented ($p<0.001$) in the sialadenitis group (Fig. 2). The representation of tobacco and diuretics use among persons with and without sialadenitis can also be seen in Fig. 2. To avoid interference of irrelevant factors when testing the differences of the representations of these two factors for statistical significance, matched controls were chosen. For use of tobacco, the controls were chosen with regards to age, sex, medication, and upper denture wearing. For diuretics, the factors age, sex, tobacco use, and upper denture wearing were included. The statistical verification for these differences were $p<0.001$ for tobacco and $p<0.01$ for diuretics use.

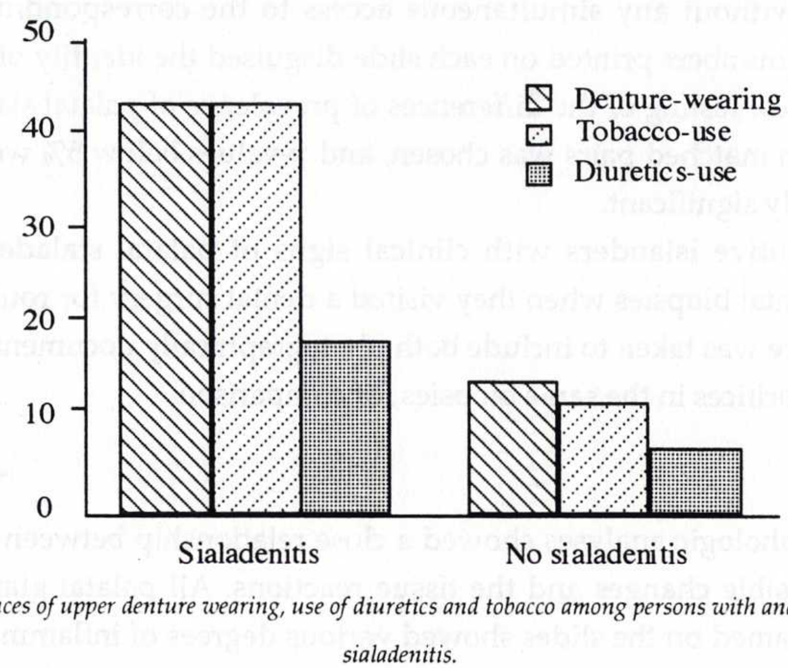

Fig. 2. Prevalences of upper denture wearing, use of diuretics and tobacco among persons with and without palatal sialadenitis.

II. STUDIES ON MINOR SALIVARY GLAND SECRETIONS USING THE PERIOTRON® METHOD

The aim of this study was to evaluate the Periotron® method when measuring the fluid output (secretion) from different oral mucosal sites in relation to age, gender, medication, use of dentures and tobacco.

Material and methods

This study was part of the longitudinal, interdisciplinary program on human response to environmental and life-style changes, the Koster Health Project. The population included in the present study consisted of 127 individuals, 61 females and 66 males (mean age 55 yrs, range 22-89 yrs), all permanent inhabitants of the Koster Islands. A total of 30 individuals were daily users of tobacco (28 smokers, 5 snuff dippers, and 3 both smokers and snuff dippers). Nine individuals were daily users of diuretics, none of which used tobacco. Twenty individuals wore upper full dentures. Measurements of fluid output were performed in the health survey during June-August 1994.

Periotron® measurements

A Periotron® 304 6000, model II (modified for minor salivary gland measurements) (Pro-Flow™, Inc.) was used to measure unstimulated secretion rates. Precut pieces of filter-paper (Munktell, Stora Kopparberg, Sweden, 10 x 15 mm in size) were used for the measurements. They produced equal results as Periopaper® (Pro-Flow™, Inc.), and were preferred because their larger size allowed the Periotron® instrument to be used as a constant reference for measured area calculation. Prior to use, the reliability of the instrument was checked. The pieces of filter-paper were loaded with known volumes of distilled water as determined by a microliter syringe for molecular biology (Eppendorf®, Hamburg, Germany). The wetted area of the paper was placed between the sensors of the device, and the three-digit readings were recorded according to the Periotron® manual. The reported values of secretion rates were calculated from the regression formula; $x = 0.00568y + 0.0421$, where x is the volume in μl, and y is the displayed digits on the instrument (Fig. 3).

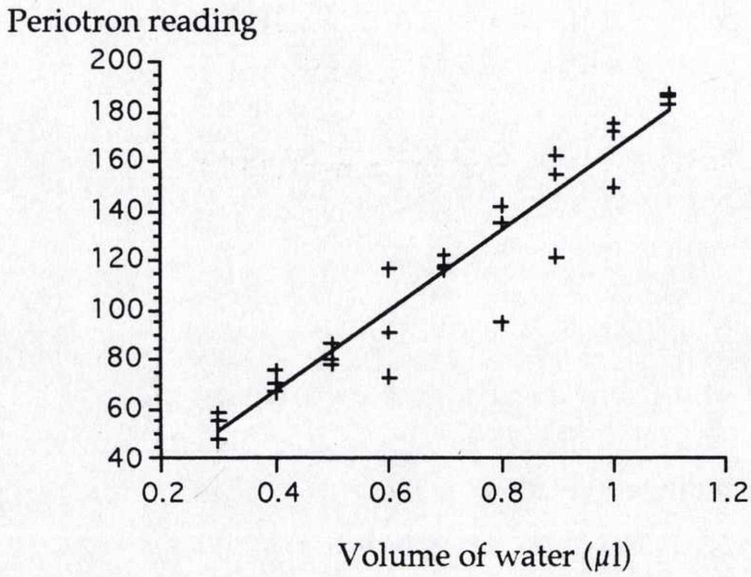

Fig. 3. Calibration curve for the used Periotron® instrument.

The sensors of the device were 9.1 mm in diameter, representing an area of 0.65 cm^2 for each reading.

The values depicted in Fig. 3, together with an additional check-up one month after the study, showed both high reproducibility (SD = 0.08 $\mu l/cm^2/min$) and stability in the readings of the instrument.

The individuals were asked not to eat or drink for at least 1 h before the measurements. The filter-paper was placed in the instrument for adjustment of zero, and after drying the actual mucosal area with a cotton gauze pad, it was placed on the mucosa. The paper strip was handled using surgical gloves and held in place with light finger pressure to assure mucosal contact and to block the paper strip from contact with moisture from the breathing air. Readings were made after the paper strips had been held in place for 30 s on the palatal, 5 s on the buccal and 15 s on the labial mucosa. Buccal secretions were measured on the left side, at the level of the parotid excretory duct and 2 cm anterior of it. Lower labial secretions were recorded near the mid-line, and 3 mm from the outer border of the labial mucosa. Palatal gland secretions were measured at three different sites; bilaterally near the first molar, and medially, near the border between the hard and soft palate.

Thirty-six individuals (20 females and 16 males) were randomly selected for double measurements in order to calculate the variance of the method. The two measurements at the different mucosal sites were performed with 20 min delay in time. The standard deviations calculated ($\mu l/cm^2/min$), including both intra-individual and method variations, were 0.57 (mean 1.34 including denture wearers) (43%) for the palatal mucosa, 2.7 (17%) for the buccal mucosa, and 0.86 (18%) for the labial mucosa. As the Periotron® method had a low method error (see above), the intra-individual variations constituted the main parts of the values.

Statistical methods

All statistical calculations were made using the originally registered readings from the Periotron® display. The calibration curve for the Periotron®, and the co-variations in fluid output between the different mucosal sites, were tested with simple regression. The influences of age, gender, upper denture wearing, use of diuretics and tobacco on the fluid output, were estimated by one-tailed, multiple regression test. Paired *t*-tests were used for the comparisons of fluid output between the different mucosal sites. The influence of mucosal fluid output on subjective feelings of dry mouth, was calculated by one-tailed, unpaired *t*-test (StatView®, Abacus, Berkeley, CA, USA). *p*-values below 0.05 were regarded as statistically significant.

Results

The mean mucosal output of fluid ($\mu l/cm^2/min$) were 16.0 for buccal, 4.8 for labial and 0.9 for palatal glands, which were highly significant differences ($p<0.001$).

The co-variation of fluid output between the various sites of the oral mucosa was calculated. While the fluid output from palatal and buccal or labial gland mucosa did not co-variate, a weak co-variation was observed for the buccal and labial mucosa ($p<0.05$). A strong co-variation ($p<0.001$), on the other hand, was observed for output from the palatal mucosa when measured at three different sites. Medial palatal sites showed, however, higher output (about 45%) compared to lateral sites ($p<0.01$).

The influences of age, gender, upper dentures, tobacco and diuretics on mucosal fluid output were investigated. Age did not influence the fluid output, while females had a 10-20% lower fluid output than males ($p<0.05$) from the measured sites (Fig. 4).

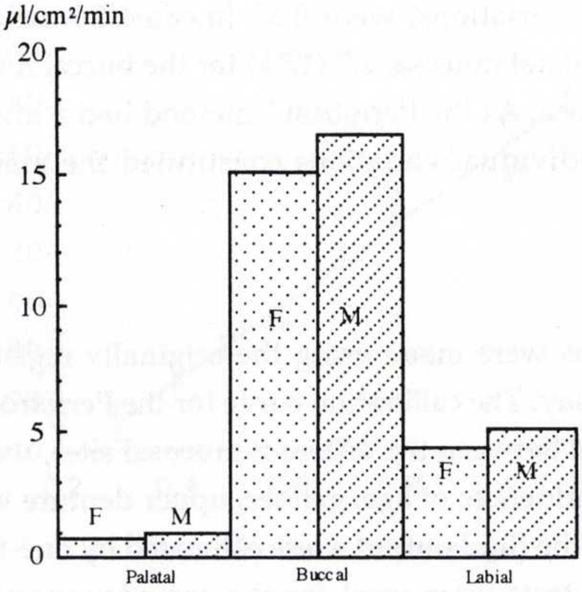

Fig. 4. Mean secretion rates (µl/cm²/min) of minor salivary glands among females and males.

Use of tobacco resulted in a significantly higher palatal fluid output (27%). In contrast, use of diuretics significantly reduced the secretion rates of buccal mucosa about 15%, while no influence on the other mucosal sites could be statistically verified. Wearing of upper dentures resulted in a major raise (300%) of the palatal gland secretion rates (Fig. 5).

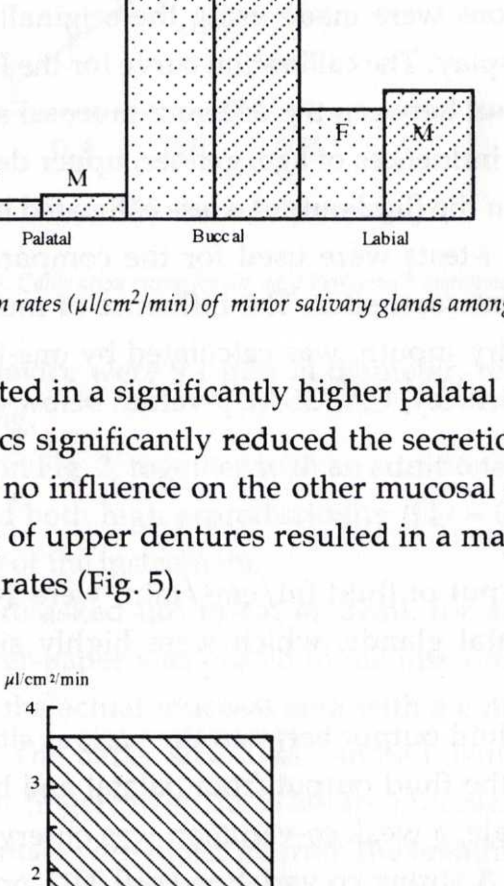

Fig. 5. Mean values of palatal gland secretion rates among upper denture wearers and non-denture wearers.

A possible relationship between mucosal fluid output and subjective feeling of dry mouth was investigated. Out of the 127 individuals, 14 experienced a feeling of dry mouth. One of them used diuretics, and 4 used tobacco. Among subjects with dry mouth labial fluid output was reduced 21% ($p<0.05$), while buccal and palatal did not show any differences.

III. THE PREDOMINANT MICROFLORA OF THE PALATAL MUCOSA IN AN ELDERLY ISLAND POPULATION

The aim of this study was to estimate the prevalence of denture stomatitis, discuss the role of sialadenitis, and to analyze the microbial colonization on the palatal mucosa in an elderly population on the Koster Islands .

Material and methods

The 184 persons investigated were medically and oral medically examined as described above, including the standardized series of macrophotographs. To avoid false diagnoses of the different stages of denture stomatitis (types I, II or III), efforts were made to eliminate the role of differences in colour reproduction of single slides. The evaluations of types I and II (62)were based on existing differences between clinically healthy and inflamed sites in the same individual, and type III (62) diagnoses were based on structural changes of the mucosal surface. The same criteria for sialadenitis was used as in Paper I.

Microbiological sampling was undertaken in 29 persons with denture stomatitis, in 8 wearers of upper dentures without stomatitis, and in 14 islanders with no dentures and clinically healthy palatal mucosa. After rinsing the mouth with water, samples were taken by scraping deep in the palatal mucosa over an area of 20 mm^2. The samples were transferred into a transport medium, described by Möller (86), and sent to the laboratory for processing. Additionally, samples for direct microscopy were taken from the same sites. The laboratory processing was made with standard procedures for detection of fungi. Growth of microorganisms was semi-quantitatively estimated as very heavy, heavy, moderate, sparse or very sparse.

Results

The prevalence of upper dentures among the Koster islanders was 27%, and the prevalence of denture stomatitis among them is presented in Fig. 6.

All cases with denture stomatitis, except for two persons in the stomatitis II and III groups with partial dentures covering only the mucosa on the alveolar ridges, showed sialadenitis in their palatal glands.

It was noted that persons with denture stomatitis had a lower prevalence of tobacco use than the other participants (p<0.05) (Fig. 7).

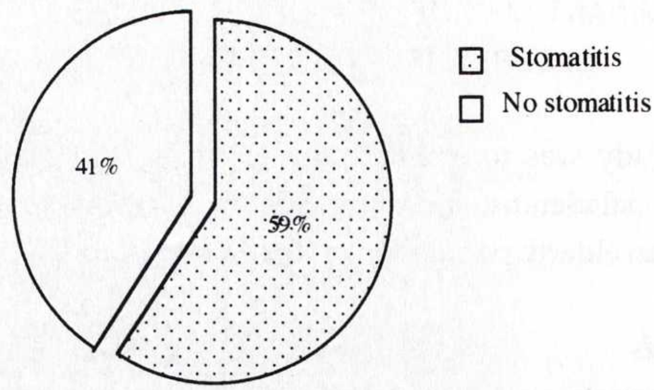

Fig. 6. Prevalence of stomatitis among upper denture wearers in the Koster population.

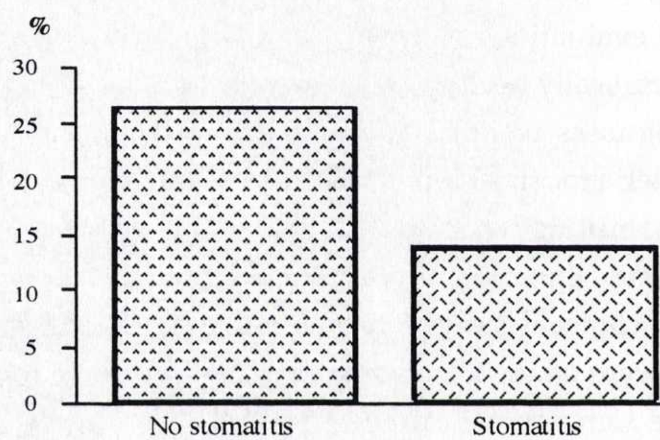

Fig. 7. Prevalences of tobacco use among the denture stomatitis and the non-stomatitis groups.

The microbiologically examined group (n=51) was divided into four categories: healthy controls without dentures (n=14), healthy controls with dentures (n=8), persons with denture stomatitis type I (n=10), and persons with denture stomatitis types II and III (n=19). Pathological colonization of micro-organisms was noted when the microbiological analyses showed moderate growth or more.

Except for oral streptococci, which were found in large numbers in all categories, persons with denture stomatitis types II and III showed an increased recovery of gram-negative species, fungi and non-oral microorganisms. Increased recovery of *Haemophilus spp., Bacteroides spp., Staphylococcus aureus, Haemophilus influenzae,* and *Klebsiella pneumoniae* was the most pronounced difference between this group and the others. Only one person in the stomatitis II and III groups showed mucosal colonization of fungi (*Candida albicans* and *glabrata*).

Discussion

In our part of the world, snuff lesions are the best known tobacco-related changes in the oral tissues. There are, however, even other mucosal changes which can be associated with the use of tobacco. Hyperkeratinisation of the palatal mucosa (87), a high prevalence of leucoedema (88) and general whitish clinical changes in the mucosa (89) are examples of such damage caused by tobacco. These tissue changes have been regarded as relatively harmless. This view has been upheld by investigations which have shown the reversibility of cell changes in snuff lesions after the individual had stopped using tobacco (90). In studies on the influence of tobacco on the salivary glands of the mucosa, extensive inflammatory and degenerative damage can be registered. Papers I and II show the presence of sialadenitis, degeneration of gland parenchyma and a changed secretion pattern of the minor salivary glands for, among others, tobacco users. The histopathological appearance of the palatal salivary glands with sialadenitis, which was described in Paper I, concurred in the main, with the earlier descriptions of the labial salivary glands of snuff users (52) and the palatal glands of denture wearers (35, 91).

Local irritation by the denture base is a probable explanation of the sialadenitis amongst denture wearers. General vaso-active mechanisms can exist with palatal sialadenitis and a changed secretion pattern in the buccal glands of diuretics users (Papers I and II). Amongst tobacco users, changes in the appearance and function of the salivary glands can have both systemic and local causes. The differences in the functional response to different stimuli (Paper II) also indicate that sialadenitis, especially in the palatal salivary glands, is an unspecific reaction.

By systematically taking clinical photographs of the oral tissues for health control purposes one can discover less noticeable mucosal changes at an early stage (Fig. 1). This is also an effective way to control the development of previously diagnosed pathological changes (92). The correlation between photographically registered and histomorphologically diagnosed sialadenitis in the palate (Paper I) also confirms the possibility of using clinical macro-photography of the oral mucous membranes as a non-invasive instrument for identifying inflammatory processes in the glands of the oral mucosa.

The Periotron® method is an improvement in comparison with older methods (as described in the introduction) of measuring the secretion rate in mucosal salivary glands. The simplicity and the accuracy of the instrument are its biggest advantages. The Periotron® system have been developed for measurements of absolute amounts. This is probably because the method was originally used for the estimations of exudate from gingival pockets in parodontological clinic and research. The Periopaper® supplied by the manufacturers for transporting the fluid (saliva) from the oral tissue to the instrument is not particularly well suited for area estimations since they are somewhat smaller than the instruments measuring surface. This gives rise to an uncertainty concerning which area one has measured. We have therefore used a filter paper which is slightly larger than the capacitor area of the instrument. This resulted in more precise measurements due to the instrument being the fixed reference for all the estimations. However, this may have certain side effects which could affect the measurements due to dampness in the filter paper outside the capacitor area. The capacitor consists of two parallel plates separated by some distance. At the edges of the plates, the electric field will not be uniform, but to some extent go outside the plates (Fig. 8). This might affect the volume estimations. However, these edge effects are very small, and if the ratio between the plate diameter (in the case of circular plates) and the distance between the plates (the thickness of the filter paper) is small, they can be neglected.

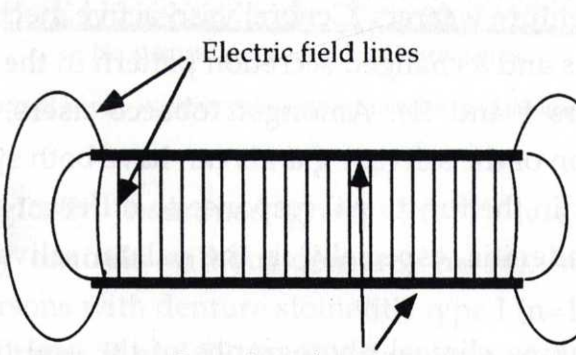

Fig. 8. Schematic view of the sensor area of the Periotron® instrument.

The lower secretion rates in the minor salivary glands in women compared with men and the fact that age doesn't seem to have an influence (Paper II) is not in agreement with other studies also using the Periotron® method (Table 1). There can be two explanations for the differences. In the other studies one has not included the life-style factor or medicinal effects. The present thesis has shown even large inter- and intra-individual variations when measuring the secretion rate in the accessory salivary glands. This makes it necessary to perform a large number of measurements in population studies. The main number of studies which are shown in Table 1 are based on relative few individuals compared to Paper II.

In accordance with previously published investigations, a large variation in the secretion rates between the different sites of mucosa was found. Even within anatomically well defined regions of mucosa there are large variations. Paper II shows that the secretion rate is greater in the medial part of the palate than in the lateral. The mean values of the secretion rates in buccal and labial areas of mucosa are different from other investigations (see Table 1). This is probably due to measuring in different parts of the mucosa and that the number of glands can vary across the surface of the separate mucous membranes. With repeated measurements of, for example, the effect of medicinal treatment on minor salivary gland function, it is therefore important to exactly define the sites used.

Some investigations have suggested that use of tobacco could increase the risk for oral *Candida* infections (for a review, see (93)). The low incidence of fungal infections amongst denture stomatitis in the population of Koster could be a result of the limited number of tobacco users in that group (Paper III). The dominating theory that *Candida albicans* should be the totally dominating pathogen for the development of denture stomatitis has, however, been questioned also in other studies (69, 94).

Saliva from minor mucosal glands have a high viscosity due to the high concentrations of mucins. Therefore they remain longer on the surfaces of the oral tissues, than saliva from larger glands. The minor salivary glands also contribute with a large portion of host-defence proteins in the oral cavity. There are, however, differences in the immunological content in saliva which is produced by the different glands (47). Saliva do also contain components which mediate bacterial adherence to tooth surfaces. Recent investigations have shown large variations in bacterial adherence to hydroxyl-apatite treated with different volumes of saliva from major and minor salivary glands (unpublished observations).

Mixed (whole) saliva consists of secretion from all the salivary glands. Normally, there is a certain balance between the various salivas. This balance could be expected to vary between the regions of the mouth depending on, for example, the secretion rate of the individual glands and the viscosity of the saliva. This might be one explanation for the findings of a different protein content in the acquired pellicle on various tooth surfaces in the mouth (95). A changed secretory pattern of individual salivary glands could then affect the content of the pellicle and thereby also oral health. This theory is supported by a recent study where a correlation between dental caries and reduced labial salivary secretion was shown (59). The status of the oral mucosa is probably also depending on the composition of the salivary film. The high prevalence of stomatitis and the tendency to a changed microflora among upper denture-wearers found in Paper III can therefore partly be due to the altered salivary conditions under the denture plates (Paper II). To be able to better understand how oral health is affected by, for example, medication and negative life-style factors, such as use of tobacco, more studies on the composition of the various salivas are, however, needed.

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