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THE BACTERIAL STATE OF DIFFERENT REGIONS WITHIN THE CLINICALLY HEALTHY GINGIVAL CREVICE

by

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The intimate relationship of bacteria to the pathogenesis of periodontal disease has been stressed by many authors (*Waerhaug* 1952, *Scherp & Burnett* 1956, *Schultz-Haudt* 1956). The question regarding the presence or absence of bacteria in the clinically healthy gingival crevice has been somewhat controversial. *Waerhaug & Steen* (1952) concluded from their investigations that the healthy crevice is, as a rule, sterile. *Boyd & Rosenthal* (1958) and *Gavin & Collins* (1961) on the other hand, stated that the healthy crevice normally contains bacteria. *Bervell* (1960) used a modification of the technique of Waerhaug & Steen and, contrary to the latter authors, found bacteria in the healthy crevice. The positive samples obtained by Bervell, after adequate disinfection of the neighbouring tooth and gingival surfaces, would appear to indicate that bacteria are normally present in the healthy crevice. This finding is in accordance with the histological observation that, even in sections of clinically healthy gingivae, varying numbers of chronic inflammatory cells can be

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noted in the connective tissue immediately beneath the crevicular epithelium (*Bernier* 1950, *Zachinsky* 1954). It has been suggested by *Schultz-Haudt* (1960) that these local inflammatory areas are caused by diffusion of various bacterial products through the crevicular epithelium and into the underlying tissue.

Clinically, gingivitis is often first detected in the interproximal area (*Fish* 1944, *Massler, Cohen & Schour* 1952, *Cohen* 1959). In manifest periodontitis the deepest pockets are found interproximally whilst the shallowest are usually situated buccally. The palatal/lingual pockets are generally deeper than the buccal ones but are not as deep as those of the inter-proximal region (*Castenfelt* 1950, *Lövdal, Arnö & Waerhaug* 1958). It is interesting to note that this variation in depth is also found in healthy gingivae (*Blumentritt* 1926, *Forsberg* 1951, *Rothner* 1954).

Previous studies with regard to the presence or absence of bacteria in the healthy crevice have been concerned with the crevice as a whole, and no attempt has been made to investigate different regions within the crevice. In view of this fact and in view of the variations in pocket depth noted, it was decided to determine whether any variation in the bacterial content could be detected between different regions within the healthy crevice.

MATERIAL AND METHOD

Seventeen personnel from the Royal Dental School, Malmö, whose ages ranged from 18 to 28 years were the subjects of the investigation. The gingival crevices of the six upper anterior teeth were studied. The associated gingivae were in a state of excellent clinical health.

The mouths were prepared with the aid of a saliva ejector and absorbant rolls to keep the area under investigation dry. Disinfection of the teeth and gingivae was carried out by applying the iodine-colofonium solution described by *Bervell* (1960). This solution dries to form a skin in 3 minutes. Bacterial samples were then taken from the gingival crevices after perforation of the disinfectant »skin» by sterile steel blades which were $5 \times 1.5 \times 0.05$ mm. They were held by sterile college dressing tweezers and carefully inserted to the base of the gingival crevice. The blades were placed in the buccal, mesial and distal areas in order to take

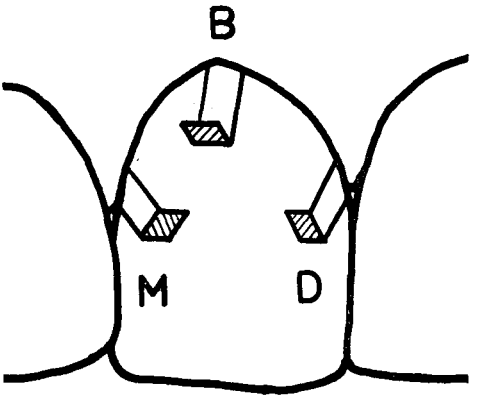

Fig. 1.
Position of steel blades when taking bacterial samples.
M—mesial, D—distal, B—buccal/palatal.

separate samples from these different regions (see Fig. 1). Samples from the palatal area were taken on a separate occasion. One week after the first samples had been taken, new samples were obtained from the buccal region.

Control samples were taken both from the surface of the disinfectant »skin» and from the tooth or gingival surfaces after the »skin» had been scraped away.

Duplicate samples were obtained from the test and control areas in order to allow aerobic (Trypticase Soy Broth*) and anaerobic (Thioglycollate**) cultures to be undertaken. When the bacterial samples had been taken, the steel blades were dropped directly into the media. The cultures were read for either growth or no growth after 5 days incubation at 37°C.

RESULTS

A total of 1,020 bacterial samples were taken from the gingival crevices. The distribution of the resulting positive and negative cultures can be seen from Table 1. Two samples (one aerobic and one anaerobic) were taken from each area in the crevice for each of the six teeth. Thus, the maximum number of positive

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Table 1

Bacterial samples from different areas of the gingival crevice.

Subject	Number of positive bacterial samples				
	Mesial	Distal	Palatal	Buccal 1st	Buccal 2nd
1	11	10	11	3	2
2	8	9	6	7	9
3	12	12	12	5	2
4	11	12	12	3	11
5	12	11	12	2	5
6	10	10	11	7	5
7	10	8	10	4	4
8	6	12	12	2	0
9	9	11	8	0	3
10	12	12	8	7	11
11	12	12	12	6	10
12	12	11	12	4	4
13	12	11	12	9	11
14	12	12	12	11	11
15	8	7	12	0	2
16	12	12	11	3	4
17	12	12	12	4	5
Mean	10.5 (90 %)	10.8 (90 %)	10.9 (91 %)	4.5 (38 %)	5.8 (49 %)

cultures which could occur for any one area in any subject is 12.

It can be seen from Table 1 that whilst 90 % of the mesial and distal, and 91% of the palatal samples were positive, only 38% of the first series of buccal samples gave growth. In view of these low buccal values further buccal samples were taken after one week. The average figure recorded this time was 49%.

172 control samples were taken. From the resultant cultures 1 was positive whilst 171 were negative. This demonstrates that the iodine-colofonium solution appears to be an effective disinfectant when applied to either tooth or mucous membrane surfaces.

DISCUSSION

Bacterial samples taken from the gingival crevice after disinfection of the teeth and gingivae showed that bacteria were regu-

larly present in the healthy crevice. When samples from the different areas were studied, however, the occurrence of positive bacterial samples was seen to vary according to the region of the crevice under investigation. Whilst about 90% of the mesial, distal, and palatal cultures gave growth only half of this number of buccal samples were positive.

The low incidence of positive cultures from the buccal region could be due to the infrequent occurrence of bacteria in this part of the crevice. It is possible that negative cultures might be due to disinfection of the crevice by penetration of the disinfecting solution before it had dried. The shallower the crevice, the greater is the possibility for disinfection to occur (see *Krasse & Brill* 1960 for further discussion). If, however, the average crevice depths for the different regions are considered, the differences in bacterial findings reported in this study do not seem likely to be explained by differences in crevice depths. *Forsberg* (1951), who measured crevice depths for upper incisors, found an average figure of 1.6 mm for the inter-proximal region and 1.1 mm for the palatal region. In the present investigation bacterial samples from the inter-proximal and palatal regions gave a similar incidence of positive cultures; thus the difference in crevice depth between these areas, 0.5 mm, appeared to have no influence on the result of the bacterial samples. The crevice depth for the buccal region was found by Forsberg to be 0.9 mm. The difference in depth between palatal and buccal regions, 0.2 mm, is comparatively small. This difference, therefore, is not considered to be the only reason for the discrepancy in bacterial findings between the palatal and buccal areas.

The low incidence of bacteria in the buccal region and their high incidence in the inter-proximal and palatal regions are interesting because they parallel the eventual distribution of disease occurring in periodontitis, as estimated by measurements of pocket depth. The greater frequency of deep periodontal pockets inter-proximally and palatally has been related to larger amounts of subgingival calculus and inefficient oral hygiene on these surfaces (*Lövdal, Arnö & Waerhaug* 1958). In thus seems reasonable to suggest that the differences in bacterial findings when the mesial, distal, and palatal regions are compared to the buccal region of the healthy crevice, may be due to the ease of access

to, and thus better cleansing of, the buccal area during oral hygiene procedures.

SUMMARY

The bacterial state of the clinically healthy gingival crevice has been investigated. Individual samples from the mesial, distal, palatal, and buccal areas were taken after disinfection of the neighbouring tooth and gingival surfaces.

Bacteria were found to be present in all the crevices studied. Some differences were found when the different areas were compared. Bacteria were found twice as frequently in the mesial, distal, and palatal regions as in the buccal area. It is suggested that these differences may be explained by the relative ease of access to the various areas during toothbrushing.

RÉSUMÉ

LES CONDITIONS BACTÉRIENNES DANS LES DIFFÉRENTES RÉGIONS DU SILLON GINGIVO-DENTAIRE CLINIQUEMENT SAIN

Les conditions bactériennes dans le sillon gingivo-dentaire cliniquement sain ont été étudiées. Des prélèvements individuels ont été faits sur les zones mésiales, distales, palatines et vestibulaire après désinfection des dents voisines et des surfaces gingivales.

La présence de bactéries a été constatée dans tous les sillons gingivo-dentaires étudiés. La comparaison des différentes régions a mis en évidence quelques différences. Les bactéries ont été trouvées deux fois plus souvent dans les régions mésiales, distales et palatines que dans les zones vestibulaires. Il est possible que ces différences puissent s'expliquer par la facilité relative d'accès aux différentes régions au cours du brossage des dents.

ZUSAMMENFASSUNG

DER BAKTERIELLE ZUSTAND IN VERSCHIEDENEN REGIONEN DER KLINISCH GESUNDEN ZAHNFLEISCHTASCHE

Der bakterielle Zustand in der klinisch gesunden Zahnfleischtasche wurde untersucht. Separate Bakterienproben von den mesialen, distalen, palatinalen und buccalen Teilen der Zahn-

fleischtasche wurden nach Desinfektion der angrenzenden Zahn- und Zahnfleischoberflächen entnommen.

In allen untersuchten Zahnfleischtaschen konnten Bakterien nachgewiesen werden. Gewisse Unterschiede konnten beim Vergleich der verschiedenen Regionen festgestellt werden. Bakterien wurden doppelt so oft in den mesialen, distalen und palatinalen Regionen als in den buccalen angetroffen. Es wird vorgeschlagen diese Unterschiede mit der relativen Leichtigkeit die verschiedenen Oberflächen während des Zähneputzens zu erreichen, zu deuten.

REFERENCES

Bernier, J. L., 1950: The histologic changes of the gingival tissue in health and periodontal disease. Oral Surg. *3:* 1194—1199.

Bervell, S. F. A. 1960: En ny teknikk ved bakteriologisk undersökelser av fysiologiske tannkjöttlommer. Norske tannlægeforen. Tid. *70:* 425—433.

Blumentritt, W., 1926: Die Tiefenverhältnisse der physiologischen Zahnfleischtasche und der physiologische Zahndurchbruch. Vierteljahrschr. f. Zahnheilk. *42:* 182—192.

Boyd, W. S. & S. L. Rosenthal, 1958: The presence of bacteria in the healthy gingival sulcus. J. dent. Res. *37:* 288—291.

Castenfelt, T., 1952: *Toothbrushing and Massage in Periodontal Disease.* Thesis. Nordisk Rotogravyr, Stockholm.

Cohen, B., 1959: Morphological factors in the pathogenesis of periodontal disease. Brit. dent. J. *107:* 31—39.

Fish, E. W., 1944: *Parodontal Disease.* Eyre & Spottiswoode, London.

Forsberg, A., 1951: *The Periodontal Tissues of the Upper Incisors.* Thesis. Acta odont. scand. *9:* suppl. 8.

Gavin, J. B. & A. A. Collins, 1961: The occurrence of bacteria within the clinically healthy gingival crevice. J. Periodont. *32:* 198—202.

Krasse, B. & N. Brill, 1960: Effect of consistency of diet on bacteria in gingival pockets in dogs. Odont. Revy *11:* 152—165.

Lövdal, A., A. Arnö & J. Waerhaug, 1958: Incidence of clinical manifestations of periodontal disease in light of oral hygiene and calculus formation. J. Amer. dent. Ass. *56:* 21—33.

Massler, M., A. Cohen & I. Schour, 1952: Epidemiology of gingivitis in children. J. Amer. dent. Ass. *45:* 319—324.

Rothner, J. T. & B. B. Saturen, 1954: The gingival sulcus: A clinical study of its depth. J. Periodont. *25:* 278—281.

Scherp, H. W. & G. W. Burnett, 1956: Host-parasite interactions in relation to dental disease. Norske tannlægeforen. Tid. *66:* 15—24.

Schultz-Haudt, S. D., 1956: Bacteria and gingivits. Norske tannlægeforen. Tid. *66:* 25—31.

Schultz-Haudt, S. D., 1960: Biochemical aspects of periodontal disease. World Health Organization report. DH 32.

Waerhaug, J., 1952: *The Gingival Pocket*. Thesis. Odont. Tidskr. *60:* suppl. 1.

Waerhug, J. & E. Steen, 1952: The presence or absence of bacteria in gingival pockets and the reaction in healthy pockets to certain pure cultures. Odont. Tidskr. *60:* 1—32.

Zachinsky, L., 1954: Range of histologic variation in clinically normal gingiva. J. dent. Res. *33:* 580—589.