# EXFOLIATIVE CYTOLOGICAL STUDY OF GERMS AND YEASTS IN PERIODONTAL DISEASE

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SUMMARY: A number of different microorganisms were detected in different stages of periodontitis. Various clinical stages of the disease were related with the presence of factors such as tobacco and alcohol use. The results obtained demonstrate a significant relationship between the variable severity of the lesion and smoking. The analysis of periodontal smears showed that the population with severe lesions who were smokers accounted for most of this difference. There was no statistically significant association between severity of periodontitis and the variables alcohol use, sex or presence of any of the microorganisms studied.

Key-words: Exfoliative cytology, Periodontitis, Microorganisms

### INTRODUCTION

Inflammation of the tissues supporting the teeth constitute in themselves nosological entities of considerable importance, owing to their frequent appearance in man (1). However, despite numerous studies, the pathogenetic mechanisms in these processes have yet to be fully clarified. Factors involved in pathogeneis include, depending on the circumstances, the cytological and proteolytic effects of bacterial plaque, immediate cell-modulated hypersensitivity reactions against bacterial antigens, and, to a similar degree, indirect lesions derived from the host's own immune response (2) as a result of the action of neutrophils, complement fractions, plasma cells and cytotoxic T cell subsets (3). This aspect of periodontal inflammation led us to investigate the presence of bacteria. yeasts and cells of the immune system in the diseased area, in an attempt to relate the severity of periodontal lesions with factors such as alcohol

and tobacco use and diabetes, which are considered to predispose the patient to such infection. We also compared the clinical findings with laboratory data to assess the disease and prognosis more accurately.

# MATERIALS AND METHODS

A total of 98 subjects were studied, of whom 78 had different degrees of periodontal disease and 20 served as disease-free controls. No attempt was made to include equal numbers of subjects with similar degrees of disease severity, but depending on the clinical findings the subjects were subsequently classified in three different groups: Mild form (inflammatory pocket less than 3 mm in diameter), Moderate form (3-6 mm in diameter) and Serious form (more than 6 mm in diameter).

An upper and a lower smear were obtained from each subject in both the study and control

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groups. The smears were taken from the most seriously affected intercanine area as near the gingival margin as possible, fixed in 1:1 alcohol-ether for 24 h and stained with the classical technique of Pappanicolau.

We studied the following parameters:

- 1) Age. The subjects were divided into three groups: 23-45 years (37,76%), 46-67 years (41,84%) and more than 68 years (20,41%). The cut-off age of 45 years for the first group was chosen as approximately representing the end of the reproductive period in women; 67 years (second cut-off) was chosen to represent retirement age; the third group comprised elderly subjects.
- 2) Sex. Males made up 53,06% of the sample population; females made up 46.94%.
- 3) Alcohol use. Thirty-two point sixty-five per cent of the participants consumed alcohol regularly.
- 4) Tobacco use. Twenty-eight point fifty-seven per cent were smokers.
- 5) Severity of periodontal disease. Controls: 20,41%; Mild form, 23,47%; Moderate form, 21,43%; Severe form, 34,69%.
- 6) Type of bacteria. Bacteria were classified according to Burnett's system (4) for bacteria in the oral cavity (1982) as endococcoid, filiform or bacillar.
  - 7) Presence of yeasts.
  - 8) Presence of inflammatory cells.

#### RESULTS

The relationship between sex and severity of periodontal disease is shown in Figure 1, which illustrates the numbers of males and females in each group (control, mild, moderate, or severe). The statistical analysis failed to demonstrate any significant association between sex and disease severity (p=0.054), although more males (24,49%) than females (10%) had severe disease.

Figure 2 shows the numbers smokers and nonsmokers with each form o periodontal disease. Significant differences were found between smokers and nonsmokers in the severity of the disease (p=0.039). These values also show that 50% of the 28 habitual smokers had severe periodontal disease, whereas nonsmokers were distributed evenly in the different groups. Of the 32 regular consumers of alcohol, 11 had severe, 9 moderate and 4 mild

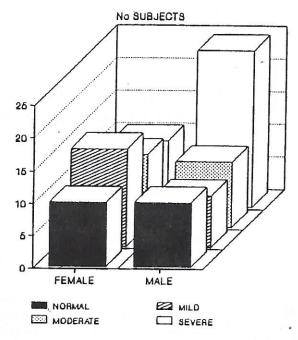


Fig. 1 — Relationship between severity of periodontal disease and sex.

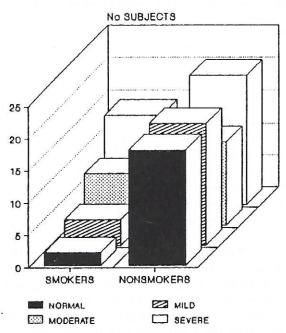


Fig. 2 — Relationship between severity of periodontal disease and tobacco use.

disease, while 11 were disease-free controls. These data indicate that alcohol use had no apparent relationship with severity of the disease (p > 0.05).

Of the 98 subjects studied, coccoid bacteria

were detected in 92, but no significant relationship was found between periodontal disease and the presence of coccoids (p > 0.05). Figure 3 shows the distribution of subjects with different forms of

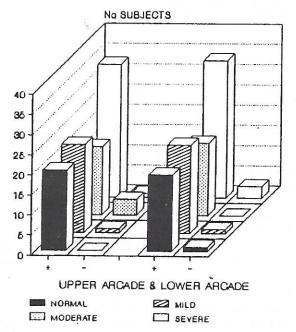


Fig. 3 — Relationship between severity of periodontal disease and presence of coccoid bacteria.

disease according to the presence or absence of coccoids in the upper and lower arcade. The numbers of subjects with coccoid, filiform or bacillar bacteria, yeast or inflammatory cells are presented in Table 1. Filiform bacteria were found in the upper arcade in only 31 of the 98 subjects, with no significant association between the presence of these germs and periodontal disease. Filiforms were found in the lower arcade in 37 of the 98 subjects, the results being similar in overall terms to those in the upper arcade. No significant relationship was found between the presence of filiforms and disease severity. Bacillar forms were detected in the upper arcade in 4 patients, and in the lower arcade in 3.

Yeasts were detected in the upper arcade in 24 subjects, and 75% of this group had severe periodontal disease. None of the control subjects had yeast-like forms in the upper arcade. In the lower arcade, yeasts were found in 37 subjects, the lmajority of whom (54%) had severe disease.

The relationship between lymphoid (inflammatory) or polymorphonuclear cells and the different froms of periodontal disease was similar to that found for other variables. Inflammatory cells were present in the upper arcade in 79 of 98 subjects, becoming more common as severity of the disease increased. Similar observations were obtained in

**Table 1.** Numbers of subjects with bacterial or yeast infections according to the severity of periodontal disease.

Agent	Arcade	Severity of periodontal disease			
		Controls	Mild	Moderate	Severe
Coccoid	Upper	20	22	17	33
	Lower	19	22	18	34
Filiform	Upper	7	2	4	7
	Lower	. 10	5	9	12
Bacillar	Upper	1	0	1	3
	Lower	0	1	0	1
Yeast	Upper	0	4	2	18
	Lower	0 2	6	9	20
Inflammatory					
cells	Upper	15	20	17	27
	Lower	20	20	21	31

the lower arcade, although no statistically significant association between the two variables was found.

#### DISCUSSION

Our results show that males have a greater tendency toward more severe forms of periodontal disease in comparison to females. This trend, which however was not statistically significant, may be due to te fact that women are more often motivated for esthetic reasons to seek dental care before the disease reaches an advanced stage. Thus of the 42 men in our study with periodontal disease, 57% had severe periodontis as compared to 27% of the women with the disease. Among patients with the mild form, 41% of the women in contrast to 19% of the men fell into this class. Ramfjord and Major (5) reported periodontal disease to be more frequent and objectifiable in women due to hormonal conditioning.

Of the other factors potentially related with periodontal disease, smoking has frequently been investigated. In our series, smoking was apparently unrelated to periodontitis when the subjects were classified according to the severity of disease.

The data given in Table 1 indicate no significant difference between controls and patients in terms of bacterial infection by coccoids or bacillar forms. This apparently contradicts the findings of Farida et al. (6), who determined the serum titers of antobodies against lipopolysaccharides of different bacteria, and is also in disagreement with Sandholm et al. (7), who found anti-actinomycetic IgG. Pertuiset et al. (8) reported that certain bacteria were pathogenetic in the site of the periodontal lesion.

Levanchy et al. (9) studied the effect of composition of the microbial flora in gingival curretages of periodontal disease in different stages, and noted a decrease in the porportion of spirella and filiforms, while coccoids increased significantly. After a certain time however, the distribution of bacterial species tended to return to its initial values. The results of our study do not suggest any significant variation in the composition of the microbial flora in any given stage of periodontitis.

As shown in Table 1, the presence of inflamma-

tory cells of the immune system did not vary significantly depending on disease severity. However, Greenspan (3) observed a higher percentage of polymorphonuclear cells and T lymphocytes in early stages of the disease. These elements were gradually replaced by activated B lymphocytes in more advanced or more severe cases.

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