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Prevalence of periodontitis and suspected periodontal pathogens in families of adult periodontitis patients

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Abstract. The aim of the present study was to investigate the prevalence of periodontopathic microorganisms and periodontal destruction in the spouses and children of adult periodontitis patients. For this study, 24 families were selected on the basis of one parent with severe periodontal breakdown and the presence of *Actinobacillus actinomycetemcomitans* and/or *Porphyromonas gingivalis* and/or more than 30% *Prevotella intermedia* subgingivally. The clinical examination of both parents and children included pocket depth and clinical attachment loss (CAL) measurements. Samples for bacterial examination were obtained from the mucous membranes, the saliva and pockets. Pocket selection was based on the most advanced periodontitis situation found in a subject. The samples were cultured for the detection of *A. actinomycetemcomitans*, *P. gingivalis* and *P. intermedia*. By phase-contrast microscopy, the % of spirochetes and motile microorganisms was assessed. The number of children within each family varied between 1 and 3. In total 49 children were investigated with a range in age of 3 months to 15 years. Results showed that under the age of 5 years, none of the children had CAL, whereas in the age group of 5-15 years, 26.5% had 1–5 sites in the primary and/or permanent dentition with 1–3 mm CAL. 3 of the spouses had no interproximal CAL. 16 of the 24 spouses had a light to moderate form of periodontitis, with at least one site with 1 to 4 mm CAL and 5 spouses had severe periodontal breakdown with sites showing at least 8 mm CAL. Spirochetes, motile microorganisms and *P. intermedia* were frequently present in all family members. 18 out of the 24 probands were positive for *P. gingivalis*. This organism was found once only in a 5-year-old boy and in 11 of the spouses. *A. actinomycetemcomitans* was detected in 13 probands; 5 children and 5 of the spouses were also positive for this bacterium. If a child harboured one of the periodontopathogens, at least 1 of the parents was also positive for that bacterium. This phenomenon may be due to transmission of microorganisms between family members. Comparison of the clinical data reported in the present study with similar clinical parameters from epidemiological studies of the Dutch population suggest that the spouses and children of adult periodontitis patients might be at relatively high risk of developing periodontal breakdown.

Key words: periodontitis; microbiology; families.

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Periodontal diseases have primarily a microbiological etiology (Loesche 1982, Socransky & Haffajee 1992). The interaction of the periodontal microbiotas with the host modulates their clinical expression and the nature of this host-parasite interaction depends on the manner that each individual has interacted with its environment since birth. If early childhood years are a critical period for the acquisition of certain bacteria, it seems likely that many infants acquire the organisms from their parents or other persons with whom they have close house-hold contact. A parent with severe adult periodontitis, who is infected with bacteria associated with periodontal disease, may function as a source of infection. Their children may be at a greater risk of becoming colonized with bacteria which are associated

with periodontal diseases, compared to children of periodontally healthy parents. If acquisition of certain periodontopathic microorganisms is possible at a later age in a stabilized oral flora, the spouses may be at risk to become colonized and subsequently develop periodontal breakdown. Many gaps still exist in the mode of transmission of periodontopathic microorganisms and the conditions that influence it. More knowledge on this subject may have direct bearing on strategies aimed at the prevention of transmission of suspected periodontal pathogens and possibly of periodontal destruction itself.

Family studies on the prevalence of periodontitis have been limited to families selected around probands of juvenile periodontitis (JP) patients (Ohtonen et al. 1983, Vandesteen et al. 1984, Spektor et al. 1985, Page et al. 1985, Beaty et al. 1987) and microbiological data in such investigations only reported on the intrafamilial distribution of *Actinobacillus actinomycetemcomitans* (Zambon et al. 1983, DiRienzo & Slots 1990, DiRienzo 1991). The results of these studies provide evidence showing familial aggregation of this specific periodontal disease and intrafamilial transmission of *A. actinomycetemcomitans*.

Besides *A. actinomycetemcomitans*, several bacterial species are considered to play a prime etiologic role in the initiation and development of periodontitis due to their frequent isolation from periodontal lesions and because of their pathogenic potential. *A. actinomycetemcomitans*, *Porphyromonas gingivalis*, *Prevotella intermedia*, spirochetes and motile microorganisms have been the subject of extensive research and appear to assume particular significance in various forms of severe periodontal disease (Zambon 1985, Greenstein & Polson 1985, Van Winkelhoff et al. 1988a). Little information is available at which time the initial establishment of these periodontopathic bacteria is possible and when this generally occurs. *P. intermedia* has been detected in edentulous children as early as one month after birth, while *A. actinomycetemcomitans* and *P. gingivalis* could not be detected in these children (Frisken et al. 1990, Könönen et al. 1992). The presence of *A. actinomycetemcomitans*, *P. gingivalis*, spirochetes and motile microorganisms in the oral cavity of 4 to 6 year old children has been reported (Alaluusua & Asikainen 1988, Alaluusua et al. 1991, Frisken et al. 1987, Sanches 1985, Loesche 1988). In clinical and microbiological studies of the oral flora in children, certain age groups are usually studied without any information on the periodontal or microbiological background of their families (Frisken et al. 1990, Frisken et al. 1987, Alaluusua & Asikainen 1988, Van Oosten 1988). It may be presumed that children obtain many microorganisms from their parents, with whom they have frequent contact. By studying the children of adult periodontitis patients, who are infected by periodontopathogens, it may be possible to obtain an impression of the youngest age at which colonization with certain periodontopathogens is possible.

The aim of the present study was to investigate the prevalence of periodontopathic microorganisms and periodontal destruction in the spouses and children of adult periodontitis patients.

Material and Methods

Selection of the families

For this study, 24 probands were selected on the basis of the following criteria: (1) untreated adult periodontitis defined as the presence of at least 10 pockets with a pocket depth (PD) $\geqslant 5$ mm and at least one pocket with clinical attachment loss (CAL) $\geqslant 4$ mm; (2) the subgingival presence of *A. actinomycetemcomitans*, and/or *P. gingivalis* and/or more than 30% *P. intermedia*; (3) no history of periodontal treatment; (4) no history of antibiotic therapy during the last 6 months; (5) part of a family with at least one child of 16 years of age or younger.

All probands had been referred to the department of Periodontology of the Academic Centre for Dentistry Amsterdam (ACTA) for diagnosis and treatment of severe periodontitis. At their first visit patients were asked to complete a questionnaire about their family. If the radiographs showed severe loss of alveolar bone support and the clinical examination revealed deep pockets which bled on probing, permission was asked for further microbiological examination. Subgingival samples were screened for the presence of *A. actinomycetemcomitans*, *P. gingivalis*, and *P. intermedia* by indirect immunofluorescence with polyclonal antisera.

If a subject fulfilled the criteria and was willing to participate in the study, an appointment was made for clinical and microbiological examination of the whole family, i.e. spouse and children.

Clinical examination

The clinical examination was performed interproximally at all teeth. The following assessments were made: plaque index (Silness & Löe 1964); colour of the gingiva (scored as: 0=no redness; 1=redness); swelling of the gingiva (scored as 0=no swelling; 1=light swelling; and 2=obvious swelling); bleeding on probing (BOP) according to the sulcus bleeding index (scored as 0=no bleeding; 1=point bleeding within 30 seconds; 2=immediate overt bleeding) (Mühlemann & Son 1971); pocket depth (PD; measurements were rounded off to the nearest mm marking); if interproximal clinical attachment loss (CAL) was present, the CAL was recorded at the sample sites (by measuring the distance from the cemento-enamel junction to the bottom of the pocket). The clinical examination of the adults was performed with a *Merrit b* (Hu Friedy) periodontal probe. The children were examined with a *Michigan no. 0* probe (Hu Friedy). The youngest children which were examined were 3 years of age. The clinical parameters were only scored when the teeth were at the level of the occlusal plane.

Microbiological procedures

Prior to any clinical measurement, samples for microbiological analysis were obtained in the following order: the buccal mucosa, the dorsum of the tongue, the tonsillar area, the saliva and a pooled sample of 4 pockets. The mucous membranes were sampled with a sterile swab and were immediately suspended in reduced transport fluid supplemented with Fildes (RTFF) (Petit et al. 1991). Approximately 1 ml of saliva was added to 1 ml of RTFF. After the clinical examination 4 pockets were chosen for bacterial sampling according to the following criteria and in the following order: (1) the deepest pocket with the greatest amount of attachment loss and BOP; (2) if no attachment loss was found, the deepest pocket which showed BOP was chosen; (3) if BOP was absent then the deepest pocket was chosen; (4) if only shallow healthy pockets were present, samples were taken mesially from the first primary and/or permanent molars. From children who were not examined clin-

ically, no pocket samples were taken. After removal of supragingival plaque, a pooled subgingival plaque sample was taken from 4 pockets by inserting 2 sterile paperpoints per pocket during 10 s. and immediate suspension in RTFF. All samples were processed in the laboratory within 2 h.

Phase-contrast video recordings of the samples were made as previously described (Petit et al. 1991). Appropriate dilutions were plated with a spiral plater (Spiral System™, Spiral Systems, Inc. Cincinnati, U.S.A.) on blood agar plates (Oxoid no. 2 Basingstoke U.K.), supplemented with 5% horse blood, hemin (5 mg/l), and menadione (1 mg/l) and Tryptic soy-Serum-Bacitracin-Vancomycin (TSBV) agar plates (Slots 1982). The blood agar plates were incubated anaerobically in 80% N_2, 10% H_2 and 10% CO_2 for up to 21 days. On the blood agar plates the total number of colony forming units (CFU) and the number of dark-pigmented colonies were counted. Representative black, brown, greenish and cream coloured colonies were streaked to purity and identified using standard techniques, including Gram stain, hemagglutination of 3% sheep erythrocytes, fermentation of carbohydrates, production of indole, and, if necessary production of specific enzymes with ATB 32A (API Biomerieux, La Balme, Les Grottes, France) (Van Winkelhoff et al. 1988b).

The TSBV plates were incubated in a CO_2 incubator, 5% CO_2 in air, at 37°C (van Steenbergen et al. 1986). After 5 and 7 days, the plates were examined for the presence of *A. actinomycetemcomitans*. The primary criteria for identification were colony morphology, the presence of a star-like inner structure and catalase production. Suspected colonies were streaked to purity and a definite identification was made on the basis of Gram stain, catalase, enzyme pattern on API-ZYM (API Biomerieux).

Statistical analysis

The Mann-Whitney *U*-test was applied to test possible differences between the various patient groups. Correlations between clinical parameters (colour and the swelling of the gingiva, BOP, PD, mean PD, % PD ≤ 3 mm, % PD ≥ 5 mm, % PD ≥ 6 mm, % PD ≥ 7 mm, % PD ≥ 8 mm and CAL at the sample site) and prevalence of selected microorganisms were investigated. To test possible differences in prevalence of specific microorganisms in different groups, the chi-square analysis was applied. Logistic regression analysis and multiple regression analyses were applied to analyze the relation of the prevalence of the different microorganisms and clinical parameters to age of the children. *P*-values <0.05 were accepted as statistically significant.

Results

Description of the families

This study included 24 families, each consisting of a proband, a spouse and one to three children. The mean age of the probands and the spouses was 38.5 and 37.7 resp., with a range from 30 to 50. In total 49 children were investigated with an age ranging from three months to 15 years. The group consisted of 20 Caucasian families born in the Netherlands, three non-Caucasian families from Surinam and one Turkish family. The mean number of years the couples were living together was 13.7 with a range from 8 to 26 years.

Periodontal condition

All probands suffered from severe (generalized) periodontitis with a mean PD of 5.3 mm and a mean maximum CAL of 9.1 mm (Table 1). The spouses had lower plaque scores, less bleeding on probing, a lower mean PD (Table 2; $p<0.0001$).

The clinical data of the children are shown in the Table 3. 9 children could not be examined clinically due to their young age (≤ 4 year) and poor cooperation. In the other 40 children the level of BOP, the mean PD and the % of PD deeper than 3 mm increased with increasing age of the children ($p<0.05$). None of the children under the age of 5 years was affected by periodontitis whereas in the group of 5–15 years 26.5% had ≥ 1 site showing a slight amount of CAL (CAL = 1–2 mm and PD ≥ 5 mm). Only 1 10-year-old girl showed 3 mm CAL affecting her primary molars. For all children, CAL was limited to interproximal sites of the primary molars, permanent first molars and the second premolars.

Microbiological evaluation

The prevalence of the selected microorganisms in the probands and the spouses is shown in the Tables 1, 2. A subject was considered positive if at least one of the samples was positive for the specific microorganism. *A. actinomycetemcomitans* and *P. gingivalis* were more prevalent in the probands than in the spouses, while *P. intermedia*, spirochetes and motile microorganisms were present in almost all adults. The adults positive for *P. gingivalis* were older than the adults negative for this microorganism ($p=0.0184$). The prevalence and the mean percentage ($\pm$SD) of the cultivable flora of the suspected periodontal pathogens per site can be seen in Tables 4 and 5. All microorganisms were frequently isolated from the oral mucous membranes and the saliva, except for the motile microorganisms and the spirochetes. These latter morphotypes were absent in the samples from the buccal mucosa. Motiles were present on the tongue in 87.5% of the adults in high percentages (probands 27.6%$\pm$18.3; spouses 36.1%$\pm$17.0) whereas spirochetes were only present in 2 adults at this site. In Table 5, it can be seen that in 10 cases, 1 of the microorganisms could be detected in the samples of the saliva or on the mucous membranes of a spouse but not in the pocket. In all probands positive for one of the selected bacteria, this specific bacterium was always detected in the pooled pocket sample.

The Tables 3 and 6 show the microbiological data of the children. *P. gingivalis* was isolated from one 5-year-old boy from the cheeks, the tonsillar area and the saliva, but not from the subgingival sample. *A. actinomycetemcomitans* was detected in 5 children of which the youngest was three years of age. If *A. actinomycetemcomitans* was detected in a child this microorganism was always present in the pocket sample. In the children, the occurrence of *P. intermedia* was related to age ($p<0.05$). The youngest child in which *P. intermedia* was detected was 3 years of age. All children of 8 years of age and older were positive for this microorganism. The youngest child colonized with motile microorganisms was 9 months of age. Motiles were found with values reaching 68% in the samples of the tongue of 37 of 44 children who were positive for these microorganisms. Motile microorganisms were regularly present in the samples of all sites of the children, except for the samples of the buccal mucosa. Thirteen children were positive for spirochetes although in low percen-

Table 1. Clinical and microbiological description of the probands

Fam.	Sex	Age (years)	LT	PI	SBI	%pd ≤3 (mm)	%pd ≥6 (mm)	Mean PD (mm)	Max LA (mm)	Presence or absence				
										Pi	Pg	Aa	spi	mmo
1	M	30	10	1.4	1.6	36	18	4.3	7	+	−	+	+	+
2	M	50	10	1.3	1.8	5	33	5.3	10	+	+	−	+	+
3	M	37	15	1.9	2.0	0	54	6.8	10	+	+	−	+	+
4	M	43	8	0.8	1.5	65	10	5.0	8	+	+	−	+	+
5	F	31	13	1.3	1.9	4	62	6.2	6	+	+	+	+	+
6	M	38	13	1.0	1.4	71	8	3.7	6	+	+	+	+	+
7	F	31	11	0.9	1.8	30	34	5.1	9	+	+	+	+	+
8	M	37	9	1.7	1.4	33	37	4.9	9	+	+	+	+	+
9	M	35	10	2.0	1.9	48	9	4.1	4	+	+	+	+	+
10	F	42	8	1.8	1.5	69	24	4.1	14	+	−	+	−	+
11	M	42	8	1.7	1.3	61	19	3.9	8	+	+	−	+	+
12	M	37	17	1.4	1.7	15	25	4.9	8	+	−	+	+	+
13	M	38	15	1.6	1.6	29	52	5.5	14	+	+	+	+	+
14	M	35	13	1.8	1.7	28	38	5.0	10	+	−	+	−	+
15	F	32	14	2.0	0.4	0	93	7.2	11	+	−	+	+	+
16	F	43	23	0.9	1.6	18	38	5.5	10	+	+	−	+	+
17	M	38	19	0.8	1.9	0	71	6.3	10	+	+	−	+	+
18	F	34	10	1.9	1.8	16	39	5.2	6	+	+	−	+	+
19	M	45	21	1.6	0.6	6	62	6.0	9	+	+	−	+	+
20	M	49	26	1.7	1.4	50	39	5.4	14	+	+	+	+	+
21	M	45	9	1.3	1.7	6	31	5.2	6	+	+	+	+	+
22	M	38	13	1.8	1.8	48	23	4.5	9	+	+	−	+	+
23	M	36	18	1.5	1.8	13	60	5.8	7	+	−	−	+	+
24	M	39	15	1.8	1.7	9	71	7.5	14	+	+	−	+	+
Mean ±SD		38.5 ±5.4	13.7 ±4.9	1.5 ±0.4	1.6 ±0.4	27.5 ±23.6	39.5 ±21.9	5.3 ±1.0	9.1 ±3.1					

Fam = family; LT = number of years the couples are living together; PI = plaque index; SBI = sulcus bleeding index; %pd = percentage pocket depth; PD = pocket depth; max LA = maximum loss of attachment; prPg = presence of *P. gingivalis*; prAa = presence of *A. actinomycetemcomitans*; prspi = presence of spirochetes; prmmo = presence of motile microorganisms. nd = not determined.

tages. The youngest child with spirochetes was 3 years of age.

Distribution of the microorganisms in relation to clinical parameters

18 probands and 11 spouses were positive for *P. gingivalis* forming 10 positive couples. Of the 6 *P. gingivalis* negative probands, one spouse was positive for this bacterium. One of the couples had a child positive for *P. gingivalis* (Table 7). There were no statistically significant differences in the clinical parameters between the spouses positive or negative for *P. gingivalis*. This was also true if the *A. actinomycetemcomitans* positive spouses were excluded from the analysis. The ten couples with both subjects positive for *P. gingivalis* had been living together for a longer period (16.2 ± 5.8 years) than the eight couples with *P. gingivalis* negative spouses (10.7 ± 2.7; $p = 0.03$). However, no correlation could be found between the number of years the couples were living together and any of the tested clinical parameters of the spouses. The trend of the presence of more *P. gingivalis* positive spouses within the group of spouses of probands positive for this bacterium in the saliva, compared to the spouses of *P. gingivalis* positive probands without this bacterium in the saliva, failed to reach the level of significance ($p = 0.054$).

The intrafamilial distribution of *A. actinomycetemcomitans* was limited (Table 8). 13 of the 24 probands were positive for *A. actinomycetemcomitans*, however, only four of them had a spouse positive for *A. actinomycetemcomitans*. There was one spouse colonized with *A. actinomycetemcomitans*, whereas the proband was negative for this microorganism. One spouse (F8), who did not harbour this microorganism at the time the samples were taken, had been treated with a combination of metronidazole and amoxicillin for *A. actinomycetemcomitans*-associated periodontitis 1 year prior to the present investigation. From the 14 families in which one or both parents were positive for *A. actinomycetemcomitans*, 5 families (18%) had children positive for *A. actinomycetemcomitans*. Within the spouses and the children, no correlation between the clinical status and the presence or absence of *A. actinomycetemcomitans* could be found. 22 probands and 18 spouses were positive for spirochetes forming 17 positive couples. If a child was positive, at least 1 of the parents was also positive but in most cases both parents were positive (Table 9). In the affected group, more children were positive for spirochetes than in the unaffected group (Table 10; $p = 0.02$). All of the families were positive for *P. intermedia* as well as motiles. No differences between the clinical status and the presence or absence of these microorganisms could be found.

Discussion

It is generally acknowledged that not all subjects within a population are equally susceptible for the development of periodontitis. Periodontal diseases have a complex and multifactorial etiology and information on the contribution of genetic and shared environmental factors that account for the differences in susceptibility, is incomplete. Colonization with specific periodontopathic microorganisms at a young age may be one of the factors which influences the initiation of periodontal destruction. However, it is not established when peri-

Table 2. Clinical and microbiological description of the spouses

Fam.	Sex	Age (years)	LT	PI	SBI	%pd ≤3 (mm)	%pd ≥6 (mm)	Mean PD (mm)	Max LA (mm)	Presence or absence				
										Pi	Pg	Aa	spi	mmo
1	F	30	10	1.7	1.5	73	2	3.4	3	+	−	+	+	+
2	F	45	10	1.2	0.9	70	6	3.4	2	+	+	−	+	+
3	F	34	15	1.6	1.1	82	7	3.4	4	+	+	−	+	+
4	F	36	8	1.2	0.5	92	0	2.9	0	+	−	−	−	+
5	M	39	13	1.7	1.4	70	10	3.7	2	+	−	−	+	+
6	F	32	13	0.7	0.5	98	0	2.4	0	+	−	−	+	+
7	M	34	11	2.1	2.0	75	2	3.3	4	+	−	−	+	+
8	F	44	9	1.0	0.1	35	15	4.9	12	+	−	−*	+	+
9	F	38	10	0.8	0.8	78	0	2.8	8	+	+	−	+	+
10	M	44	8	1.4	1.9	73	7	3.4	3	+	−	−	−	+
11	F	32	8	0.9	1.1	88	0	2.9	1	+	−	−	+	+
12	F	36	17	0.9	0.6	54	28	4.4	10	+	−	−	+	+
13	F	34	15	1.1	1.1	84	0	3.1	1	+	+	−	−	+
14	F	35	13	0.8	0.8	70	0	3.2	2	+	−	+	+	+
15	M	36	14	1.4	0.6	76	0	3.0	2	+	+	+	+	+
16	M	44	23	1.9	1.5	93	0	2.7	1	+	+	−	+	+
17	F	36	19	0.9	0.5	96	0	3.0	2	+	+	−	−	+
18	M	38	10	1.0	1.7	41	26	4.4	9	+	+	−	+	+
19	F	45	21	1.6	1.0	89	0	2.9	1	+	+	−	+	+
20	F	50	26	0.8	1.2	46	9	3.9	9	+	+	+	+	+
21	F	40	9	1.3	1.2	85	0	2.9	0	+	−	−	−	−
22	F	37	13	0.3	0.8	77	5	3.4	3	+	+	−	+	+
23	F	33	18	1.2	1.0	88	0	2.9	2	+	−	−	−	−
24	F	33	15	1.0	1.2	86	0	2.9	4	+	−	+	+	+
Mean ±SD		37.7 ±5.2	13.7 ±4.9	1.2 ±0.4	1.0 ±1.0	75.7 ±16.7	5.0 ±8.1	3.3 ±0.6	3.5 ±3.4					

For legends, see Table 1.

* A treated *A. actinomycetemcomitans*-associated periodontitis patient.

odontopathic bacteria colonize the oral cavity and from what source these bacterial originate. It has been shown for *A. actinomycetemcomitans* in families with a high occurrence of JP, that parents may be the primary source (Zambon et al. 1983, DiRienzo 1991). Therefore, adult periodontitis patients may function as a source of infection for their family members, i.e. children and spouse.

Most of the microbiological studies in children prior to 1980 mentioned the presence of black-pigmented gram-negative anaerobes, that were classified as '*Bacteroides melaninogenicus*' (De Araujo & MacDonald 1964, Bailit et al. 1964, Mackler & Crawford 1973). These bacteria now include two new genera, *Porphyromonas* and *Prevotella* (Shah & Collins 1988, 1990); therefore it is difficult to find out which of these species were studied by previous investigators. In more recent studies *P. intermedia*, but not *P. gingivalis*, has been detected in edentulous children as early as one month after birth (Frisken et al. 1990, Könönen et al. 1992). Culture studies on the prevalence of black-pigmented gram-negative anaerobes in healthy children in the age range of 5 to 16 years, show that *P. intermedia* is quite frequently found as part of the oral flora, whereas *P. gingivalis* is rarely detected (Delaney et al. 1986, Yanover & Ellen 1986, Frisken et al. 1987, Wojcicki et al. 1987, Ashley et al. 1988, Van Oosten et al. 1988, Frisken et al. 1990). In the present study we isolated *P. gingivalis* only from 1 out of 36 children, all having *P. gingivalis* positive parents. This finding strengthens the view that colonization with *P. gingivalis* usually does not occur at an early age.

The results of the present study concerning the time of colonization and prevalence of *A. actinomycetemcomitans* in young children are in agreement with previous studies in which this bacterium was not detected in study populations under the age of 2.5 years (Frisken et al. 1990, Könönen et al. 1992) and could be detected in the primary dentition of healthy children from 4 to 7 years of age (Alaluusua & Asikainen 1988). Intrafamilial transmission of *A. actinomycetemcomitans* in families with a high prevalence of juvenile periodontitis has been suggested by several authors (Zambon 1983, DiRienzo & Slots 1990). In families of adult periodontitis patients only in 5 out of the 14 *A. actinomycetemcomitans* positive families (5 out of 28 children) was one child colonized with *A. actinomycetemcomitans*. In a similar study by Alaluusua et al. (1991) in 2 out of 9 families, children harbouring *A. actinomycetemcomitans* were found, which is comparable to our data. Therefore, transmission of the bacterium between parents and children may be possible but does not seem to occur easily. If a child was positive for *A. actinomycetemcomitans* at least 1 of the parents was positive for this microorganism.

Relatively little information is available on the prevalence of spirochetes in children. Loesche (1988) has detected spirochetes in the dental plaque of about 44% of the 3- to 5-year-old children and in about 50% of the 6- to 13-year-old children examined. Their numbers were less than 0.5% of the flora. In a study by Mikx et al. (1986) in almost all 6- to 12-year-old Dutch and Tanzanian children examined, spirochetes were detected in their plaque and the numbers and proportions were greater in plaque samples obtained from sites which bled on probing. The prevalence of spirochetes in our results are more in accordance with Asikainen et

Table 3. Clinical and microbiological description of the children

Fam	Sex	Age (years)	PI	SBI	%pd ≤3 (mm)	%pd ≥6 (mm)	Mean PD (mm)	Max LA (mm)	Presence or absence				
									Pi	Pg	Aa	spi	mmo
1	F	10	1.23	0.65	72.9	4.2	3.44	3	+	−	+	+	+
	M	5	0.90	1.12	85.7	0	3.24	1	+	−	−	+	+
	F	0	nd	nd	nd	nd	nd	nd	−	−	−	−	+
2	M	1	nd	nd	nd	nd	nd	nd	−	−	−	−	−
3	M	4	nd	nd	nd	nd	nd	nd	+	−	−	−	+
	M	1	nd	nd	nd	nd	nd	nd	−	−	−	−	−
4	M	4	0.38	0.20	97.5	0	2.60	0	−	−	−	−	+
5	F	8	0.94	0.85	100	0	2.60	0	+	−	−	−	+
	F	6	0.81	0.60	95.8	0	2.50	0	+	−	−	−	+
6	M	3	1.17	1.19	91.7	0	2.99	0	−	−	+	−	+
	M	1	nd	nd	nd	nd	nd	nd	−	−	−	−	+
7	F	10	1.63	2.00	68.8	0	3.33	1	+	−	−	+	+
	M	7	1.06	1.56	87.0	0	2.67	1	+	−	+	+	+
	F	0	nd	nd	nd	nd	nd	nd	−	−	−	−	−
8	F	8	1.06	0.92	87.5	0	2.73	0	+	−	+	−	+
9	F	9	1.57	1.72	100	0	2.72	0	+	−	−	+	+
	M	7	1.64	0.84	86.4	0	2.90	0	+	−	−	−	+
10	F	2	0.22	0.31	100	0	1.97	0	−	−	−	−	+
	M	0	nd	nd	nd	nd	nd	nd	−	−	−	−	−
11	F	15	1.46	1.29	75.0	0	3.29	1	+	−	−	+	+
	M	9	0.88	1.06	100	0	2.17	0	+	−	−	−	+
	F	3	1.88	0.78	100	0	2.08	0	+	−	−	+	+
12	M	13	1.54	1.86	73.2	0	3.61	1	+	−	−	−	+
	M	11	1.35	1.83	78.8	0	3.48	0	+	−	−	−	+
	F	1	nd	nd	nd	nd	nd	nd	−	−	−	−	+
13	M	5	0.20	0.30	100	0	2.15	0	−	+	−	−	+
14	F	5	0.79	0.63	100	0	2.29	0	−	−	+	+	+
	M	3	0.82	0.26	100	0	2.11	0	−	−	−	−	−
15	F	13	1.70	1.43	82.1	0	3.00	0	+	−	−	+	+
	F	8	0.92	0.86	100	0	2.42	0	+	−	−	−	+
16	F	15	1.05	0.89	96.4	0	2.90	0	+	−	−	−	+
	F	11	0.79	0.89	92.9	0	2.95	1	+	−	−	+	+
17	M	14	0.51	0.92	89.3	0	3.09	0	+	−	−	−	+
	F	11	1.52	1.85	89.6	0	2.90	0	+	−	−	−	+
18	F	7	1.02	1.33	89.6	0	2.79	0	+	−	−	−	+
	M	5	0.78	0.55	100	0	2.43	0	+	−	−	−	+
19	M	11	1.42	1.71	83.3	0	2.79	0	+	−	−	−	+
	M	10	0.96	1.10	100	0	2.33	0	+	−	−	−	+
20	F	12	1.02	1.04	83.9	0	3.13	2	+	−	−	−	+
21	F	8	0.95	0.84	100	0	2.25	0	+	−	−	+	+
	F	4	0.48	0.33	100	0	2.25	0	+	−	−	+	+
22	F	9	0.36	0.59	87.0	0	3.13	0	+	−	−	+	+
	F	7	0.50	0.65	87.5	0	2.42	0	+	−	−	−	+
	M	4	nd	nd	nd	nd	nd	nd	−	−	−	−	+
23	M	13	0.73	0.45	90.5	0	2.76	0	+	−	−	−	+
	F	10	0.84	1.18	97.7	0	2.50	1	+	−	−	−	+
	F	7	1.00	0.23	100	0	2.23	0	−	−	−	−	+
24	M	10	0.25	0.48	95.8	0	2.58	0	+	−	−	−	+
	M	7	0.52	0.39	90.9	0	2.45	0	+	−	−	−	+

For legends, see Table 1.

al. (1986) who found spirochetes in 13% (in our study 18%) of the plaque samples of 100 healthy teenagers.

Investigating in this study the relationship between the microbial flora and the periodontal condition in children, a correlation was found between the presence of spirochetes and CAL. Such an association could not be found between the clinical parameters and the other investigated bacteria. The prevalence of CAL within the age group 5–15 years was 26.5%, which seems to be rather high, if it is compared with the prevalence of 5% CAL in 15- to 16-year-old adolescents, which was found in an epidemiological study in Amsterdam by Van der Velden et al. (1989). This suggests that children of adult periodontitis patients may have a greater risk to develop periodontal destruction than other children.

Offenbacher et al. (1985) examined cross-sectionally the subgingival microflora in husband and wife cohabitant pairs to determine whether there are greater similarities in bacterial darkfield morphotypes from the periodontal sulci of couples living together than between other individuals. The results indicated that the flora was better matched between spouses, suggesting a risk of transmission within couples. In the pres-

Table 4. Prevalence of suspected periodontal pathogens per sampled site of the probands ($N=24$)

	N positive[a]	Tongue	Buccal mucosa	Tonsils	Saliva	Pocket
Pg	18	7[b]	14	11	10	18
		3.3±6.2[c]	3.5±4.5	1.4±1.3	3.5±6.7	26.9±19.1
Aa	13	11	11	5	11	13
		0.01±0.03	0.4±1.3	0.2±0.5	0.1±0.1	1.7±2.4
Pi	24	23	22	23	23	24
		1.1±3.0	3.2±4.2	2.2±4.4	6.6±14.0	9.1±9.4
spir	22	2	0	1	5	22
		0.8±0.3	0	1.64	0.5±0.2	5.5±5.4
mmo	24	21	0	8	17	24
		27.6±18.3	0	3.7±2.2	10.4±9.4	20.3±14.3

[a] Number of positive subjects for the specific microorganism; [b] Number of the positive subjects positive at the specific site; [c] Mean proportion±standard deviation of the microorganisms of the positive subjects.
Pg=*P. gingivalis*; Aa=*A. actinomycetemcomitans*; Pi=*P. intermedia*; spir=spirochetes; mmo=motile microorganisms.

Table 5. Prevalence of suspected periodontal pathogens per sampled site of the spouses ($N=24$)

	N positive[a]	Tongue	Buccal mucosa	Tonsils	Saliva	Pocket
Pg	11	7[b]	9	7	10	9
		0.39±0.69[c]	0.63±0.61	7.8±13.2	1.5±2.1	29.3±22.7
Aa	5	3	4	2	3	5
		0.01±0.01	0.04±0.07	0.01±0.01	0.5±0.5	1.1±1.9
Pi	24	20	19	20	21	23
		0.2±0.2	1.2±1.5	2.7±6.7	1.3±1.7	6.1±7.0
spir	18	0	0	4	3	16
		0	0	3.6±4.3	3.1±3.7	4.9±4.6
mmo	24	21	0	4	15	19
		36.1±17.0	0	5.7±3.3	21.4±15.6	12.1±10.5

For legends, see Table 4.

Table 6. Prevalence of suspected periodontal pathogens per sampled site of the children ($N=49$)

	N[a]	Tongue ($N=45$)	Buccal mucosa ($N=49$)	Tonsils ($N=39$)	Saliva ($N=49$)	Pocket 1 ($N=35$)	Pocket 2 ($N=31$)
Pg	1	0[b]	1	1	1	0	0
		0	0.009	4.2	1.4	0	0
Aa	5	1	3	2	3	4	3
		0.01	0.3±0.6[c]	0.003±0.004	2.7±4.6	0.6±0.4	0.1±0.009
Pi	34	25	18	22	31	23	23
		0.4±0.8	0.7±1.0	0.7±1.3	0.8±1.6	3.0±3.5	3.7±5.4
spir	13	7	0	1	3	5	6
		1.7±1.0	0	21.5	1.8±1.7	7.1±6.5	3.2±2.9
mmo	44	36	2	17	24	13	11
		21.7±17.8	29.3	10.2±15.0	14.5±10.3	12.8±10.8	7.8±6.1

For legends, see Table 4
Pocket 1=pooled sample of pockets of the primary dentition; pocket 2=pooled sample of pockets of the permanent dentition.

ent study, we found a low percentage of *A. actinomycetemcomitans* positive spouses, which indicates that transmission of *A. actinomycetemcomitans* between spouses does not seem to occur easily. In contrast, Sixou et al. (1991) suggested on the basis of biotyping and serotyping that transmission of *A. actinomycetemcomitans* may occur frequently between JP patients and their spouses. This difference in results may be due to the fact that the probands in the study of Sixou et al. were younger in age and were diagnosed as JP patients, possibly harbouring higher numbers of *A. actinomycetemcomitans*.

Van Steenbergen et al. (in press) indicated the possibility of transmission of *P. gingivalis* between spouses, on the basis of the presence of distinct Restriction Endonuclease Patterns (REPs) of *P. gingivalis* isolates from unrelated individuals and the presence of indistinguishable REPs of *P. gingivalis* isolates within the couples. These couples were derived from the couples presented in this study. If the high % of *P. gingivalis*-positive spouses is due to transmission of *P. gingivalis* between the spouse and the proband, the question then arises which factors influence transmission. The probands of the couples with both subjects positive for *P. gingivalis* had a higher prevalence of *P. gingivalis* in the saliva than the *P. gingivalis* positive probands with negative spouses. Person to person transmission of this bacterium may occur more easily if the bacterium is present in the saliva. The oral colonization or outgrowth of *P. gingivalis* in the spouses of the *P. gingivalis* positive probands was correlated with an increasing number of years of cohabitation of the couples. As colonization with *P. gingivalis* was also correlated with increasing age definite conclusions can not be drawn. The significance of the oral colonization of the spouses with *P. gingivalis* may be questioned, since the periodontal condition of the *P. gingivalis* positive spouses was not different from the periodontal condition of the *P. gingivalis* negative spouses. It may be possible that the positive spouses have been carriers for many years. On the other hand they may have been infected only recently and periodontal destruction may develop in the course of time.

The couples in this study had been living together for on the average 14 years. It can be questioned to what extent the severely inflamed periodontal

Table 7. Presence (+) or absence (−) of *P. gingivalis* in 24 couples and their children

Couples ($N=24$)	Children ($N=49$)	
	children$^+$	children$^-$
proband$^+$ spouse$^+$ ($n=10$)	1	17
proband$^+$ spouse$^-$ ($n=8$)	0	16
proband$^-$ spouse$^+$ ($n=1$)	0	2
proband$^-$ spouse$^-$ ($n=5$)	0	13

Table 8. Presence (+) or absence (−) of *A. actinomycetemcomitans* in 24 couples and their children

Couples ($N=24$)	Children ($N=49$)	
	children$^+$	children$^-$
proband$^+$ spouse$^+$ ($n=4$)	2	6
proband$^+$ spouse$^-$ ($n=9$)	3	15
proband$^-$ spouse$^+$ ($n=1$)	0	2
proband$^-$ spouse$^-$ ($n=10$)	0	21

Table 9. Presence (+) or absence (−) of spirochetes in 24 couples and their children

Couples ($N=24$)	Children ($N=49$)	
	children$^+$	children$^-$
proband$^+$ spouse$^+$ ($n=17$)	10	26
proband$^+$ spouse$^-$ ($n=5$)	2	7
proband$^-$ spouse$^+$ ($n=1$)	1	1
proband$^-$ spouse$^-$ ($n=1$)	0	2

Table 10. Prevalence of *A. actinomycetemcomitans*, *P. gingivalis*, *P. intermedia*, spirochetes and motile microorganisms in at least one of the sampled sites of the children 5–15 years of age in relation to periodontal breakdown

	Loss of attachment		
	Present ($N=9$)	Absent ($N=25$)	
A. actinomycetemcomitans	2 (22%)	2 (8%)	
P. gingivalis	0	1 (4%)	
P. intermedia	9 (100%)	21 (84%)	
spirochetes	6 (67%)	5 (20%)	$p=0.02$
motile microorganisms	9 (100%)	25 (100%)	

condition of the probands influenced the periodontal condition of the spouses. Since a proper control group is missing we can only compare the prevalence of periodontitis in the spouses to the prevalence of periodontitis in the same age group of the Dutch population. Therefore we compared our results to an epidemiological study on the periodontal condition in the Netherlands (Truin et al. 1989). The prevalence of the presence of a pocket $\geqslant 6$ mm, which is the CPITN score 4, i.e., the most severe periodontal condition, was about 10%. In our spouses we found a prevalence of about 38% of CPITN score 4. This difference is suggestive for an influence of the periodontal condition of the proband on the periodontal condition of the spouse. More studies on this subject are necessary to investigate the role of a severe periodontitis patient as a source of infection.

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Zusammenfassung

Prävalenz von Parodontitis und verdächtigen Parodontalpathogenen in Familien von Erwachsenenparodontitis-Patienten

Das Ziel der vorliegenden Studie war die Untersuchung der Prävalenz von parodontalpathogenen Mikroorganismen und parodontaler Zerstörung bei den Ehegatten und Kindern von Erwachsenenparodontitis-Patienten. Für diese Studie wurden 24 Familien auf Grund der Tatsache ausgewählt, daß ein Elternteil eine schwere Parodontitis hatte und subgingival *Actinobacillus actinomycetemcomitans* und/oder *Porphyromonas gingivalis* und/oder mehr als 30% *Prevotella intermedia* vorhanden waren. Die klinische Untersuchung der Eltern und Kinder bestand aus Messungen der Sondierungstiefe und des klinischen Attachmentverlusts (CAL). Die Proben zur bakteriellen Untersuchung wurden den Schleimhäuten, dem Speichel und den Taschen entnommen. Die Auswahl der Tasche erfolgte nach der am meisten fortgeschrittenen Läsion, die bei einem Patienten vorgefunden wurde. Die Proben wurden zum Nachweis von *A. actinomycetemcomitans*, *P. gingivalis* und *P. intermedia* kultiviert. Mittels Phasenkontrast-Mikroskopie wurde der prozentuale Anteil von Spirochäten und beweglichen Mikroorganismen bestimmt. Die Anzahl der Kinder in einer Familie schwankte zwischen 1 und 3. Insgesamt wurden 49 Kinder im Alter von 3 Monaten bis zu 15 Jahren untersucht. Die Ergebnisse zeigten, daß bei einem Alter unter 5 Jahren kein Kind CAL aufwies, während in der Altersgruppe von 5–15 Jahre 26.5% 1–5 Flächen im Milch- und/oder bleibenden Gebiß mit 1–3 mm CAL vorhanden waren. 3 der Ehegatten hatten keinen inderdentalen CAL. Sechzehn der 24 Ehegatten hatten eine leichte bis mäßige Form der Parodontitis mit wenigstens einer Zahnfläche mit 1 bis 4 mm CAL. Fünf Ehegatten hatten starken parodontalen Abbau mit Flächen von wenigstens 8 mm CAL. Spirochäten, bewegliche Mikroorganismen und *P. intermedia* waren bei allen Familienmitgliedern häufig zu finden. 18 der 24 Probanden waren positiv hinsichtlich *P. gingivalis*. Dieser Organismus wurde nur einmal bei einem 5 Jahre alten Jungen und bei 11 der Ehegatten gefunden. *A. actinomycetemcomitans* wurde bei 13 Probanden nachgewiesen; 5 Kinder und 5 der Ehegatten waren auch positiv bezüglich dieses Bakteriums. Dieses Phänomen könnte durch eine Übertragung zwischen den Familienmitgliedern zustande gekommen sein. Aus dem Vergleich der vorgestellten klinischen Daten der vorliegenden Studie mit ähnlichen klinischen Parametern aus epidemiologischen Studien der holländischen Bevölkerung läßt sich vermuten, daß die Ehegatten und Kinder von Erwachsenenparodontitis-Patienten ein höheres relatives Risiko hinsichtlich der Entwicklung von parodontalem Abbau haben könnten.

Résumé

Prévalence de la parodontite et des organismes soupçonnés pathogènes pour le parodonte dans les familles de patients atteints de parodontite de l'adulte

Le but du présent travail était d'étudier la

prévalence des microorganismes pathogènes pour le parodonte et de la destruction parodontale chez les conjoints et chez les enfants de patients atteints de parodontite de l'adulte. A cet effet, 24 familles ont été sélectionnées parce qu'un des parents présentait une sévère destruction parodontale avec présence sous-gingivale d'*Actinobacillus actinomycetemcomitans* et/ou de *Porphyromonas gingivalis*, et/ou plus de 30% de *Prevotella intermedia*. L'examen clinique des parents ainsi que des enfants comprenait l'enregistrement de la profondeur des poches et de la perte d'attache clinique (CAL=clinical attachment loss). Des échantillons ont été prélevés pour l'examen bactériologique au niveau des muqueuses, de la salive et des poches. Les poches choisies étaient celles où on trouvait la parodontite la plus avancée chez le patient en question. Les échantillons ont été mis en culture pour y rechercher *A. actinomycetemcomitans*, *P. gingivalis* et *P. intermedia*. La proportion des spirochètes et des microorganismes motiles a été établie au microscope à contraste de phase. Le nombre d'enfants par famille allait de 1 à 3. L'examen a porté sur un total de 49 enfants âgés de 3 mois à 15 ans. Les résultats ont montré qu'aucun des enfants âgés de moins de 5 ans ne présentait de CAL, tandis que 26.5% des enfants âgés de 5 à 15 ans avaient 1–5 sites ayant 1–3 mm de CAL dans la denture temporaire et/ou dans la denture permanente. Chez 3 des conjoints, on ne constatait pas de CAL interproximale. Chez 16 des 24 conjoints, il existait une parodontite de forme légère à modérée, avec au moins 1 site ayant une CAL de 1 à 4 mm, et chez 5 des conjoints, il existait une sévère destruction parodontale, avec des sites ayant une CAL d'au moins 8 mm. Des spirochètes, des microorganismes motiles et *P. intermedia* étaient souvent présents chez tous les membres d'une famille. Pour 18 des 24 probants, le sujet était positif pour *P. gingivalis*. Cet organisme a été trouvé une seule fois chez un garçon de 5 ans et chez 11 des conjoints. *A. actinomycetemcomitans* a été trouvé chez 13 des probants; 5 enfants et 5 des conjoints étaient aussi positifs pour cette bactérie. Lorsqu'un enfant abritait un des organismes pathogènes pour le parodonte, au moins un de ses parents était aussi positif pour cette bactérie. Ce phénomène peut être dû à la transmission de microorganismes entre les membres d'une famille. La comparaison entre les données cliniques dont il est rendu compte dans cette étude et les paramètres cliniques similaires obtenus dans des études épidémiologiques de la population hollandaise semble indiquer que les conjoints et les enfants de patients atteints de parodontite de l'adulte sont exposés à un risque relativement élevé d'être touche's par la destruction parodontale.

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